The Hereditary Haemolytic Anaemias

The Croonian Lecture, 1973

J. V. DACIE, MD, FRCP, PRCPath, FRS, Professor of Haematology, Royal Postgraduate Medical School, University of London

This review of the hereditary haemolytic anaemias (HHAs) makes it obvious that their more complete understanding at the present time has stemmed very largely from the increasing application of biochemical knowledge and techniques to medicine. The HHAs illustrate, too, how well-recognised clinical syndromes are being broken down into more and more separate entities, and they provide a remarkable illustration of the relationship between molecular variation and disease. The subject is now a very large one, and I can do no more than deal rather superficially with it.

Dr William Hunter was perhaps the first to coin the term haemolytic. In his book *Pernicious Anaemia*, published in 1901, he referred to the presence of yellow spherical microcytes in the blood; he also described how he had been able to produce similar bodies by the addition of destructive agents to normal blood and concluded that their presence marked the anaemia—in pernicious anaemia—as being due to excessive destruction of blood and not to deficient formation and that 'they denote the anaemia to be haemolytic, not haemogenic, in its origin'. It was about this time that the disorder now referred to as hereditary spherocytosis (HS) was becoming clinically recognised. Wilson had described, in 1890, patients with hereditary splenomegaly and jaundice; later, in a follow-up report published with Stanley in 1893, the presence of anaemia was mentioned. One of the patients died, and death was considered as 'due to active haemolysis of splenic origin'.

The first detailed account of hereditary spherocytosis in the Continental literature seems to be that of Minkowski (1900) but it was Chauffard (1907) in Paris who described the increased red cell osmotic fragility—hence the eponym Minkowski-Chauffard which was used for many years to distinguish hereditary (or congenital) haemolytic anaemia from the acquired form of Hayem and Widal. The microcytosis of HS had in fact been described (and illustrated) many years earlier by Vanlair and Masius (1871) in a paper published in Belgium entitled 'De la Microcythémie'. They incidentally referred to the microcytes as 'globules atrophiques' and suggested that excess

bile pigment was derived from them. Thus, the main facts about the clinical syndrome of hereditary haemolytic anaemia were being gathered together at about the turn of the nineteenth century. At that time there was no suggestion that hereditary haemolytic anaemia (or congenital icterus) comprised more than one disorder.

It was not until 1910 that the next highly significant event in the history of the hereditary haemolytic anaemias took place. This was the publication by Herrick of a paper entitled 'Peculiar elongated and sickle-shaped red blood corpuscles in a case of anaemia'. It was the first description of sickled red cells. The term sickle-cell anaemia was, however, not introduced until 1922, and it was in this paper that Mason suggested that the disorder might be confined to the negro race.

In 1928 there was a further advance: Hijmans van den Bergh described the association of elliptical red cells and haemolytic anaemia. Thus, when I became a medical student in the early 1930s, there were three known types of hereditary haemolytic anaemia: hereditary spherocytosis, hereditary elliptocytosis and sickle-cell anaemia. Almost nothing was known of their pathology and pathogenesis, and one major group, the important non-spherocytic group, had been overlooked. It was not until 1947 that Russell Haden of Cleveland published his well-known paper entitled 'A new type of hereditary hemolytic jaundice without spherocytosis'; Haden reported, incidentally, that splenectomy, carried out on one patient, had failed to alter the course of the disease. Since 1947 the pace of advance in knowledge has been accelerating and in the last decade or so it has become almost breathtaking. Table 1 illustrates the growth of knowledge over the last 100 years or so.

It is now conventional and useful to classify the HHAs into three groups:

