Sickle cell disease in Saudi Arabs in early childhood

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SUMMARY Haemoglobin electrophoresis screening of 2341 infants from the oases of eastern Saudi Arabia, performed in an attempt to detect cases early and then to follow up and give better management to patients with sickle cell disease, showed 20% with S-trait and 43 with sickle cell disease (37 HbSS and 6 S- β^0 thalassaemia). On follow-up from birth (or from 3 months) for a mean of 3½ years there was more morbidity and mortality than in matched Saudi Arab controls, but these rates were lower than for affected black infants in the US or Jamaica. Serial haemoglobin levels were 1 g/dl lower than for controls; height and weight increases were the same. Mean fetal haemoglobin level was 32% at 4-5 years. Half of a subsample of 23 cases had evidence of coexistent α -thalassaemia which appeared to protect against functional asplenia, judged by the presence of Howell-Jolly bodies; however there was no difference in degree of anaemia or clinical course. Early screening is recommended for infants at risk for sickle cell disease, and comprehensive care should be given even if the infant has the less severe type.

There has been recent emphasis¹ on the need for prospective studies in sickle cell disease (SCD), especially in infancy and early childhood when morbidity and mortality rates are believed to be high. Several studies¹⁻³ indicate there is considerable serious illness and death during the first few years.

Homozygous sickle cell anaemia (HbSS) in Saudi Arabs from the oasis areas of the Eastern Province of the country is characterised by an associated unusually high level of fetal haemoglobin (HbF), and this is believed to account for the relatively mild clinical course.⁴⁻⁵ It was suggested recently that the HbSS in these people represents a useful model for what might be achieved if HbF production could be manipulated after birth,⁶ so a full understanding of the natural history of HbSS in Saudi Arabs is of interest. The present study was designed to complement and extend our understanding⁵ of the natural history of SCD in 270 Saudi Arab subjects of all ages. An overall review suggested there was a mild clinical course even in infancy, but early morbidity and mortality rates were not studied. A prospective study, based on early detection by means of screening all infants before significant morbidity or mortality occurs, is

needed to evaluate the problem in infants and children.

We report here the results of such a study, which includes the clinical and haematological findings of the 43 SCD subjects (37 HbSS) identified by a screening programme and followed up for a mean of 3½ years. We compare the patients with SCD with an Arab control group, and with reported findings in blacks. The screening programme also gives data on the frequencies of the haemoglobinopathies occurring in Saudi Arabs from the Qatif oasis area.

Subjects and methods

The Arabian American Oil Company (Aramco) provides medical care for its employees and their families. A screening programme for identifying and following up all infants from the oasis areas, and therefore at special risk of having a haemoglobinopathy, has been conducted at the Maternal Child Health Clinic of Dhahran Health Centre since April 1973. Because nearly one-quarter of infants are born at home, and still others are born in facilities other than the main hospital in Dhahran, cord

blood screening was impracticable. All Aramco-dependent infants born at home (or in outside facilities) are routinely followed up in the clinic, the mean age for the first visit being 3 months, and as significant morbidity and mortality from SCD occurs after age 6 months,^{2 7-8} 6 months was the time chosen for screening all infants.

A capillary blood sample was obtained for the initial haemoglobin electrophoresis. Whole blood was haemolysed by freezing or by saponin 0.1% and applied to cellulose acetate in tris EDTA borate and barbital buffers at pH 8.9 using 250 volts over 30 minutes. This method⁹ adequately separates haemoglobins A and S, and S and F. Follow-up quantitative electrophoresis on cellulose acetate (Gelman Sepraphore III System, Gelman Instrument Company, Ann Arbor, Michigan) using a Beckman R110 microzone densitometer (Beckman Instruments, Fullerton, California) was later carried out on infants found to have an SF pattern, indicative of SCD. We have shown that, in the absence of HbA, this method gives a more accurate HbF estimate than alkaline denaturation, when the HbF level is higher than 15% as in our patients.⁶ Previous studies⁴⁻⁶ showed the expected β^6 Glu $\rightarrow$ Val substitution; the HbS band on electrophoresis in this population is invariably associated with a positive sickling test; agar-gel electrophoresis (pH 6.0) did not show HbD; and the distribution of HbF, evaluated by acid elution or immunofluorescence, was always heterocellular.

