

Sickle Cell Disease: Demystifying the Beginnings

by Clarice Reid and Griffin Rodgers

The Precursors

As a global health problem, sickle cell anemia affects many world populations. Since its initial description in the United States almost a century ago, scientists and medical researchers have continued to better understand the pathophysiology of this inherited disease, while simultaneously attempting to find more effective therapies and ultimately a cure.

The pathology of Sickle Cell Disease (SCD) is complicated, in addition to moderate to severe anemia, it often includes infarction and resorption of the spleen during childhood, which results in diminished immune function. Growth and development are delayed, and the ability to produce offspring is often lessened. Bone and eye disorders may occur. Vaso-occlusive “crises” or painful episodes punctuate the course of the disease in many patients, and they may be characterized by unbearable pain in the legs and arms, back, chest, or abdomen. Also affected are the cardiovascular, pulmonary, and renal systems.

The first case report in United States literature occurred in 1910, when James B. Herrick, a cardiologist at Rush Presbyterian Hospital in Chicago, described the physical attributes of the disease as “peculiar, elongated and sickle-shaped erythrocytes” causing “distortion of red blood cells . . . by the aggregation of abnormal hemoglobin molecules.” One of his patients, a 20-year-old student at the University of Chicago Dental School, presented years earlier at Rush Presbyterian Hospital with a leg ulcer and, later, pneumonia. On the patient’s peripheral blood smear, marked anemia and crescent-shaped erythrocytes were noted. An intern, Ernest Irons, then described these cells and drew pictures of the peculiar sickle-shaped red blood cells; these original drawings provided the classic description of SCD. Herrick postulated that these abnormal cells might play a major role in the patient’s illness, which prompted Herrick to report his findings in the Archives of Internal Medicine.¹

Historically, the disease had been known for generations in Africa, and according to reports of clinical syndromes in Western Africa, the disease had numerous local names. Following Herrick’s observations, a number of significant studies were done on these peculiar cells, and several prominent hematologists theorized that the problem might lie within the hemoglobin.

Lemuel Diggs, a pathologist at the University of Tennessee, concluded from autopsy findings that the occlusion of the microvasculature might be a factor in the painful episodes linked to the disease.² In 1949, Linus Pauling and his colleagues demonstrated the electrophoretic abnormality in sickle hemoglobin (HbS), which migrated differently than normal hemoglobin in an electric field. Pauling thus recognized that SCD must be a disease of the hemoglobin molecule.³ In contrast to sickle cell anemia, both sickle hemoglobin and normal hemoglobin were present in the sickle cell trait. This was the first example of understanding a human disease in terms of a specific protein abnormality, and led to the identification of protein abnormalities in several other genetic diseases, especially hemoglobinopathies and enzyme deficiencies.

At about the same time, James V. Neel of the University of Michigan reported that he had examined the parents of 29 patients with sickle cell anemia and found that the red blood cells could be induced to sickle in every parent.⁴ James Neel and E. A. Beet independently reported that the mode of inheritance was a recessive gene, which made it clear that hemoglobin was the culprit.⁵ In 1957, Vernon Ingram ascribed the molecular abnormality to a substitution of valine, an amino acid, for glutamic acid at the $\beta 6$ position of the globin chain; this discovery was based on the development of a fingerprinting technique used to identify changes in amino acids.⁶

Over the next decade, a number of other abnormal hemoglobins were identified, such as HbC, which in combination with sickle cell hemoglobin is usually a milder form of the disease. Currently, there are over 1,200 abnormal hemoglobins, with most lacking any clinical significance. The field was further advanced by Max Perutz who elucidated the three-dimensional structure of the hemoglobin molecule by X-ray diffraction, for which he received the Nobel Prize for Physiology and Chemistry in 1967.⁷

The 1970s heralded new and important developments in both sickle cell anemia and thalassemia through a revolution in molecular biology brought about by recombinant DNA technology. Also, new techniques of nucleic-acid sequencing were applied, which illustrated that the nucleotide change in the DNA for sickle hemoglobin resulted an A \rightarrow T change in the DNA, a mutation which alters the codon GAG (glu) to GTG (val). Sickle cell anemia thus became the first human disease whose abnormality was recognized at the level of the single nucleotide mutation in the gene. Many of these alterations have also been identified for other genetic diseases, especially thalassemia.

At the national level, sickle cell anemia was the first targeted program for a genetic disease, and offered a point of reference in setting the stage for the incredible progress of the past three decades. These achievements are even more noteworthy given the label of sickle cell anemia as an “orphan disease” in the United States, and the limited support of research by the pharmaceutical industry. Many of these advances will be discussed in detail in later chapters.