TABLE 1. Growth of knowledge of the hereditary haemolytic anaemias

Pre-1900	Early clinical accounts	Vanlair and Masius, 1871; Wilson, 1890; Wilson and Stanley, 1893
1900–1939	Complete description of clinical and haematological findings. Recognition of sickle-cell anaemia and hereditary elliptocytosis	Minkowski, 1900; Chauffard, 1907; Herrick, 1910; Meulengracht, 1921; Mason, 1922; Gännslen *et al.*, 1925; Hijmans van den Bergh, 1928
1940–1959	Early studies on pathogenesis. Recognition of hereditary non-spherocytic haemolytic anaemias. Discovery of molecular basis for Hb-S	Ham and Castle, 1940; Dacie and Mollison, 1943; Haden, 1947; Pauling *et al.*, 1949; Cathie, 1952; Selwyn and Dacie, 1954
1960–1973	Enormous expansion in knowledge; description of many new types of HHAs. Recognition of the unstable Hb diseases	Valentine *et al.*, 1961; Grimes and Meisler, 1962; Jacob and Jandl, 1964; Jacob, 1972

those associated apparently with red cell membrane abnormalities, enzyme deficiency HHAs, and haemoglobinopathies, respectively. HS, non-spherocytic HHA and sickle-cell anaemia are the prototypes. Each group comprises several to very many entities.

GROUP 1. HHAs ASSOCIATED WITH RED-CELL MEMBRANE DEFECTS (Table 2)

Hereditary spherocytosis, *la microcythémie* of Vanlair and Masius, the familial acholuric jaundice of British authors, is by far the most important entity in this group. However, determination of the exact nature of the red-cell membrane abnormality or abnormalities has proved a difficult task. Spherocytosis is a dynamic change. The youngest red cells in HS, the reticulocytes, are probably almost normal in size and shape, i.e. disc-like; it is as the cells circulate in the blood, and particularly if they become arrested in the spleen pulp, that they lose surface and to some extent shrink and become more and more spheroidal, i.e. spherocytic, and the haemoglobin concentration within the cells rises.

TABLE 2. Hereditary haemolytic anaemias: Group 1

Red cell membrane defects
Hereditary spherocytosis, ? not homogeneous
Hereditary elliptocytosis
Hereditary ovalocytosis, ? distinct from HE
Other rarer defects, e.g., hereditary stomatocytosis

Spherocytosis explains the increased osmotic fragility and the lack of plasticity of the red cells, so well demonstrated by LaCelle and Weed (1969). It is this lack of plasticity (i.e. abnormal rigidity) which leads to the sequestration of the red cells in the spleen where the retention of the cells in an unfavourable metabolic environment leads to their premature death, and the signs of increased haemolysis in the patient.

The central problem in HS is the cause and mechanism of the loss of cell surface leading to the spherocytosis. After a number of false trails it now seems likely, according to Jacob (1972), that the fundamental structural defect involves the microfilament protein components of the red cell membrane, a concept which, not involving any enzyme defect, fits in well with the dominant inheritance of HS. Ultracentrifuge studies of aggregated solubilised membrane protein have pointed to at least two types of protein abnormality.

Jacob suggested that the normal biconcave disc-like shape of red cells results from the interplay of two types of microfilaments—a lattice work of fine structural filaments and contractile actomyosin-like filaments. An orderly non-tetanic contraction of the latter elements upon a normal lattice framework produces, he suggests, the normal plastic red cell. Abnormality in either type of protein would, he considers, produce relatively rigid spherocytes (Table 3).

TABLE 3. Pathology and pathogenesis of hereditary spherocytosis (after Jacob, 1972)

Mutation leading to abnormality in filamentous membrane protein

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Loss of red cell surface
by invagination or
by external budding of fragments (lipid loss)

↓

Microspherocytosis

↓

Decreased membrane plasticity

↓

Sequestration within pulp of spleen

↓

Failure of Na pump compensation
(? lack of substrate glucose)

↓

Osmotic swelling and lysis

N.B. Increased Na permeability is probably secondary to membrane abnormality. In the blood stream increased Na pumping can compensate for this

An interesting feature of HS is the variability of its clinical expression; this ranges from severe haemolytic anaemia producing haemolysis in early life, and even neonatal kernicterus, to a mild, continuous and well-compensated haemolysis producing hardly detectable jaundice in an adult, only discovered accidentally on routine medical examination. Usually, although not invariably, the expression of the disease is consistent within families. The questions arise as to whether HS is one or several (or many) diseases, caused by one, several (or many) mutations, which give rise to a whole variety of subtly different structural abnormalities resulting in different degrees of damage to the red cells, and to what extent the expression of the disorder in an individual affected with the same genetic abnormality can be markedly modified by other genes, including those of the apparently normal parent of an affected heterozygote? The existence of a variety of different abnormalities, presumably each based on a distinct mutation, can be solved only by demonstrating clear biochemical differences between sibships; as already mentioned, Jacob (1972) has evidence suggesting on this basis at least two types of HS.