In 2341 infants screened there were 43 cases of SCD, 37 with HbSS and 6 with sickle cell- β^0 thalassaemia (S- β^0 thal), and there were 467 (20%) infants with S-trait (Table 1). The presence of S- β^0 thal was established by family studies, the only reliable method in this population; we had previously found that 8 out of 14 patients with proved S- β^0 thal did not have raised Hb A₂ levels owing to the high levels of HbF with which Hb A₂ is inversely correlated.¹⁰ All the fathers of the HbSS patients were shown to have an S gene as were all the mothers, except 2 who were not available for checking.

Table 1 Results of screening programme for haemoglobinopathies in 2341 infants, 1973-8

Haemoglobin pattern	Diagnosis	Infants n = 2341	
		No	%
(F)A	Normal*	1824	77.9
S(F)A	Sickle cell trait	467	20.0
SF	Homozygous sickle cell anaemia	37†	1.58
SF	Sickle- β^0 thalassaemia	6‡	0.25
C(F)A	Presumed haemoglobin C trait	7	0.29

*Includes β -thal trait, †includes one set of identical twins, ‡includes three siblings in one family.

Controls were matched for sex, year of birth, and village or town of origin, and had either an AA or AS haemoglobin pattern. Both subjects and controls were routinely followed up in the clinic at intervals of about 1 to 3 months for a mean of 3½ years (range 1 to 5 years). Serial blood haemoglobin (Hb), haematocrit, and reticulocyte values were assessed; the presence or absence of Howell-Jolly bodies was determined by search of about 3000 red cells, and red cell indices were measured by means of a Coulter Counter S model for 23 cases of HbSS. Weights and heights were taken by the clinic nurses; routine physical examination was by clinic physicians; paediatric specialist consultation was readily available for any problem in outpatients or inpatients. The 6 S- β^0 thal cases are combined with the HbSS cases for clinical evaluation unless otherwise stated; their haematological data are excluded from the tables showing these values.

Statistical comparisons have used the standard Student's *t* test, or if the exact probability is given, Fisher's exact 2-tailed test for 2 × 2 tables.

Results

Haematological data. The anaemia, during the first 3 years, was particularly pronounced at ages 9 to 12 months when the mean blood Hb was 8.6-8.8 g/dl. At 6 months and during the 27- to 36-month period the mean blood Hb level was 9.4-9.6 g/dl. The mean haematocrit was 29.6-30.9% on serial measurements from 6 to 36 months. Controls had a 1 g/dl higher blood Hb level. Reticulocytosis in HbSS patients was greater from age 12 to 36 months than before 12 months, mean values being 5.4 compared with 3.6%; considerable individual variation was noted. About half the HbSS infants who had their red cell indices measured, at a mean age of 44 months, exhibited a distinctly low mean corpuscular haemoglobin of less than 23 pg and a low mean cell volume. This microcytic hypochromic group, with presumed coexistent α -thalassaemia (α -thal), is compared with those with normal indices (Table 2). Both groups had unusually high HbF levels ranging from 9 to 41.5% but the mean HbF of 29% in the microcytic, hypochromic group was significantly lower than the mean level of 35% in the group with normal indices ($P < 0.02$). There was no significant difference between the two groups in blood Hb, reticulocytosis, or, interestingly, mean corpuscular haemoglobin concentration. The haemoglobin, reticulocyte, and HbF values of the 6 infants with S- β^0 thal were not significantly different from the HbSS cases.