The National Sickle Cell Disease Program

The 1970s launched unprecedented attention to SCD with both increased public awareness and mixed enthusiasm. This was sparked in 1971, when President Nixon, in his health message to Congress, targeted sickle cell anemia as a major health problem and encouraged greater support at the national level. An earlier article by Robert Scott, Medical College of Virginia, appearing in the *Journal of the American Medical Association*, compared federal resources for SCD with less prevalent genetic diseases found predominantly in non-black populations (e.g., cystic fibrosis, muscular dystrophy), further fueling this national debate.⁸ An editorial in the same *Journal* stated that “the level of general ignorance concerning the nature of sickle cell anemia remains depressingly high despite substantial scientific advances.” The National Institutes of Health (NIH) support for SCD at that time was approximately one million dollars in three institutes.

In the African-American community, the political climate was one of considerable unrest concerning civil equality. Skepticism and cynicism abounded at the news that a program focused on a virtually unknown genetic disorder in African-Americans had been elevated to national priority level. Further compounding the problem were the prevailing myths and misconceptions about this disorder, the confusion of sickle cell trait with SCD, and the dilemma posed by follow-up counseling.⁹ Screening programs proliferated; well-intentioned legislators passed laws mandating premarital and pre-school screening; insurance companies increased premiums for individuals with sickle cell trait; and the Air Force denied trait carriers occupational opportunities as pilots or co-pilots. The community viewed many of these new policies as genocidal, with overtones of political exploitation. These interdictions were soon reversed; the program transcended these barriers and SCD moved to a position of prominence, attracting top scientists throughout the world to work on this genetic disease.¹⁰ This was a major triumph for medicine and science.

The public health response was immediate. Secretary Elliott Richardson of the Department of Health, Education and Welfare (DHEW) appointed the National Sickle Cell Disease Advisory Committee; its professional and lay members recommended a broad-based program of research and service. A national program was established in the Public Health Service in 1972, with the NIH designated as lead agency, and program responsibility was assigned to the National Heart, Lung and Blood Institute (NHLBI). Basic research — including studies into globin molecular genetics, red cell metabolism, hematopoiesis and hematopoietic stem cells — was and continues to be supported by the Division of Kidney, Urologic and Hematologic Diseases of the National Institute of Diabetes and Kidney Diseases (NIDDK). NIDDK has also fostered a strong translational research program in iron-chelators and in non-invasive assessment of iron-overload. For the past 33 years, the NHLBI has been the major source of funding support for sickle cell anemia.

Roland Scott, “patron saint” of SCD in the United States, spearheaded many of the initial efforts to obtain federal attention for the disease, and testified at Congressional hearings on this disorder.¹¹ In 1972, Congress passed the National Sickle Cell Control Act (PL.92-294), mandating federal programs for diagnosis, control, treatment, and research in sickle cell anemia.¹² This was the first federal legislation to focus on a genetic disorder. No appropriations were made under this legislative authorization; funds were from the budget of the NHLBI/NIH. The legislation expired in 1975. John Hercules provided the major leadership for advancing sickle cell disease scientific research programs at the NHLBI.

The Health Services Administration (HSA) carried out the service component, with 19 Screening and Education Clinics funded as demonstration projects to hospitals, universities and community health centers, and to freestanding organizations. All played a key role in lessening hysteria, mobilizing the community to participate in selected screening programs, and providing accurate information and follow-up counseling. Education of the public, including health educators and health care providers, was a high priority for the national program during the early years. In many cases, these programs served as entry points for individuals into the health care delivery system.

In 1978, federal support for the Screening and Education Clinics administered by the Genetics Program at the Health Resources and Services Administration (HRSA) was transferred to the states via State Block Grants. The Genetic Services Branch, Maternal and Child Health Bureau, continued to support services through its appropriation for Special Projects of Regional and National Significance (SPRANS). HRSA played a major role in funding newborn screening and follow-up programs, and in collecting valuable demographic data through the support of the Council of Regional Networks (CORN). In recent years, the Maternal and Child Health Bureau, HRSA, have received a

direct appropriation to support a community-based demonstration project for sickle cell newborn screening.

The comprehensive sickle cell centers have been the major component of the national program, established in geographical areas with large at-risk populations. The original center concept focused on the patient, while embracing community involvement and the integration of services in an academic research environment. This was one of the early program models to accommodate the emerging emphasis on prevention, education, and control programs supported by the NIH. In this setting, the previously impersonal, fragmented, and episodic care for sickle cell patients was drastically changed. A cadre of trained personnel — clinicians, nurses, social workers, educators, psychologists, nutritionists, and counselors — worked closely in a team approach to patient care. Emergency room protocols were developed, while parents and caregivers were trained to detect important signs and symptoms of early complications. Sickle cell programs at non-NIH funded medical centers, using similar models, have played a pivotal role in improving overall patient care. A minimum of ten centers was congressionally mandated by the Orphan Drug Act, Public Law 97-414, in 1983. Over the years, the focus of the center program has shifted more toward basic and clinical research, with limited support of non-research and community activities.