But there is collateral evidence, too, for the alternative mechanism—variable expression of the same abnormal gene. In hereditary elliptocytosis (HE), generally agreed to be very similar to HS, Jenssen *et al.* (1967) in Iceland traced 50 cases and provided evidence that the patients had all probably inherited the abnormality from a single ancestor. Despite this, the expression of the disorder varied from a harmless trait to active haemolytic anaemia and the morphology of the red cells, i.e. the degree of elliptocytosis, varied to some extent. The nature of the red-cell abnormality of HE has not been so fully studied as that of HS. But the basic abnormality or abnormalities seem likely to be analogous, although elliptocytosis *per se* appears often to be harmless from the point of view of red cell survival. It is interesting to note that reticulocytes are round in contour in HE. How a round reticulocyte transforms itself into a markedly elliptical cell on maturation is an intriguing problem for which an answer has not yet been provided.

GROUP 2. THE ENZYME DEFICIENCY HEREDITARY HAEMOLYTIC ANAEMIAS

Although it is true that familial cases of hereditary haemolytic anaemia in which the osmotic fragility of the red cells was not increased and in which splenectomy did not result in clinical cure had been recognised in the 1930s, it was not until after Haden's publication in 1947 that the hereditary non-spherocytic haemolytic anaemias became widely recognised as entities clearly distinguishable from HS and HE. Studies in the early 1950s based on the autohaemolysis test provided evidence of a metabolic abnormality and for heterogeneity in non-spherocytic cases (Selwyn and Dacie, 1954), but it was not until about 1960 that the first chronic HHAs based on enzyme defects were described—the G-6-PD type by Newton and Bass (1958) and pyruvate-kinase (PK) deficiency by Valentine *et al.* (1961). These provide the prototypes of the two main groups of enzyme-deficiency HHAs (Table 4).

The discovery of chronic haemolytic anaemias due to G-6-PD and PK deficiency has been followed by a flood of new observations. Many different types of non-spherocytic haemolytic anaemias are now known to exist, based on deficiencies of one or other of most of the enzymes active in the Embden-Meyerhof pathway and the hexose monophosphate shunt. It is now known, too, that the apparent enzyme deficiency, as measured in laboratory tests, is brought about in most cases, perhaps in all, by the presence of molecular variants of a particular enzyme, of impaired efficiency, rather than by failure of production of the normal enzyme. In the case of G-6-PD, very many different enzyme types have been defined, each presumably due to a discrete amino acid substitution. It is important to note that the molecular variants

TABLE 4. Hereditary haemolytic anaemias: Group 2

The enzyme-deficiency haemolytic anaemias

Defective enzymes of the Embden-Meyerhof pathway
- e.g. pyruvate kinase deficiency
- other rarer deficiencies—many types

Defective enzymes of the hexose-monophosphate shunt
- e.g. G-6-PD deficiency—many types
- other rarer deficiencies
  - e.g. glutathione synthetase deficiency and other types

of G-6-PD bringing about chronic hereditary haemolytic anaemia are distinct from the common allotype causing drug sensitivity and acute haemolysis (but not chronic haemolytic anaemia) in negro populations. Some recently described types of enzyme deficiency HHA are listed in Tables 5 and 6.

TABLE 5. Hereditary non-spherocytic haemolytic anaemias (HNSHA) due to defects of enzymes involved in anaerobic glycolytic (Embden-Meyerhof) pathway

Defective enzyme	*First described as cause of HNSHA by:*
Pyruvate kinase	Valentine *et al.*, 1961
Triosephosphate isomerase*	Schneider *et al.*, 1965
Hexokinase	Valentine *et al.*, 1967
Phosphoglyceratekinase†	Kraus *et al.*, 1968
Glucosephosphate isomerase	Baughan *et al.*, 1968
Phosphofructokinase‡	Tarui *et al.*, 1969
2,3-diphosphoglyceromutase	Schröter, 1970

* Associated with neuromuscular disorder
† May be associated with neurological disorder
‡ May be associated with myopathy

Review references: Jaffé (1970); Valentine (1970, 1971, 1972).