The age of onset and frequency of functional asplenia was assessed in 23 HbSS infants (ages

Table 2 Haematological findings in a subsample of 23 infants with HbSS

Age (years)	Hb (g/dl)	MCH (pg)	MCV (fl)	MCHC (%)	Reticulocytes (%)	HbF (%)
<i>Microcytic hypochromic group</i>						
3 5/12	9.0	19.8	61	32.4	4.2	29.2
2 6/12	10.0	21.7	62	35.2	2.4	30.6
1 6/12	9.0	21.0	63	33.1	3.0	36.0
2 11/12	9.6	19.5	64	30.6	3.6	34.8
4 6/12	10.2	19.8	64	30.9	2.0	39.3
3 9/12	8.0	20.1	64	31.3	10.0	17.0
3 9/12	9.2	20.2	64	31.9	5.2	26.4
3 3/12	10.3	21.6	67	32.5	3.2	21.8
5 6/12	12.0	22.3	67	33.5	4.8	27.1
5	9.6	22.6	67	33.9	3.8	32.0
3 3/12	9.7	22.3	68	32.9	4.2	24.3
1 7/12	8.0	22.1	75	29.8	3.2	29.7
<i>Mean (±SD)</i>						
3 5/12	9.55 (±1.08)	21.08 (±1.15)	65.50 (±3.71)	32.33 (±1.53)	4.13 (±2.07)	29.0 (±6.24)
<i>Normochromic normocytic group</i>						
1 8/12	8.2	26.1	75	34.7	12.3	32.0
3 1/12	11.4	25.5	78	32.5	3.8	9.0
5 2/12	10.6	25.8	77	33.8	2.0	34.0
5	10.8	25.9	77	34.0	4.4	40.0
5 3/12*	7.2	25.9	79	33.2	11.8	30.2
2 4/12	10.6	24.1	81	31.1	3.2	31.8
4 6/12	10.0	26.0	83	31.4	—	35.2
2 6/12*	8.6	27.7	86	32.3	18.2	34.0
4 9/12	9.8	29.3	86	33.7	4.9	41.5
3 6/12*	8.3	28.5	87	33.1	3.2	35.7
5 6/12*	9.1	30.5	92	33.5	22.0	21.4
<i>Mean (±SD)</i>						
3 11/12	9.51 (±1.32)	26.86 (±1.93)	81.91 (±5.32)	33.03 (±1.11)	8.58 (±7.09)	34.98 (±3.7)
<i>Mean (±SD) for 20 controls†</i>						
	24.53 (±2.38)	74.15 (±6.21)	32.74 (±1.23)			

MCH=mean corpuscular haemoglobin, MCV=mean cell volume, MCHC=mean corpuscular haemoglobin concentration, HbF=fetal haemoglobin. *Cases with Howell-Jolly bodies present. †Inpatients without known blood disease and matched for age. They also came from the oases; their values for red cell indices are midway between those for the microcytic and the normocytic groups as would be expected if half had α -thal.

18–66 months) by examining blood smears for Howell-Jolly bodies. Only 4 showed these red cell inclusions, 2 of whom had splenomegaly. Slight splenomegaly was found in one-quarter of all 37 infants with HbSS, most often between ages 1 and 2½ years although 3 patients had palpable spleens between ages 6 and 12 months.

Complications in the first 3 years. Complications specific for SCD and significant infections during the first 3 years of life were studied in 42 patients (27 boys, 15 girls), and in 42 matched controls. One infant with HbSS was excluded from the review of clinical problems because he also had a large patent ductus arteriosus which probably was responsible for his frequent pneumonias. The ductus was ligated at age 30 months with a subsequent pronounced decrease in the number of infections.

Vasculo-occlusive crises occurred rarely and were generally slight. Two patients had the hand-foot syndrome, one of whom was admitted to hospital.