The hemoglobinopathy training program at the Centers for Disease Control (CDC) in Atlanta was a prominent part of the early national sickle cell program. It was established to train laboratory personnel in the use of up-to-date technologies to detect abnormal hemoglobin species. Laboratories with federal funding had to participate in the proficiency-testing program of hemoglobin diagnosis; this monitoring of laboratory procedures resulted in better quality assurance and accuracy of laboratory diagnoses. Early focus of the National Program was on emergency room management and the development of treatment protocols to ensure adequate care for all patients. A consensus of experienced clinicians in the field led to the publication of “Management and Therapy,” outlining the basics of routine care, and for common medical and surgical problems. This document has been updated over the years and continues to be a handy reference for the management of sickle cell patient.¹³

The Sickle Cell Disease Association of America (SCDAA), founded by Charles Whitten in 1972 as the National Association of Sickle Cell Disease, is the key lay organization directed toward improving the quality of life for sickle cell patients and their families, and ultimately finding a cure. It is the national umbrella organization for 57 community-based associations located across the country that provide public and professional health education, patient services, community outreach, and newborn screening follow-up. Since its inception, through a community-based approach, the SCDAA has been in the forefront of advancing public awareness, correcting misinformation, and providing counseling and patient advocacy for thousands with the disease and their families for the past three decades. These activities had a major impact on changing the earlier climate for sickle cell programming at the local and national levels.

As an ongoing partner with the National Sickle Cell Disease Program, the SCDAA is a strong advocate for obtaining federal funds for sickle cell research and services, and works closely with federal agencies. In its early years, the SCDAA provided research funding to the ten Comprehensive Sickle Centers, and later initiated a Summer Research Apprentice Program that allowed promising high school seniors to gain early exposure to research. The SCDAA has established a Post-Doctoral Research Fellowship Program aimed at developing young investigators beginning their biomedical and psychosocial research careers. In fiscal year 2002, as a result of the legislative efforts of the SCDAA, an unprecedented \$4 million was earmarked for the budget of the Maternal and Child Health Bureau, HRSA, to fund sickle cell activities. This support is scheduled to continue to fiscal 2008.

Clinical Advances

Early clinical investigations to improve the outcomes of sickle cell anemia patients targeted the recurrent painful episode that was the clinical hallmark of the disease. In the mid-1960s, Makio Murayama hypothesized that the polymerization of the deoxyhemoglobin S molecule was dependent upon the existence of hydrophobic bonds between molecules.¹⁴ This hypothesis led to *in vitro* studies in 1971 by Robert Nalbandian that demonstrated that the compound urea could interfere with these bonds, and thereby prevent and reverse sickling.¹⁵ A small, uncontrolled study reported efficacy in ameliorating both severity and duration of pain when urea was administered intravenously to sickle cell patients in a solution of invert sugar; however, these findings were not confirmed in a controlled, double-blind, randomized study funded by the NIH. A companion study also revealed that alkali was not beneficial in the treatment of pain, as had been suggested by other reports.^{16,17}

Various additional therapies were thus introduced to inhibit polymerization, swell red blood cells, and modify hemoglobin, but with little or no success. Anthony Cerami and James Manning noted that the small amounts of cyanate which formed in urea solutions might have anti-sickling effects after completing *in vitro* and animal studies on cyanate, which showed that anti-sickling properties were due to an increased affinity for oxygen.¹⁸ Clinical trials were initiated in a small number of medical centers; the observed benefits, however, were offset by neurotoxicity and other complications when higher doses were utilized.¹⁹ Thus these studies were terminated. Yet the drug seemed to hold much promise, for it appeared that the drug would be safe if cyanate reacted only to red blood cells, with the extra cyanate being eliminated. Funding from the NIH enabled testing of an instrument similar to a hemodialysis device that delivered cyanate by the extracorporeal route of administration.²⁰ This process of carbamylation proved cumbersome, and while it demonstrated potential feasibility, problems arose with hyperviscosity. Therefore, the project was discontinued.

These clinical studies on urea and cyanate were quite important in underscoring the necessity for supporting basic clinical research on therapeutic strategies for sickle cell anemia; they also highlighted the need to understand the natural history of the disease in order to assess the therapeutic efficacy for various complications. During this period, a quantum leap in the application of molecular techniques occurred which greatly increased the understanding of sickle cell anemia at molecular, cellular, and clinical levels. Studies on fetal hemoglobin synthesis in erythroid cells catalyzed the field of molecular biology and became a foundation for the development of therapeutic approaches based on increasing the level of fetal hemoglobin in those afflicted with sickle cell anemia.

Another significant development in the 1970s was prenatal diagnosis of the disease by Yuet W. Kan and Andrea Dozy.²¹ Earlier techniques for prenatal diagnosis required fetal cells for the biochemical analysis of globin chains to determine the genotype of the fetus. This approach was replaced by gene mapping of DNA fragments using restriction endonuclease enzymes and identification of polymorphisms (restriction fragment length polymorphism or RFLP analysis) adjacent to the β -globin locus. The largest utilization for prenatal diagnosis has been in other populations, especially β -thalassemia. In contrast, the broader application of RFLP has found its way to forensics, agriculture and genomics.

