Neonatal screening

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Introduction

Though based firmly on the "old fashioned" disease of phenylketonuria, whole population blood based neonatal screening is a technique with many potential uses. Most never enter general service despite their technical feasibility, reflecting mainly the perceived balance between costs and benefits. For historical reasons there is an underlying assumption that neonatal screening should make a "profit".¹ Hitherto, such considerations have not been widely applied to other branches of medicine, but with the NHS reforms this is changing. A broader perspective on cost effectiveness, together with advances in treatment and the increased importance being attached to genetic information, may lead to greater enthusiasm for screening. New tests are still being developed and recent instrumental advances may make it practicable to screen for groups of disorders where this has previously been too difficult or too time consuming.

Screening in the United Kingdom

PHENYLKETONURIA

The foundations of the current neonatal screening programme in the United Kingdom were laid in 1969, when it was recommended that nappy testing with Phenestix should be abandoned in favour of blood based screening for phenylketonuria.² HM(69)72 instructed that laboratory examination of blood specimens should be centralised to the greatest possible extent, on a regional or even supra-regional basis.² Screening centres were to have full facilities for carrying out biochemical confirmatory tests and should be associated with the specialised service responsible for treatment. Relatively few regions complied fully with these suggestions and in 1979 there were 40 laboratories involved in screening for phenylketonuria, only 13 of which screened more than 25 000 babies a year.³ The introduction of an external quality assessment scheme in 1980 highlighted gross inadequacies and several laboratories with low workloads stopped screening altogether. Nevertheless, there were still 26 laboratories measuring phenylalanine for screening purposes in 1988.⁴

Various methods for detecting increased phenylalanine concentrations are used. The greatest number of babies are still screened for phenylketonuria by the Guthrie test (45% of infants).⁵ Next most popular are chromatographic methods (36%). These are more time consuming than the Guthrie test, their attraction being their ability to detect other amino acid disorders such as maple syrup urine disease. In most areas of the United Kingdom, however, the incidence of these conditions is very low and care is needed not to cause problems by reporting harmless conditions

such as histidinaemia. Automated fluorometry (19% of infants) has the advantage of sensitivity: (a) patients with tetrahydrobiopterin deficiencies are more likely to be detected⁴ and (b) most cases of galactosaemia may be picked out by the increased concentration of phenylalanine in their blood secondary to liver damage.⁶

All these methods essentially date from the early 1960s and instrument availability is becoming a problem for those using automated fluorometry. Adaptations of the fluorometric method to microtitre plates have recently been described, as have microplate assays using phenylalanine dehydrogenase, but so far none of these has entered routine use for large scale screening in the United Kingdom.

CONGENITAL HYPOTHYROIDISM

Screening for congenital hypothyroidism had been introduced in many parts of the United Kingdom well before its formal recommendation in June 1981.² The first large scale scheme was in East Anglia. This was based on a two stage screen which had been used in pioneering studies in North America. The first step was assay of thyroxine and samples giving results below the 20th centile which were then re-assayed for thyrotropin. This method is time consuming but has the advantage of detecting secondary hypothyroidism. All subsequent schemes in the United Kingdom followed later European practice and used immunoassay of thyrotropin as a single step screen.⁷ The sensitivity of screening assays is too low to permit detection of secondary hypothyroidism but it is argued that this is an acceptable limitation given the low incidence (2-3% of that of primary hypothyroidism) and its rather different clinical features.

QUALITY ASPECTS

The performance of the United Kingdom neonatal screening programme for phenylketonuria has recently been reviewed by Smith and colleagues.⁴ It has been largely effective, with a detected false negative rate over recent years of under 1%. Coverage in some places is far from complete, however, and there has been resistance to incurring additional costs just to ensure inclusion of the last 1% or so of babies. Some districts still do not have any effective checking system^{4,8} even though negative reporting is now recommended policy and the national test card/request form (HMR 101/6) was redesigned with flimsy copies in 1985 to facilitate this.

