Clinical Topics

Evaluation of eight and a half years of neonatal screening for haemoglobinopathies in Birmingham

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Abstract

A pilot neonatal screening programme for haemoglobinopathies linked with screening for phenylketonuria and congenital hypothyroidism was reviewed. During 1978 to December 1986 137 000 neonates were tested. There were improvements in the detection rate and accuracy of diagnosis for homozygotes and mixed heterozygotes, mainly associated with the introduction of citrate agarose gel electrophoresis as a follow up procedure on all specimens showing any abnormality on the initial cellulose acetate electrophoresis.

We recommend that the programme is continued on a service basis.

Introduction

Sickle cell disease is a serious health problem among the Afro-Caribbean population, with an incidence of about 1 in 300.¹ Data from the United States suggest that diagnosis in the neonatal period followed by counselling and usually preventive treatment with penicillin reduce early mortality and morbidity.²

In 1978 a pilot screening programme for the detection of sickle cell disease using cellulose acetate electrophoresis was set up in Birmingham, and preliminary results were reported in 1982.³ More recently neonatal screening programmes have been set up elsewhere in the United Kingdom in Brent⁴ and Camberwell,⁵ but no audit of such programmes has been published.

We reviewed the results from the Birmingham programme after eight and a half years of screening and evaluated the laboratory performance.

Methods

Screening for haemoglobinopathies was added to the existing neonatal screening programme for phenylketonuria in April 1978. Since then a test for congenital hypothyroidism was added. Capillary whole blood samples are collected from all newborns between 6 and 10 days of age and sent to the department of clinical chemistry at this hospital. All babies are tested as it is not possible to select out the "at risk" specimens. In our population 9.4% of

births are of Afro-Caribbean origin—that is, approximately 1500 births a year (1982 statistics).

In the laboratory the blood is centrifuged to separate the cells and plasma. The plasma is used to screen for phenylketonuria and congenital hypothyroidism. A cut off length of red cells is added to a tube containing deionised water. This is shaken and centrifuged to produce a haemolysate.

Before June 1984 the only test carried out on this haemolysate was cellulose acetate electrophoresis.³ Using this system HbS and HbD cannot be distinguished from each other, and similarly HbC and HbE cannot be separated. Since June 1984 all haemolysates showing an abnormality on cellulose acetate electrophoresis were tested further by citrate agarose gel electrophoresis at pH 6.0 followed by staining with tetramethylbenzidine.⁶ This method is more sensitive and also separates HbS from HbD and HbC from HbE, thereby allowing all bands to be fully identified by a combination of the two methods.

PATIENT FOLLOW UP

All babies who were thought to be homozygotes for HbS, HbD, HbC, or HbE, or mixed heterozygotes—that is, a combination of two abnormal haemoglobins, such as HbSC, or a combination of abnormal haemoglobin with β thalassaemia, such as HbS/ β thalassaemia—were followed up with hospital appointments to confirm the diagnosis and provide counselling about the disorder. Since April 1986 preventive treatment with penicillin has been offered.

To assess the reliability of the screening method to detect homozygotes and mixed heterozygotes missed cases were searched for by reviewing records at this hospital and also in haematology departments at other Birmingham hospitals. Screening records of all known cases of children presenting with these disorders and born since 1978 were then checked to see if the children had been tested as newborns and what results were obtained. This search was not comprehensive as some children may have been under the care of other hospitals in the city and others may have moved out of the city.

Although the programme was not set up to identify heterozygotes, an internal laboratory recording system recorded the presence of haemoglobins S, D, C, and E. We therefore decided to assess retrospectively the ability of the method to detect them. All children who were heterozygotes for HbS, HbD, HbC, and HbE, born since 1978, and known at this hospital were identified. The neonatal screening records were searched to check whether they had been tested as newborns and what result was obtained.

Results

A total of 137 002 neonates had been tested. Table I gives the number of babies recalled yearly as possible homozygotes or mixed heterozygotes and the outcome of these cases (SS or S β thalassaemia, SC, and other haemoglobinopathies: DD, D β thalassaemia, CC, C β thalassaemia, EE, and E β thalassaemia).