The 1980s ushered in an era of great optimism under the leadership of Clarice Reid, Chief, Sickle Cell Disease Branch, via a series of clinical reports and an abundance of publications containing heretofore unknown clinical data related to sickle cell anemia. The predominant source of these reports was the National Institutes of Health study known as the Cooperative Study of Sickle Cell Disease (CSSCD) organized in 1979. Although there was a great body of knowledge on the molecular basis of SCD, there was a paucity of information on its clinical course and natural history. Much of the information in the medical literature only described the most severe cases, usually isolated

in a hospital setting; the reports of clinical outcomes were mostly retrospective or anecdotal. The variability of clinical severity, ranging from mild to debilitating and with the same genotype and hematologic patterns, challenged the clinician and the researcher. For the most part, information on the natural history was only available for the first 10 years within the United States from Darlene Powars and the Jamaican sickle cell anemia pediatric cohort of Graham Sergeant.^{22,23}

To improve understanding of the natural history and the variable severity of sickle cell disease, the CSSCD, a large-scale, multi-institutional epidemiological study, became of great importance.²⁴ Over 4000 patients were recruited, representing the major phenotypes of the disease, from infants to adults in the sixth decade of life. The infants in this study represented an unbiased population that was enrolled based on early diagnosis, and patient recruitment extended beyond hospital-based programs in which patients were the most severely affected; those involved in community-based health departments and under the care of physicians were also included. A number of important papers published by CSSCD investigators resulted from this prospective, longitudinal study and included patient demographics, growth and development patterns in children, pregnancy, transfusion and alloimmunization, infection, pain, mortality, major organ dysfunction, and others. A comprehensive summary of the many CSSCD publications has recently been published by Duane Bonds.²⁵

The landmark study from this period was the Prophylactic Penicillin Study led by Marilyn Gaston. Prior to the 1980s, 30% of deaths were due to pneumococcal infections, mostly in children under five years of age. A randomized, controlled clinical trial²⁶ found that the daily oral administration of penicillin reduced the rates of infection from *S. pneumonia* by 84% in young children with SCD (Fig. 1). There was a strong public health response to these compelling results showing that prophylactic penicillin could save lives. The NIH convened a consensus development conference on Newborn Screening for Sickle Cell Disease and other Hemoglobinopathies to examine the merit of

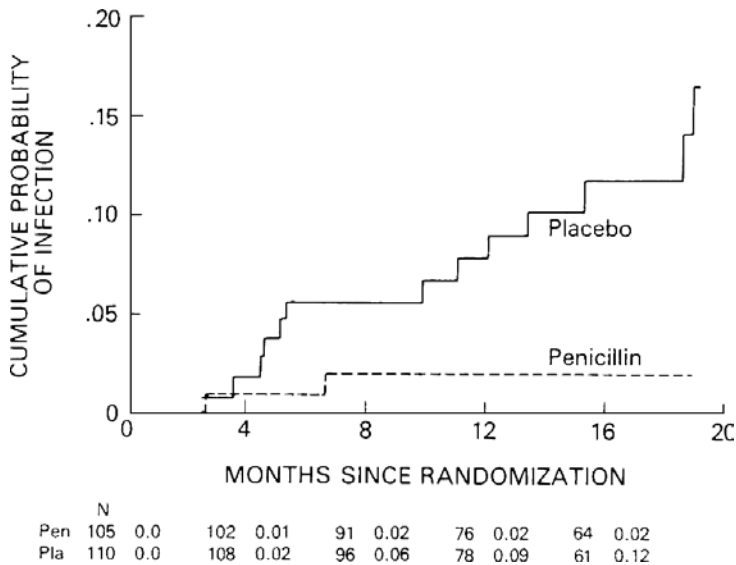


Fig. 1. Cumulative infection rates for all patients in the prophylactic penicillin study. Numbers of patients at risk and infection rates in the penicillin (Pen) and placebo (Pla) groups, at four-month intervals, appear below the curves (one-tailed p value = 0.003). By permission from Gaston MH *et al.* (1991). *N Engl J Med* **314**:1593–1599.

widespread newborn screening and its inherent controversies. The panel included biomedical scientists, clinicians, other health professionals, and representatives of the public. This led to a number of recommendations including that all newborns in the United States be screened for SCD and placed on prophylactic penicillin by the age of three months.²⁷ This was a major public policy change.

Newborn screening the administration of prophylactic penicillin, and early comprehensive care for infants with SCD are considered as general standards within the medical community, serving as the first steps toward prevention. More than 46 United States states screen at birth for SCD. Although the technology for screening of umbilical cord blood for SCD had been available for many years, the recommendation as policy for newborns was not formally adopted until the findings gained from the penicillin study.²⁸ Prevention of infection in infants and children now includes routine pneumococcal vaccination. It is safe to discontinue prophylactic penicillin in most children with sickle cell anemia after the age of five years.²⁹

For the first time, the CSSCD report on the epidemiology of painful events showed a correlation of pain rates with early death in sickle cell patients over the age of 20 (Fig. 2). The rate of pain was shown to be positively influenced by the levels of fetal hemoglobin (HbF), resulting in decreased pain rate and accompanying decreased morbidity. It was noted that 38% of the patients did not experience a painful event during a given five-year period between 1979 and 1988. In addition, patients with more frequent pain histories (3–10 events per year) represented only 5.2% of the population, but contributed 33% of the painful events to the analysis.³⁰