Hypoplastic crises sufficiently severe to require blood transfusion occurred in one patient during the first 3 years, and a slight fall of blood haemoglobin of about 1.5 g/dl, in association with infections, was noted in several other patients but resolved without transfusion. Two patients, identical twins, each had an acute febrile illness associated with severe hypoplastic crises but at an older age—4½ years. In general a fall in blood Hb level in the SCD patients (requiring transfusions in 2) was more often associated with an increased reticulocytosis (above the steady state level) and was probably related to a coexistent G6PD deficiency which was found in 19 of 38 SCD patients and in 15 of 35 controls. In most of these instances no specific cause of the haemolysis, other than infection, was noted. Thus altogether 3 patients required blood transfusions for Hb <6 g/dl, 2 of whom had blood once when less than 12 months of age, and 1 of whom had a transfusion on three occasions, at ages 15, 17, and 25 months. There was no case of acute splenic sequestration crisis in the present study,

Table 3 Conditions resulting in admittance to hospital for 14 out of 42 patients with sickle cell disease under age 3 years

Condition	No of times admitted
Gastroenteritis	8
Respiratory (acute)*	5
Septicaemia†	2
Hand-foot syndrome	1
Hypoplastic crisis	1
Anaemia and fever	1
Osteomyelitis	1
Other‡	7

*Includes pneumonia, bronchiolitis, croup with otitis media. †Proved by blood culture 1; presumed 1. ‡Includes cellulitis, viral hepatitis, malnutrition.

Figure Serial weight and height measurements in 14 Saudi Arabian boys with sickle cell anaemia (SS) and matched controls. A similar pattern of normal growth was found in 7 girls with SS.

although one typical case has been observed in the Aramco population.

One patient had osteomyelitis and one had salmonella septicaemia. Another patient with presumptive septicaemia died suddenly. Gastroenteritis and acute respiratory infections were the most common causes for being admitted to hospital (Table 3). Fourteen of 42 patients with SCD were in

hospital because of illness during the first 3 years, compared with 6 of the controls. The difference is not significant ($P = 0.07$), but the difference is significant if the 37 HbSS patients alone, of whom 14 were in hospital, are compared ($P = 0.02$). The first and second years of life appeared to be equally at risk for illness leading to admittance to hospital. All except 7 patients received antibiotics for illnesses treated in the outpatient clinic. Both the patients and controls were treated in the clinic with antibiotics for an average of $3\frac{1}{2}$ different illnesses during the first 3 years. Growth, in the infants with HbSS, as reflected by the mean weights and heights of 14 boys and 7 girls (all having satisfactory measurements at least to age 2 years) was not significantly different from that in controls (Figure). Clinical assessment of development did not show any difference from the controls. Only 3 out of 27 infants with SCD who had chest x-rays had cardiac enlargement, compared with 1 out of 14 controls.

There were 2 deaths. One child, at age 25 months, had vomited for 6 hours and appeared severely ill with drowsiness, cold clammy skin, and pallor; he died 30 minutes after admission. The spleen was not enlarged; a single blood culture was negative, but septicaemia seemed likely on clinical grounds. The other fatal case, who died at age 35 months, had had diarrhoea and fever for 5 days without treatment, was greatly dehydrated, and was dead on arrival, probably from gastroenteritis.

Discussion

Serious complications and also functional asplenia, in blacks with SCD, are seen only after age 6 months,^{1 3 8 11} making it practical to screen during the first half year as well as at birth. Although we were unable to perform cord blood screening, which may be ideal, this was not a serious problem for this prospective study since patients were followed up from birth in hospital-delivered cases (three-quarters of all), and from about age 3 months in the remainder.

We had previously noted⁴⁻⁶ the presence of an unusually high level of HbF (mean 22 to 26%) in Saudi Arabs (children and adults) with HbSS; the present study shows the presence of an even higher level during the first few years of life (Table 2). The mean level of HbF of 32% even after age 3 years is in striking contrast to that in most other populations.^{1 12} In most other populations the HbF, even though falling more slowly after birth than normally, was generally less than 15% by age 1 year and less than 9% by 3 years.¹² Lower levels of HbF have been associated with lower blood Hb levels, increased numbers of irreversibly sickled cells, and

the presence of Howell-Jolly bodies.^{1 12-13} The likely role of the high level of HbF in the relatively mild disease in Saudi Arabs has already been reviewed.⁴⁻⁵