There are noticeable regional differences in the age at which babies with classic phenylketonuria have started treatment,⁴ and it is clear that an effective screening programme requires much more than an analytical ser-

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vice. Neonatal screening involves midwives, health visitors, family practitioners, community and hospital paediatricians, and a variety of clerical and administrative staff. The screening laboratory, whether district based or regional, forms the hub of the enterprise and effective communications with the various professional groups and an active role in maintaining the overall quality of the programme are essential.

OTHER DISORDERS

Few United Kingdom screening laboratories screen specifically for additional disorders, though laboratories using chromatographic methods for phenylketonuria will detect other amino acid disorders. Scotland screens for galactosaemia using a microbiological assay. One centre screens for homocystinuria.⁵ Pilot schemes for congenital adrenal hypoplasia and for biotinidase deficiency have been discontinued.

Screening for sickle cell disease, which provoked extreme controversy when mandated in parts of the United States of America in the early 1970s, is now recognised as medically useful because early diagnosis means prophylaxis against infection. Neonatal screening for sickle cell disease is performed in parts of London and Birmingham. Further expansion seems likely but in much of the United Kingdom the incidence in the population as a whole is considered too low to merit universal screening and the practical and political difficulties associated with selective screening too daunting. Preconceptual screening for heterozygotes could be a more effective measure⁹ but the ethical and organisational aspects have yet to be fully addressed.

It is possible to detect most cases of cystic fibrosis on the basis of increased immunoreactive trypsin in the neonatal blood spot.¹⁰ There is disagreement over the clinical value of such a screen. Significant financial savings have been reported but this is likely to be highly dependent on the local pattern of medical care. Four laboratories in the United Kingdom screen for cystic fibrosis, three reporting highly satisfactory results in terms of sensitivity and specificity. All use a two stage screen; babies showing increased immunoreactive trypsin in the initial screening sample are retested at about 4 weeks of age. In all cases the diagnosis must be confirmed by a sweat test. The need for retesting may be substantially reduced by DNA analysis for the commonly occurring F508 mutation which can be performed on the initial blood spot using the polymerase chain reaction. In the South Australia protocol¹¹ the second immunoreactive trypsin test is eliminated entirely, but at the cost of a greatly increased number of negative sweat tests. A more conservative protocol requires the second immunoreactive trypsin assay to be performed only on F508 heterozygotes. In both these schemes the small proportion of patients who have only non-F508 cystic fibrosis mutations will be missed.

International perspectives

Neonatal screening programmes exist in many countries. Throughout most of Europe,

North America, Australia, and in parts of the Far East there is equal emphasis on phenylketonuria and congenital hypothyroidism. In the developing world screening for congenital hypothyroidism is seen as a practical and cost effective measure, but lack of treatment facilities and relatively low incidence makes screening for phenylketonuria unattractive. Considerable dedication and ingenuity has been devoted to some such schemes. Cuba, for example, has enzyme linked immunosorbent assays (ELISA) based screens for thyrotropin, α -fetoprotein (in pregnancy), and IgE, with reagents and instrumentation being manufactured indigenously.

Viewed on a global scale, there is a bewildering variety of different screens in use. Some, such as those for glucose-6-phosphate dehydrogenase deficiency and tyrosinaemia type I, reflect disorders particularly common on a local basis. Others have arisen because of the interests of an individual scientist. More systematic differences do exist, however, implying differing judgments of the medical value of the tests concerned. Thus in the United States of America 44 out of 51 states screen all newborns for galactosaemia, and 23 and 21 screen for maple syrup urine disease and homocystinuria, respectively. These last two disorders have incidences of 1 in >200 000 in many of the populations concerned, and even as "add-ons" must be considered at the very limits of viability. Nevertheless, international comparisons leave the impression that in the United Kingdom there is a particularly austere approach to the assessment of screening and a general reluctance to support innovation.

Outlook

Despite the apparently slow progress in neonatal screening, with only two tests introduced on a national basis in the United Kingdom over a period of 25 years, there is great potential for further development.

NEW DISEASES

Most of the more recently discovered inherited metabolic disorders do not lend themselves to neonatal screening, either because of their rarity, or lack of adequate laboratory methods, or effective treatment. An exception is biotinidase deficiency which, though relatively uncommon with a global incidence averaging 1 in 60 000, is readily detectable in dried blood spots and can be easily and cheaply treated.¹² It is screened for routinely in 15 states of the United States of America and in several European countries.