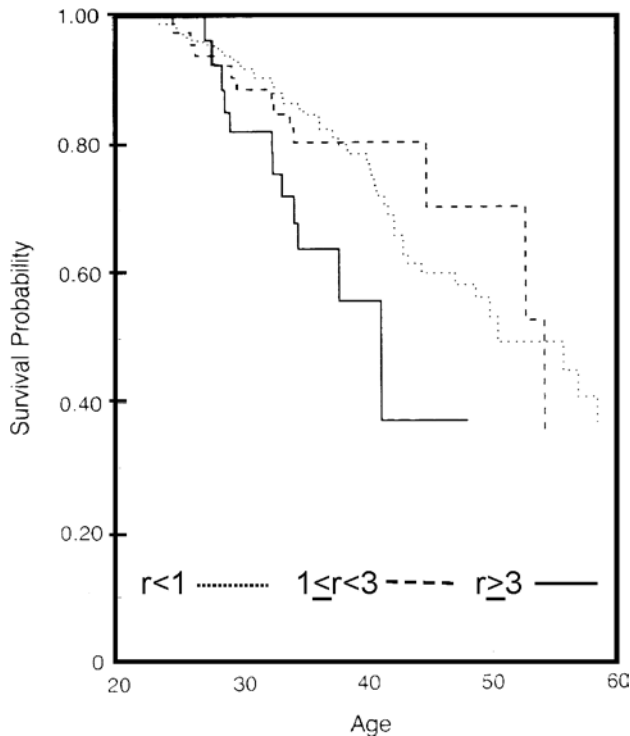


Fig. 2. Survival of patients with sickle cell anemia (≥ 20 years old at entry) who had different pain rates. The letter “r” denotes the number of episodes of paid per patient-year. By permission from Platt OS *et al.* (1991). *New Engl J Med* **325**:11–16.

Fortunately, the CSSCD data provided encouraging mortality information on both pediatric and adult patients with sickle cell anemia. Early hematology literature declared that patients rarely lived to adulthood. Survival curves, stratified by gender and the age of patients with SS and SC diseases in the CSSCD, indicated that the median life span was about 42 years for SS males, 48 years for SS females, 60 years for SC males, and 68 years for SC females.³¹ Although the medians differ by many years from other African-Americans, they reflect a significant improvement in survival for this population. The life expectancy of sickle cell patients has doubled since passage of the National Anemia Sickle Cell Control Act (Fig. 3). More recently published survival data³² on children with SCD concluded that “childhood mortality is decreasing, the mean age at death is increasing, and a smaller proportion of deaths are from infection.” This improvement allows long-range planning for education, careers, professional development, job opportunities, and the prospects of children and families.

With the advent of the 1990s, molecular medicine evolved from the bench to the bedside; basic research on globin gene regulation and how it functions also greatly improved.³³ This classic example of translation research was evidenced in the use of pharmacologic agents to reactivate dormant genes and ameliorate pain in severely affected sickle cell anemia patients. HbF is known to have a sparing effect on sickle hemoglobin, and those with high levels of fetal hemoglobin also were mildly affected. The findings by Joseph DeSimone, Timothy Ley and others that 5-azacytidine increases gamma globin synthesis led to the search for other less toxic cell-cycle specific agents exhibiting the same effects.^{34–36} One of the agents known to increase HbF is hydroxyurea.³⁷ Following small but promising proof-of-concept studies in non-human primates and in patients, Griffin Rodgers, Arthur Neinhuis and colleagues performed a pivotal phase I/II study on sickle cell patients admitted to the clinical center at the NIH for periods of three to four months.³⁸ This study demonstrated the safety and efficacy of hydroxyurea, and defined the optimal dosages to achieve HbF augmentation. After