The inheritance of the enzyme-deficiency HHAs differs from that of HS and HE. Except in the G-6-PD HHAs, which are sex-linked, the enzyme-deficiency HHAs are manifest clinically only in homozygotes—although enzyme deficiency may be demonstrable as a laboratory phenomenon in heterozygotes. In G-6-PD deficiency, males (hemizygotes) are affected, and the incidence of overt haemolytic anaemia is thus many times greater in males than in the much rarer homozygous females. The other enzyme deficiencies are determined by abnormal autosomal genes, and male and female

TABLE 6. Hereditary non-spherocytic haemolytic anaemias (HNSHA) due to defects of enzymes involved in aerobic glycolysis (hexose monophosphate shunt)

Defective enzyme	*First described as cause of HNSHA by:*
Well-established conditions	
G-6-PD*	Newton and Bass, 1958
GSH deficiency	Oort *et al.*, 1961
GSH synthetase	Boivin *et al.*, 1966
γ glutamyl-cysteine synthetase	Konrad *et al.*, 1972
GSH peroxidase	Necheles *et al.*, 1969
Less well-established conditions	
GSH reductase	Löhr and Waller, 1962
6-PGD	Lausecker *et al.*, 1965

* Many different variants of G-6-PD have by now been described in association with HNSHA

heterozygotes, and female heterozygotes for G-6-PD deficiency, do not usually suffer from overt haemolysis because the presence of one normal gene results in sufficient normal enzyme production for red cell metabolism to continue almost, if not quite, unimpaired.

Other tissues usually escape the enzyme deficiency that impairs red cell metabolism. Thus, the enzyme activity in leucocytes in PK deficiency homozygotes seems to be normal, so much so that red-cell enzyme assays have to be carried out on blood carefully freed from leucocytes. This indicates that the PK in red cells and leucocytes are iso-enzymes which are under different genetic control. But this is not apparently true of all the enzymes that may be defective, and some types of G-6-PD deficiency affect leucocytes and other tissues in addition to red cells. Clinical evidence of involvement of systems other than the blood in certain enzyme deficiencies is additional evidence of this. Thus, deficiencies of triosephosphate isomerase and phosphoglycerate kinase in red cells have been associated with serious neuromuscular or neurological disorders.

PK deficiency is undoubtedly the commonest cause of a hereditary non-spherocytic haemolytic anaemia due to a defect of an enzyme of the Embden-Meyerhof pathway. Clinically, its expression is variable, but anaemia is often severe and most cases are diagnosed in infancy or early childhood. Exchange transfusion in the neonatal period and/or periodic transfusion in infancy and early childhood are required much more frequently than in HS and HE.

The way in which PK deficiency brings about haemolysis is not entirely understood, but failure to metabolise glucose and to maintain an adequate supply of ATP must be an important factor. Curiously enough, PK red cells

do not become notably spherocytic before their demise, although some may become markedly crenated and contracted; this is particularly noticeable after splenectomy. Another interesting feature of PK deficiency post-splenectomy is the very large number of reticulocytes (up to 80 per cent) that may be present. There is, in fact, reason to believe that reticulocytes survive much better than adult red cells. Not only is PK activity higher in reticulocytes than in mature red cells, but reticulocytes also have the advantage of being able to metabolise glucose via the Krebs cycle (Keitt and Bennett, 1966).

GROUP 3. THE HAEMOGLOBINOPATHIES

As already mentioned, our present knowledge of the chemical and molecular basis of the haemoglobinopathies stems from the work of Pauling *et al.* (1949). In their paper 'Sickle cell anemia, a molecular disease' they reported that they had found in haemoglobin solutions derived from sickle-cell disease patients an abnormal haemoglobin component, which they named Hb-S, that separated from normal haemoglobin when submitted to ultra-centrifugation in a Tiselius apparatus. This finding acted as a tremendous stimulus to research and now, a little less than 25 years later, more than 200 abnormal haemoglobins have been isolated and the responsible substitutions in the haemoglobin amino acid chains pin-pointed. But by no means all the abnormal haemoglobins lead to disease, and of those which do, not all lead to haemolytic anaemia. The haemoglobin Ms, for instance, cause congenital methaemoglobinaemia and certain high oxygen-affinity haemoglobins, for example Hb-Heathrow (White *et al.*, 1973), familial polycythaemia.