Thirty eight patients were recalled with a possible diagnosis of SS or S β thalassaemia or SC disease, of which 29 were confirmed, while in four follow up was not possible. There were five false positives, which all occurred before 1982, one of which seems to have been an error in recall. In the other four (of which three were shown to be heterozygotes) the initial

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TABLE 1—Outcome of patients recalled as possible HbSS or HbS β thalassaemia, HbSC, and other haemoglobinopathies (DD, D β thalassaemia, CC, C β thalassaemia, EE, and E β thalassaemia)

Year of birth	No recalled			No not followed up			No confirmed			No of false positives		
	HbSS or S β thalassaemia	HbSC	Other	HbSS or S β thalassaemia	HbSC	Other	HbSS or S β thalassaemia	HbSC	Other	HbSS or S β thalassaemia	HbSC	Other
1978	3	0	2	3 (2 left country; 1 cot death)	0	1 (refused appointment)	0	0	0	0	0	1 (A+D)
1979	2	2	1	0	0	1	1	1	0	1 (A+S)	1 (AA)	0
1980	0	2	0	0	0	0	0	2	0	0	0	0
1981	4	0	1	0	0	0	1	0	1 (DD)	3 (A+S; A+AA)	0	0
1982	1	0	1	0	0	0	1*	0	0	0	0	1 (AA)
1983	2	4	2	0	0	0	2	4	1 (DD)	0	0	1 (A+C)
1984	1	1	1	0	0	0	1	1	1 (CC)	0	0	0
1985	7	5	3	1 (refused appointment)	0	0	6	5	3 (DD; DD; D β thal)	0	0	0
1986	1	3	0	0	0	0	1	3	0	0	0	0
Total	21	17	11	4	0	2	13	16	6	4	1	3

*Combined HbS and hereditary persistence of fetal haemoglobin.

electrophoretic strip was incorrectly interpreted and citrate agarose gel electrophoresis was not available as an aid.

Detecting homozygotes and mixed heterozygotes—Table II gives the screening results for all known cases of homozygotes and mixed heterozygotes. Fifteen of the 20 missed cases were recorded as heterozygotes, and one was missed as a result of the recall error referred to above. Most of the missed cases might

TABLE II—Screening results of all known homozygotes and mixed heterozygotes

Year of birth	No	No not tested	No detected	No missed
1978	4	2	0	2
1979	6	0	2	4
1980	10	1	2	7
1981	4	2	2	0
1982	8	3	1	4
1983	10	0	7	3
1984	4	1	3	0
1985	15	1	14	0
1986	4	0	4	0
Total	65	10	35	20

have been correctly identified if citrate agarose electrophoresis had been available at the time. Before this was introduced 96 557 babies were tested and 14 cases were detected (one in 6897). All 20 cases that were missed, however, occurred in this period. Since the introduction of this additional test 40 445 babies have been tested and 21 cases detected (one in 1926), and no cases are known to have been missed.

Detecting heterozygotes—One hundred and thirty five heterozygotes for HbS, HbD, HbC, and HbE were identified from records at this hospital. Of these, 30 were born outside Birmingham and were therefore not screened. Of the remaining 105 who were screened, 90 had been correctly identified but 15 had been missed, all before 1984. The numbers since 1984 are still small, however. In the two full years for which citrate agarose electrophoresis has been available—1985 and 1986—139 and 134 probable heterozygotes for HbS were detected respectively.

Costs—The cost of providing the service for the five Birmingham districts above the cost of the existing neonatal screening service including labour, equipment, and consumables is 33 pence per baby tested.

Discussion

The detection rate of homozygotes and mixed heterozygotes and the accuracy of the diagnosis for these disorders have improved over the eight and a half year period. This improvement has coincided with the introduction of citrate agarose gel electrophoresis. We believe, however, that much of the improvement is also due to better interpretation in the light of experience. Moreover, the quality of electrophoresis has improved since 1985 when the present operator took over, whereas there had been frequent changes in the previous years.

It is possible that missed cases since 1984 may not yet have come to light. But the incidence of cases detected by screening has increased since citrate agarose gel electrophoresis was introduced and although the numbers are still small, they are close to that expected based on gene frequency in the population.

Although we did not intend to detect heterozygotes, our limited retrospective review of 135 known cases suggests that the programme can be extended to detect heterozygotes. No missed cases have been identified since 1984, and although the numbers in this period are small, this also suggests that the technique and interpretation have improved.

The finding of sickle cell trait in nearly a tenth of the Afro-Caribbean births for 1985 and 1986 is consistent with the findings of other workers^{1,5} and suggests that this method is sufficiently sensitive to detect most carriers.

When evaluating a screening procedure it is important to consider several criteria outlined by Whitby.⁷ These are as follows:

(1) Is the abnormality adequately defined? Clearly sickle cell disease satisfies this criterion.

(2) What population should be screened? Evidence has been presented by others⁵ that attempts to select out the babies at risk on the basis of ethnic origin leads to missed cases. We would therefore advocate screening the total neonatal population in areas where the incidence of sickle cell disease is high.

(3) What is the incidence of the condition? This has been shown to be about one in 300 in the Afro-Caribbean population and is much higher than phenylketonuria or congenital hypothyroidism, or even cystic fibrosis, in the white population.