Medium chain acyl-CoA dehydrogenase deficiency seems to be about as common as phenylketonuria.¹³ It is a life threatening condition but its natural history is not well understood and screening would have to be based on DNA analysis or tandem mass spectrometry (see below), either of which would be costly.

NEW TREATMENTS

Screening for sickle cell disease has been revived by the general acceptance that early vigorous treatment of the disorder is beneficial. Screening for cystic fibrosis is also

increasingly seen as a logical extension of the general move to more intensive treatment and will become essential once more fundamental treatment, perhaps based on gene replacement, has been developed.

Screening for Duchenne's muscular dystrophy, based on creatine kinase in dried blood spots, has been possible for many years. A "pilot" study in Lyon, France, has tested 320 000 babies since 1975, and a commercial service, paid for by the parents themselves, has been available in Germany since 1977 and has screened over 350 000 babies. Despite the obvious genetic implications, this screen has not proved popular, due largely to the lack of any useful early treatment. As with cystic fibrosis, a clearer understanding of the biological nature of the condition has raised hopes of more effective approaches and should these be realised there will be a pressing need for routine screening of all male babies.

NEW LABORATORY TECHNIQUES

Immunoassay techniques continue to develop apace in both sensitivity and specificity. Perhaps the most important development from the neonatal screening point of view is the adaptation of dissociation enhanced lanthanide fluoroimmunoassay (DELFLIA) to assay up to four different labels simultaneously. A demonstration system allowed simultaneous screening for congenital hypothyroidism, congenital adrenal hyperplasia, cystic fibrosis, and muscular dystrophy (using thyrotropin, 17- α -hydroxyprogesterone, immunoreactive trypsin and creatine kinase MM, respectively) in a single 5 mm diameter dried blood spot.¹⁴ Given that the main objection to screening for congenital adrenal hyperplasia is the cost, to which labour makes a major contribution, such technical developments and the high level of automation that accompanies them may well permit the introduction of this and other screening tests hitherto considered too marginal to warrant consideration.

A very different development, that of fast atom bombardment tandem mass spectrometry with continuous flow injection, may permit broad spectrum screening (screening screening) for a wide range of organic acid and amino acid disorders.¹⁵ Though many of the disorders covered by this screen are relatively rare, the inclusion of phenylketonuria and virtually the whole range of organic acid defects, including medium chain acyl-CoA dehydrogenase deficiency, may make this a viable proposition despite the high capital cost.

SCREENING FOR INFECTION

The worldwide HIV epidemic has given a major impetus to the use of neonatal screening spots for epidemiological studies. Maternal antibodies to HIV may be detected in neonatal blood spots by a variety of methods and this is all that is required to monitor trends in prevalence. At present most programmes, including those in the United Kingdom, are for unlinked anonymous testing, but as treatment is improved the pressure for named testing could become stronger.

SCREENING FOR GENETIC INFORMATION

So far neonatal screening to provide genetic information has been used primarily to estimate prevalence, either of the condition in question or of a specific mutant gene (heterozygote frequency). The large scale provision of specific genetic information—for example, identifying individuals carrying the F508 cystic fibrosis mutation—raises many practical and ethical problems. It is technically feasible to obtain such information as part of the routine neonatal screening programme, and this would have operational and financial advantages over schemes targeted at older age groups. However, there are major reservations about whether reporting such information in the neonatal period would be appropriate or useful.

POST-NEONATAL SCREENING

For some diseases screening in the neonatal period is not effective and a growing number of schemes use neonatal screening technology to examine older children. Screens for Wilson's disease (at 18 months), dyslipidaemias (neonatal or 18 months), neuroblastoma (3 weeks to 6 months), renal disease (6 months) and preclinical insulin dependent juvenile diabetes (5–7 years) were presented at a recent symposium.¹⁶ Perhaps it is not too fanciful to envisage the time when the neonatal screen is just the first of a series performed during childhood and forming a major tool in preventive medicine.

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