In the present study the possible role of α -thal as an ameliorating factor was also considered, with the knowledge that half of the oasis population has high Hb Bart's at birth which is indicative of α -thal.¹⁴ The microcytic hypochromic blood findings in half of those HbSS cases in which indices were measured is therefore not unexpected. A similar proportion with presumed α -thal was found in age-matched inpatients without blood disease. Additional evidence in favour of the low mean corpuscular haemoglobin and mean cell volume being due to coexistent α -thal is the finding of a low HbS level (range 23 to 31%) in at least one parent of all but a single case in the microcytic hypochromic group. Iron studies were only available in a few; 8 of the 12 patients had been given iron treatment empirically some time before the indices were obtained. Comparison of mean blood Hb levels and of other indicators of severity of the HbSS—such as complications and illnesses leading to being admitted to hospital—failed to show any significant differences between those with and those without coexistent α -thal. However, a large number needs to be evaluated for valid conclusions.¹⁵ Certainly coexistent α -thal is not the cause of the unusually high HbF levels, and the finding of a significantly lower HbF level in the microcytic hypochromic group would suggest α -thal actually reduces the increase in HbF that occurs in these cases of SCD. Interestingly and significantly ($P = 0.037$), none of the α -thal group had Howell-Jolly bodies present, and those with these inclusions had significantly higher mean corpuscular haemoglobin and mean cell volume compared with the remaining 19 subjects ($P < 0.001$). The 4 cases with Howell-Jolly bodies had significantly lower blood Hb and higher reticulocytosis, but the mean corpuscular haemoglobin concentration was no different from other subjects. It is possible that coexistent α -thal facilitates the removal of Howell-Jolly bodies from the red cells or protects against functional asplenia. The report¹⁶ of the rare presence of irreversibly sickled cells in 5 black HbSS cases associated with α -thal supports the latter hypothesis. Despite this observation no amelioration of the clinical course was evident. Three other reported cases¹⁷⁻¹⁸ of α -thal associated with HbSS had a mild clinical course but also had raised HbF levels.

The presence of Howell-Jolly bodies is considered a reliable and clinically practicable indicator of loss of splenic function.^{1 12} While only one-sixth of the Saudi Arab infants with HbSS had Howell-

Jolly bodies when studied at age 18 months or older (mean 44 months), patients of African descent with HbSS are reported to have lost their splenic function in two-thirds to four-fifths of cases after age 1 year.^{1 12} This helps to explain the disparity in mortality during the first two years, reported as 13% for Jamaicans,³ compared with none for Saudi Arabs ($P = 0.04$). It is, nevertheless, of concern if any proportion of patients are without splenic function.

In the present study there were 2 patients with either proved or presumed bacteraemia: one died shortly after admission and the other had successful treatment of the salmonellosis with antibiotics. Each was between 2 and 3 years of age. The fatal case had an unusually severe course during his second year, being in hospital three times for infections and once for a hypoplastic crisis. The control group suffered no deaths, and only 6 needed to be admitted to hospital, distinctly fewer than for the HbSS subjects.

We showed previously that there is a 34-fold increased risk (compared with the general population) of pneumococcal meningitis for the Saudi Arab HbSS subject considering all age groups.⁵ The importance of pneumococcal bacteraemia and meningitis in HbSS cases in the US is well known.^{11 19-20} Since there is good evidence²¹⁻²² for the efficacy of polyvalent pneumococcal vaccine in subjects aged at least 2 years, we began a vaccination programme, in 1978, using a 14-valent preparation (Pneumovax). Penicillin prophylaxis from age 6 months up to age 6 years has been recommended²³ because of the crucial period before the pneumococcal vaccine is effective (less than 2 years) and also because of vaccine failures. The effectiveness of such prophylaxis, even when compliance is not a problem, is unproved; trials are in progress.²³ We have not used penicillin prophylactically, and thus far have had no vaccine failures in 118 cases (all ages).

It is worth stressing that early screening for SCD is important, and that continuous comprehensive care is required if early detection is to be of value. This is true even for the less severely affected subject such as the Saudi Arab child. It is evident that vigilance, family education, and a trial of pneumococcal vaccine are all indicated.

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Received 4 December 1979