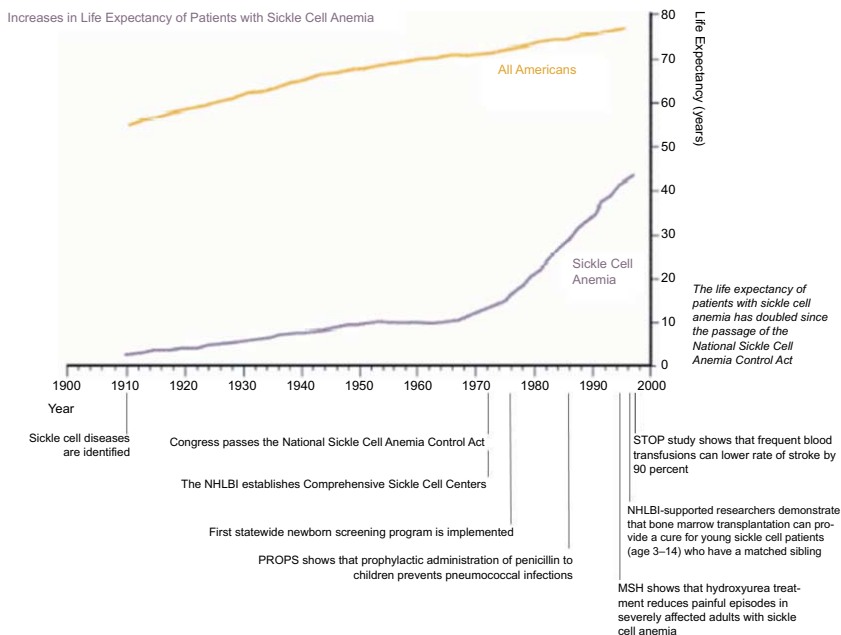


Fig. 3. Increases in life expectancy of patients with sickle cell anemia. The life expectancy of patients with sickle cell anemia has doubled since 1970. (From the National Heart Lung and Blood Institute, NIH publication No. 02-5214, September 2002.)

other phase II reports corroborated the NIH safety and clinical efficacy results, a prospective, multi-center, randomized clinical trial involving 299 patients using hydroxyurea was undertaken through the efforts of clinical investigator Samuel Charache, and Duane Bonds from the NHLBI. It demonstrated a 50% reduction in vaso-occlusive episodes in adults with sickle cell disease who were treated with this agent. There were also significant reductions in transfusion requirements, hospitalization, and acute chest syndrome.³⁹ Follow-up data available for up to nine years in 233 of the original subjects indicated, among other findings, that taking hydroxyurea was associated with a 40% reduction in mortality.⁴⁰

Hydroxyurea in children has been investigated in a safety and dosing study showing that the drug could be used in children between the ages of 5 and 15 years, accompanied by a significant increase in HbF and hemoglobin concentration.⁴¹ Subsequent reports noted that hydroxyurea was well tolerated in an even younger population of sickle cell infants with a median age of 15 months.⁴² The definitive Phase III trial of hydroxyurea in children to determine clinical efficacy in preventing end organ damage has been initiated at the NHLBI.

The investment in basic research and controlled clinical studies has proven to be of enormous benefit to adults with SCD. In addition, the study of this disease and other globin disorders has illuminated broader principles applicable to other genetic diseases, and has facilitated technological developments with applications to many more disorders. A testament to this conclusion is demonstrated by the many “firsts” associated with the study of SCD (Table 1) in which studies illuminated broader principles that resulted in more extensive applications to other fields of science and medicine.

Many of the young children with SCD have crippling central nervous system complications, including stroke, which was the second cause of death in the CSSCD pediatric population. Chronic blood transfusions to maintain the level of sickle hemoglobin below 30% have been successful in preventing recurrent strokes in children.⁴³ A major goal for preserving cerebro-vascular competence is to prevent the first stroke and its debilitating sequelae. The Stroke Prevention Trial in Sickle Cell Anemia (STOP) study demonstrated that such prevention is possible using non-invasive techniques to detect large cerebral vessel stenosis, identify children at-risk for stroke, and treatment with regular blood transfusions.⁴⁴ An additional challenge to the clinician is to prevent the problems of iron-overload that are secondary to treatment with transfusions in these patients.

Table 1. “Firsts” associated with the study of sickle cell anemia.

Authors	Year	Seminal contribution
J. Herrick	1910	First Clinical Report in the US
L. Pauling	1949	Application of protein electrophoresis
J. Neel	1949	Genetic nature of disease
M. Perutz	1951	X-ray crystallography of large protein
A. Allison	1954	Concept of “balance polymorphisms”
V. Ingram	1957	Two dimensional gel “fingerprint”
Y. W. Kan and A. Dozy	1976	Pre-natal diagnosis of a disease
A. Deisseroth	1977	Chromosomal localization of globin genes
Y. W. Kan and A. Dozy	1978	Restriction Fragment Length Polymorphism (RFLP) and DNA Haplotypes
J. DeSimone and P. Heller	1982	Reactivation of fetal (dormant) gene expression
R. Saiki and K. Mullis	1985	Clinical application of PCR

Summary

A number of seminal developments marked the pathway from the early clinical description of sickle cell anemia in 1910 to the unparalleled advances of the past three decades. The first federal program for a genetic disorder, launched in a political climate of civil unrest in the 1970s, continues to impact SCD research and services at the national level. This achievement is noteworthy and reflects the leadership role of the NIH in partnership with other federal agencies and the SCDA. The commitment to invest in basic and clinical research has led to major changes in public health policy and other interventions to prevent and/or decrease serious complications and save lives.

Tremendous progress has been made at all levels of SCD over the past three decades, and provides the catalyst for even more vigorous research. There is an optimistic outlook for patients, their families, the caring physician, and the vigilant scientist. Yet there remains an imbalance between what is known about SCD and translating this knowledge into effective therapies. There is only one approved drug for treatment, and a cure is not currently available for most patients. A limited number of primary care physicians and hematologists are caring for adult sickle cell patients. Nonetheless, comprehensive care and improved management have extended the quality of life and longevity of these patients. The absence of established genetic modifiers and biomedical markers of clinical severity limit the ability to identify patients who would benefit most from specific therapies, which becomes very important when a therapy has potentially serious or even life-threatening side-effects.