Hb-S is by far the most important of the abnormal haemoglobins that lead to haemolysis and anaemia. Fortunately, however, it is only in the homozygous state or when combined with thalassaemia or another abnormal haemoglobin such as Hb-C that important symptoms arise. Homozygous Hb-S disease, or sickle-cell anaemia, is an extremely serious affection and the cause of much suffering. Although at one time thought almost always to cause death in childhood or adolescence, an increasing proportion of patients now live to adult life, helped mainly by the improving facilities for general medical care. A recent potentially important line of research is that concerned with the possibility of the prevention of sickling by interfering by chemical means with the process of the lining-up of haemoglobin molecules at low oxygen tensions, which is the physical basis of sickling. A number of substances, e.g. urea and cyanate, can do this in the laboratory; the problem is to find a chemical that can be given in sufficient doses *in vivo* to be effective and that does not at the same time cause damage by binding on to normal proteins.

In fact, the results of trials now being carried out with sodium cyanate in the United States seem promising (Gillette *et al.*, 1972). Eventually, in the normal course of events, a proportion of the red cells of a patient with homozygous sickle-cell disease become irreversibly sickled. Irreversibly sickled cells are particularly dangerous in that their life-span is short and because their rigidity leads to blockage of capillaries and organ infarction.

The way in which Hb-S leads to a reduction of red-cell life-span is not wholly clear. In heterozygotes the cells survive normally, and sickling, although easily produced *in vitro*, does not occur to a significant extent *in vivo* except under exceptional circumstances, as, for example, by exposure to low oxygen partial pressure at high altitudes. In homozygous sickle-cell disease haemolysis is always present to a varying extent. The patients' main symptoms, such as abdominal or bone pain, usually stem from infarction due to vascular blockage rather than from anaemia due to haemolysis. The increased rigidity of sickled cells probably plays a part in the haemolysis and the abnormal haemoglobin may also interfere with red cell membrane function, but in exactly what way is obscure. The increased haemolysis in homozygous Hb-C disease also probably stems from increased membrane rigidity; this results from the diminished solubility of Hb-C and the increased viscosity of the abnormal haemoglobin in solution.

Hb-S and Hb-C are comparatively stable molecules, as are most of the haemoglobin variants. However, another important group of abnormal haemoglobins is notably unstable; and it is their instability that leads to haemolysis. These haemoglobins differ from the stable varieties in giving rise to haemolysis in heterozygotes. No homozygous cases have been described; indeed such an occurrence seems likely to be incompatible with life. The discovery of the unstable haemoglobins makes a fascinating story. The associated haemolytic anaemias were first described as Heinz-body anaemias, of apparent congenital origin, not, seemingly, being accountable by exposure to any toxic drug or chemical (Cathie, 1952). Later it became clear that these Heinz-body anaemias might occur in families and that they were inherited as a dominant; it was realised, too, that the Heinz bodies could be seen only in preparations of fresh blood if the spleen had been removed, and that, without splenectomy, the blood picture was that of a hereditary non-spherocytic haemolytic anaemia. Next it was demonstrated that, in certain cases of Heinz-body anaemia, heating red cell lysates at 50°C led to a marked precipitation of haemoglobin. From this observation it was deduced that an abnormal haemoglobin was present, and it was concluded that it was the instability of the haemoglobin (as demonstrated by the heating test) that led to the formation of Heinz bodies (Grimes and Meisler, 1962). Once it was clear that a peculiar type of

abnormal haemoglobin was present, the way was open for the demonstration of the amino acid substitution responsible. These studies led to the description by Carrell *et al.* (1966) of Hb-Köln in which methionine is substituted for valine at the FG5 (98) position of the β chain.

In the last few years at least 45 examples of unstable haemoglobins have been discovered and it is now obvious that the unstable haemoglobin diseases or congenital Heinz-body anaemias comprise a group of HHAs which, although rare, are by no means as rare as was once thought. Some very large sibships have been studied.