(4) How do screening methods compare in efficiency and cost? We have shown that cellulose acetate electrophoresis alone is not sensitive enough for the reliable detection of sickle cell disease. The addition of citrate agarose gel electrophoresis has greatly improved the efficiency of the programme at little extra cost. The use of isoelectric focusing has been shown to be equally effective by others⁸ but is more expensive.

(5) Are diagnostic facilities available for the follow up of abnormalities shown by the screening procedure and is there an acceptable treatment for the condition? Patients detected by the screening programme are followed up in a haemoglobinopathy outpatient clinic where investigations, counselling, and family studies can be completed. We believe that counselling about potential problems has been well received and acted upon by the parents of our patients. Regular preventive treatment with penicillin has not, however, been accepted by them all.

(6) Is the natural history of the disease favourably influenced by the screening procedure and early treatment? The results of the cooperative study of sickle cell disease suggest that detection in the neonatal period followed by treatment with penicillin reduces morbidity and mortality.²

(7) Is it cost effective? The costs of adding screening for sickle cell disease onto an existing programme for other disorders are low. The

cost benefit of the programme could be quantified by a reduction in the need for hospital treatment, but there is little evidence to support this argument.

(8) What should be done about findings that are neither clearly normal nor obviously abnormal? Sick cell trait clearly falls into this category. We now need to look further at how efficient our programme is since the introduction of citrate agarose electrophoresis to confirm that sickle cell trait is accurately and reliably detected. If so, we would then need to consider how this information could best be used. Counselling and follow up for all these babies would be a major undertaking and there are implications for the provision of prenatal diagnosis.

We use the neonatal capillary blood sample already collected for phenylketonuria and congenital hypothyroid screening. Thus the advantages are pre-existing blood collection, processing, and laboratory facilities and a negligible failure rate, unlike cord blood sampling.¹ The additional cost is kept to a minimum. Moreover, a computer system linked to the local child health register ensures that every baby is tested, although we have to record some babies as "not tested" because they have received a blood transfusion which would interfere with the interpretation. We are evaluating in the West Midlands Region which health districts could justify neonatal screening in this form. Several health districts have so few babies at risk that this is clearly not justified. In districts where there are few heterozygotes for sickle cell disease, identification and testing of the

babies at risk only in each maternity unit will be more cost effective than comprehensive population screening.

Extending such a screening programme to other parts of the UK would depend upon the local incidence of sickle cell disease. In some regions an additional sample may be required as many screening laboratories use dried blood spot samples on filter paper cards for phenylketonuria and congenital hypothyroid screening. Although it is possible to elute haemoglobins from dried blood spots, the specimen is less stable and interpretation is more difficult.

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Medical History

Maurice Ravel's illness: a tragedy of lost creativity

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Abstract

Maurice Ravel had been subject to psychiatric disorder for many years when signs of organic brain disease appeared at the age of 52. Aphasia, apraxia, agraphia, and alexia became established some five years later. Musical creativity was lost. Alajouanine diagnosed cerebral atrophy with bilateral ventricular enlargement. Though Ravel's condition deteriorated progressively, generalised dementia was not apparent. He died in December 1937, after a craniotomy performed by Clovis Vincent, possibly from a subdural haematoma. Vincent's operative findings are described here. The likely cause of Ravel's illness was a restricted form of cerebral degeneration.

Observations on composers long dead are often of limited scientific value, for they depend on evidence from times when medical

practice was less precise than it is today and when laboratory tests were virtually unknown. Thus although Meyer and Slater were able to construct an acceptable explanation of Robert Schumann's mental illnesses,¹ Henson and Urich found medical reports prepared in the early 1840s inadequate when they tried to assign a cause to the paralysis of the composer's hand.² Systematic examination of the nervous system had yet to become established. With composers dying in more recent years some difficulties of fact may be overcome, but other problems remain, including lack of important information—a natural deficiency in retrospective studies—and an understandable reticence of family and friends. Moreover, observations by untrained witnesses may be unreliable or misleading.

What is the justification for turning intimate personal matters into public property? We can fairly claim that knowledge of the sufferings of composers heightens our appreciation of them and their music. This carries no implication that neurological dissection of creative artists illuminates the sources behind the power of their art.

Maurice Ravel (1872-1937) developed progressive neurological symptoms over the later years of his life. The history of his consequent decline has been told by many friends and biographers. His recorded disabilities are susceptible to neurological analysis, though methods of studying the brain by imaging and biochemical techniques were largely undeveloped in the 1930s, and neuropsychology was barely poised for advance. There are conflicting and even erroneous published accounts of Ravel's illness, but it is now

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