Improvements in preparative regimens for hematopoietic stem cell transplantation from sibling donors and the ability to use unrelated matched HLA donors in the future offer the best prospect to cure a limited number of patients with sickle cell disease. Gene therapy for SCD is the “holy grail” of cure, yet its application has been formidable due to the difficulty in transducing hematopoietic stem cells, and the necessity for erythroid specific, high level, and balanced globin gene expression. Despite these challenges efforts to move forward with gene therapy continue. Current genomic studies should provide insights on more pre-emptive strategies to resolve these therapeutic challenges.

References

1. Herrick JB (1910). Peculiar elongated and sickle-shaped red blood corpuscles in a case of severe anemia. *Arch Int Med* **6**:517–521.
2. Diggs L, Ching RE (1934). Pathology of sickle cell anemia. *South Med J* **27**:839–845.
3. Pauling L, Itano H, Singer SJ, Wells IC (1949). Sickle cell anemia, a molecular disease. *Science* **110**: 543–548.
4. Neel JV (1949). The inheritance of sickle cell anemia. *Science* **110**:164–166.
5. Beet EA (1949). The genetics of the sickle cell trait in a Bantus tribe. *Ann Eugen* **14**:279–284.
6. Ingram VM (1957). Gene mutations in human hemoglobin: the chemical difference between normal and sickle cell hemoglobin. *Nature* **180**:326–328.
7. Perutz MF, Rossman MG, Cullins AF, Muirhead H, Will G, North ACT (1960). Structure of haemoglobin: a three dimensional Fourier synthesis at 5.5, a resolution obtained by X-ray analysis. *Nature* **185**:416–422.
8. Scott R (1970). Health care priority and sickle cell anemia. *JAMA* **214**:731–734.
9. Whitten CF (1973). Sickle cell programming — an imperiled promise. *N Engl J Med* **288**:318–319.
10. Culliton B (1972). Sickle cell anemia: the route from obscurity to prominence. *Science* **178**:140.
11. Scott R (1983). Historical review of legislative and national initiatives for sickle cell disease. *Amer J Pediatric Hematology/Oncology* **5**(4):349.
12. The National Sickle Cell Disease Control Act (1972). Public Law, 92–294.
13. Management and Therapy of Sickle Cell Disease (2002 Revised). US Dept of Health and Human Services. NIH Publication, Rockville, MD. No. 02-2117.