The clinical and haematological findings in the unstable haemoglobin diseases are of unusual interest (White and Dacie, 1971). The patients present with varying degrees of haemolysis, the severity of which depends on the site of the amino acid substitution and the nature of the substituent amino acid. If severe, haemolysis is usually observed in the first year of life. Severely affected patients often pass dark, almost black, urine, and may also be obviously cyanosed. The dark coloration of the urine is due to the presence of mesobilifuscins—dipyrrols derived from haem—and the cyanosis can be accounted for by the presence of small amounts of met- and sulphaemoglobin and by unusual amounts of reduced haemoglobin if, as sometimes happens, the abnormal haemoglobin has a reduced oxygen affinity. In the patient whose abnormal haemoglobin was named Hb-Hammersmith, the reduced oxygen affinity of her abnormal haemoglobin is a major factor in allowing her to lead a normal active life despite a haemoglobin level of about 7·5 g/100 ml and active haemolysis.

As already referred to, Heinz bodies are seen only in freshly drawn blood if the patient's spleen has been removed. *In vivo*, it seems that the Heinz bodies are removed from the red cells as the cells pass through the narrow communications, 'stomata', between the vascular spaces of the spleen pulp and the splenic sinuses. Heinz bodies are relatively rigid structures and it is thought that the spleen removes them by a process of 'pitting' in much the same way as it removes other inclusions from red cells such as large siderotic granules. The bodies stain well in Giemsa-stained sections of spleen; they do not stain for iron (White and Dacie, 1971).

The unstable haemoglobin disorders provide remarkable examples of the effects of molecular variation. Not only do they demonstrate once again how the substitution of a single amino acid by another in a chain of 146 amino acids—as in the β chain of haemoglobin—may result in most serious consequences for the patient, but instances are known in which the replacement of a single amino acid by several different substituents results, according to the chemical structure of the substituent, in distinct clinical syndromes (Table 7).

TABLE 7. Different effects of substitution of one amino acid by different substituents (from White and Dacie, 1971)

	Valine E 11 (β67) (*non-polar*) *substituted by:*	
Alanine (non-polar)	Side chain too short to make haem contact	Hb-Sydney; mild haemolysis
Glutamic acid (polar)	Side chain charged and long, reaches Fe^{++} of haem and oxidises it to Fe^{+++}	Hb-M Milwaukee; congenital methaemoglobinaemia
Aspartic acid (polar)	Side chain charged but too short to reach surface; gross distortion of haem pocket	Hb-Bristol; severe haemolysis

The essential feature of an unstable haemoglobin, which leads to its instability, is the presence of a substituent amino acid that either impairs the firm binding of haem within the haem pocket of a globin chain or interferes with the normal contacts between the globin chains. Loss of haem, or the entrance of water into the haem pocket, or the separation of globin chains, leads to a break-up of the haemoglobin molecule and the precipitation of the separated or haem-depleted globin chains as Heinz bodies.

It should be added that a number of types of HHA have been described in recent years the pathology of which is largely unknown and which do not fit comfortably in the three major groups of disorders. Some may depend upon abnormalities of the red cell membrane; if they do, little or nothing is known about the nature of the lesions. The disorders listed in Table 8 are all rare but

TABLE 8. Recently described types of hereditary haemolytic anaemia

Miscellaneous types		
'Stomatocytosis'	Lock *et al.*, 1961	Dominant
Adenosine-triphosphatase (ATP-ase) deficiency	Harvald *et al.*, 1964	? Dominant
Disorder ? distinct from stomatocytosis, with 'cold autohaemolysis'	Miller *et al.*, 1965	?
Disorder ? distinct from stomatocytosis, with extensive cation permeability	Nathan *et al.*, 1966	?
Disorder associated with increased K permeability	Brain *et al.*, 1968	?
Disorder with increased red cell phosphatidyl choline content	Jaffé and Gottfried, 1968	Dominant
Disorder with high ATP	Busch and Heimpel, 1969	?
Adenylate kinase deficiency	Szeinberg *et al.*, 1969	Autosomal recessive
Disorder with low ATP	Paglia *et al.*, 1970	Dominant

their existence should be borne in mind when considering an unusual case. The titles are descriptive and tentative.