14. Murayama M (1967). Structure of sickle cell hemoglobin and molecular mechanism of the sickling phenomenon. *Clin Chem* **13**:578.
15. Nalbandian RM, Henry RL, Barnhart MI, Nichols BM, Camp FR Jr, Wolf PL (1971). Sickle reversed and blocked by urea in invert sugar. *Am J Pathol* **65**:405–422.
16. McCurdy PR, Mahmood L (1971). Intravenous urea treatment of the painful crisis of sickle cell disease. *N Engl J Med* **285**:992–994.
17. (No authors listed) (1974). Therapy for sickle cell vaso-occlusive crises. Controlled clinical trials and cooperative study of intravenously administered alkali. *JAMA* **228**:1129–1131.
18. Cerami A, Manning JM (1971). Potassium cyanate as an inhibitor of the sickling phenomenon. *Proc Natl Acad Sci* **68**:1180–1183.
19. Harkness D, Roth S (1975). Clinical evaluation of cyanate in sickle cell anemia. *Progr Hematol* **9**:157–184.
20. Deiderich DA, Trueworthy RC, Gill P, Cader AM, Larsen WE (1976). Hematologic and clinical response in patients with sickle cell anemia after chronic extracorporeal red cell carbamylation. *J Clin Invest* **58**:642–653.
21. Kan YW, Dozy AM (1978). Antenatal diagnosis of sickle-cell anemia by D.N.A. analysis of amniotic fluid cells. *Lancet* **2**:910–912.
22. Serjeant GR, Grandison Y, Lowrie Y, Mason K, Phillips J, Serjeant BE, Vaidya S (1981). The development of haematological changes in homozygous sickle cell disease: a cohort study from birth to six years. *Br J Haematol* **48**:533–543.
23. Powars DR (1975). Natural history of sickle disease: the first ten years. *Semin Hematol* **12**:267.
24. Gaston M, Rosse WF and the Cooperative Group (1982). The cooperative study of sickle cell disease: review of study design and objectives. *Am J Pediatr Hematol Oncol* **4**:197–201.
25. Bonds DR (2005). Three decades of innovation in the management of sickle cell disease: the road to understanding the sickle cell disease clinical phenotype. *Blood Reviews* **19**:99–110.
26. Gaston MH, Verter JI, Woods G, Pegelow C, Kelleher J, Presbury G, Zarkowsky H, Vichinsky E, Iyer R, Lobel JS (1986). Prophylaxis with oral penicillin in children with sickle cell anemia: a randomized trial. *N Engl J Med* **314**:1593–1599.
27. NIH Consensus Conference (1987). Newborn screening for sickle cell disease and other hemoglobinopathies. *JAMA* **258**:1205–1209.
28. Pearson HA, O'Brien RT, McIntosh S, Aspnes GT, Yang MM (1974). Routine screening of umbilical cord blood for sickle cell diseases. *JAMA* **227**:420–421.
29. Falletta JM, Woods GM, Verter JI, Buchanan GR, Pegelow CH, Iyer RV, Miller ST, Holbrook CT, Kinney TR, Vichinsky E (1995). Discontinuing penicillin prophylaxis in children with sickle cell anemia. *J Pediatr* **127**:685–690.
30. Platt OS, Thorington BD, Brambilla DJ, Milner PF, Rosse WF, Vichinsky E, Kinney TR (1991). Pain in sickle cell disease. Rates and risk factors. *N Engl J Med* **325**:11–16.
31. Platt OS, Brambilla DJ, Rosse WF, Milner PF, Castro O, Steinberg MH, Klug PP (1994). Mortality in sickle cell disease: life expectancy and risk factors for early death. *N Engl J Med* **330**:1639–1644.
32. Quinn CT, Rogers Z, Buchanan G (2004). Survival of children with sickle cell disease. *Blood* **103**:4023–4027.
33. Grosveld F, Dillon N, Higgs D (1993). The regulation of human globin gene expression. *Balliere's Clinical Hematology* **6**:31–55.
34. DeSimone J, Heller P, Hall L, Zwiers D (1982). 5-Azacytidine stimulates fetal hemoglobin synthesis in anemic baboons. *Proc Natl Acad Sci USA* **79**:4428–4431.
35. Ley TJ, DeSimone J, Noguchi CT, Turner PH, Schechter AN, Heller P, Nienhuis AW (1983). 5-Azacytidine increases gamma-globin synthesis and reduces the proportion of dense cells in patients with sickle cell anemia. *Blood* **62**:370–380.
36. Charache S, Dover G, Smith K, Talbot CC Jr, Moyer M, Boyer S (1983). Treatment of sickle cell anemia with 5 aza-cytidine results in increased fetal hemoglobin production and is associated with nonrandom hypomethylation of DNA. *Proc Natl Acad Sci USA* **80**:4842–4846.
37. Platt OS, Orkin SH, Dover G, Beardsley GP, Miller B, Nathan DG (1984). Hydroxyurea enhances fetal hemoglobin production in sickle cell anemia. *J Clin Invest* **74**:652–656.

38. Rodgers GP, Dover GJ, Noguchi CT, Schechter AN, Nienhuis AW (1990). Hematological responses in patients with sickle cell anemia treated with hydroxyurea. *N Engl J Med* **322**:1037–1045.
39. Charache S, Terrin ML, Moore RD, Dover GJ, Barton FB, Eckert SV, McMahon RP, Bonds DR (1995). Effect of hydroxyurea on the frequency of painful crises in sickle cell anemia. *N Engl J Med* **332**:1317–1322.
40. Steinberg MH, Barton F, Castro O, Pegelow CH, Ballas SK, Kutlar A, Orringer E, Bellevue R, Olivieri N, Eckman J, Varma M, Ramirez G, Adler B, Smith W, Carlos T, Ataga K, DeCastro L, Bigelow C, Sauntharajah Y, Telfer M, Vichinsky E, Claster S, Shurin S, Bridges K, Waclawiw M, Bonds D, Terrin M (2003). Effect of hydroxyurea on mortality and morbidity in adult sickle cell anemia: risks and benefits up to 9 years of treatment. *J Am Med Assoc* **289**:1645–1651.
41. Kinney TR, Helms RW, O’Branski EE, Ohene-Frempong K, Wang W, Daeschner C, Vichinsky E, Redding-Lallinger R, Gee B, Platt OS, Ware RE (1999). Safety of hydroxyurea in children with sickle cell anemia: results of the HUG-KIDS Study, a phase 1/11 trial. *Blood* **94**:1550–1554.
42. Wang WC, Wynn LW, Rogers ZR, Scott JP, Lane PA, Ware RE (2001). A two year pilot of hydroxyurea in very young children with sickle cell anemia. *J Pediatr* **139**:790–796.
43. Russell MO, Goldberg HI, Hudson A, Kim HC (1984). Effect of transfusion therapy on arteriographic abnormalities and on recurrence of stroke in sickle cell disease. *Blood* **63**:162–169.
44. Adams RJ, McKie VJ, Hsu L (1998). Prevention of first stroke by transfusion in children with sickle cell anemia and abnormal results on transcranial doppler ultrasonography. *N Engl J Med* **339**:5–11.
45. Saiki RK, Scharf S, Faloma F, Mullis KB, Horn GT, Erlich HA, Arnheim N (1985). Enzymatic amplification of beta-globin genomic sequences and restriction site analysis for diagnosis of sickle cell anemia. *Science* **230**:1350–1354.