DIAGNOSIS AND TREATMENT

Some clinical points that may enable the clinician to make a shrewd clinical diagnosis have already been mentioned. These include the sex of the patient, the age of onset, the patient's racial origins and the apparent inheritance pattern. The presence or absence of pain in the abdomen or elsewhere, the size of the spleen, the passing of very dark urine, and the presence of cyanosis may also provide clues to the nature of the patient's illness. However, confirmation of the diagnosis has to rest on laboratory observations, e.g., the blood count, red cell morphology, osmotic fragility, and autohaemolysis tests in HS and allied disorders; the results of screening tests and definitive enzyme assays in the enzyme deficiency haemolytic anaemias, and tests for sickling, haemoglobin electrophoresis, and haemoglobin instability tests in the haemoglobinopathies. It is only by applying the appropriate tests and by possessing an understanding of the basis of the tests, and by having some experience in the interpretation of the results they give, that the haematologist can arrive at the diagnosis.

A correct diagnosis is not only a matter of intellectual satisfaction; it has a bearing, too, on prognosis and treatment. In relation to treatment the question that most often arises is whether or not the spleen should be removed and, if so, at what age. It is well known that patients with HS derive great benefit from splenectomy; they are almost always clinically cured, and the same seems true of patients with HE. In the enzyme-deficiency haemolytic anaemias splenectomy is much less helpful, but it may not be without value, and in PK deficiency there is increasing evidence that the operation is almost always followed by some improvement. Splenectomy should certainly be seriously considered in any PK-deficient patient requiring regular transfusions to maintain the haemoglobin at a reasonable level. Fortunately, patients with PK deficiency tolerate low haemoglobins remarkably well, an important reason for this being the shift to the right of the oxygen dissociation curve of haemoglobin resulting from the increased content of 2,3,DPG in their red cells. Therefore, in coming to a decision to undertake splenectomy, the patient's symptoms rather more than his haemoglobin level should be taken into account. Splenectomy should never, however, be undertaken lightly in any type of case, for at least two reasons. Although the immediate mortality of the operation must be very small, post-operative complications, notably sepsis and/or thrombo-embolism, are not infrequent. In relation to thrombo-embolism an important point is whether or not the increased haemolysis is

likely to cease after splenectomy, and the answer to this depends upon making a correct diagnosis. There is evidence that the height of the post-splenectomy platelet count and the persistence of thrombocytosis are directly related to the degree to which increased erythropoiesis persists after splenectomy, and it seems likely that persistent thrombocytosis is a factor in the genesis of post-operative thrombo-embolism. Our own experience is that serious episodes of thrombo-embolism are most frequent in patients who do not achieve normal haemoglobin levels and have high reticulocyte counts after splenectomy; and and it is in this group of patients that persistent thrombocytosis is found (Hirsh and Dacie, 1966).

The second reason for being cautious in advocating splenectomy is the well-known risk of the occurrence of serious sepsis, including septicaemia, subsequently; this risk seems to be greatest in infants and young children, in whom the spleen is perhaps relatively more important in the defence against infection than it is in later life.

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Waggoners at Sea

The College library is well known for the breadth of learning to be found on its shelves. The President, with his interest in sailing, can browse through the earliest printed charts of the British coast. These were first published in 1584 by Lucas Waghenaer, a retired Dutch pilot whose navigational findings were engraved in Amsterdam. His work was published in England by Philip Ashley who translated the text and had new plates engraved. The engraving was the finest to be done in England at that time and was carried out by engravers from the Low Countries, then the seat of superlative engraving. Depending on the degree of political and religious odium they incurred, the engravers commuted across the Channel. The *Mariner's Mirrour* was published in 1588, a few months after the defeat of the Armada. The charts were accurate as well as decorative. Distances were given in English, Dutch and Spanish leagues, presumably to avoid nationalist offence. They also showed the coast-line in profile, a device still used by the Admiralty. The success of the charts was immediate and for the next 150 years British sailors referred to any sea chart as a 'Waggoner'.