

Newborn screening for cystic fibrosis

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Since the late 1970s when the potential of the immunoreactive trypsinogen assay for early identification of infants with cystic fibrosis was first recognised, the performance of newborn blood spot screening (NBS) has been continually assessed and its use has gradually expanded. NBS for cystic fibrosis is a cost-effective strategy and, if standards of care are fully implemented and robust management pathways are in place, has a positive effect on clinical outcomes. In the past decade, NBS has undergone rapid expansion and an unprecedented number of infants with cystic fibrosis have access to early diagnosis and care. Cystic fibrosis NBS has now moved on from the development phase and is entering an era of consolidation. In the future, research should focus on the rationalisation and optimisation of existing programmes, with particular attention to bioethical implications such as unwanted detection of carriers and inconclusive diagnoses.

Introduction

A proactive multidisciplinary approach has long been promoted for the care of people with cystic fibrosis.^{1,2} In the past three decades, increasing evidence that well-nourished and active patients enjoy a better quality of life, respiratory function, and survival has emerged to support this strategy.^{3,4} Early researchers in cystic fibrosis believed newborn blood spot screening (NBS) was a valid strategy to help achieve these goals. The characterisation of the immunoreactive trypsinogen (IRT) assay in 1979 provided laboratories with a potential screening test to identify infants with cystic fibrosis in the first weeks of life, often before they presented with clinical signs.⁵ The IRT assay seemed to have better sensitivity than other screening tests that had previously been used to identify infants with cystic fibrosis (eg, measuring meconium albumin or lactase levels)^{6,7} and was included in the NBS schedule already established for other diseases such as phenylketonuria. However, concerns were raised that screening of infants for cystic fibrosis might not have sufficient effect on clinical outcomes to meet the criteria required to advocate universal screening. This concern prompted a call for evidence to show that NBS was beneficial for children with cystic fibrosis.

Previous research

Clinical trials and epidemiological studies

Two pseudo-randomised clinical trials were initiated in the 1980s. A trial in the UK screened newborn babies for cystic fibrosis on alternate weeks.⁸ After 4 years, no substantial differences were reported in the nutritional or respiratory outcomes of 58 infants with cystic fibrosis identified by screening compared with 44 children diagnosed clinically.⁸ Simultaneously, in a trial in Wisconsin, a substantial nutritional benefit was noted in the cohort of 56 infants who were diagnosed early through screening compared with the 40 infants in the cohort diagnosed clinically.⁹ Furthermore, evidence of increased airway damage on chest radiographs was reported in the screened cohort, possibly as a result of earlier *Pseudomonas* infection.¹⁰ Issues with the clinical management of these infants were described for both

clinical trials, notably infrequent visits to a cystic fibrosis centre and suboptimum segregation policies that might have had an adverse effect on clinical outcomes.

Despite the scarcity of definitive evidence of improved outcome from these clinical trials, NBS for cystic fibrosis has expanded worldwide, particularly in the past decade.¹¹ This expansion was based on evidence from epidemiological sources, including patient registries, which provided information on health-care costs and longer term outcomes.^{12,13}

Long-term results from cohort studies

A major difficulty in evaluation of the clinical effects of cystic fibrosis NBS is that positive outcomes can take many years to become apparent. Compelling evidence has come from the long-term follow-up of children enrolled in the Sydney cohort study.¹⁴ Data available for the progress of these patients in their third decade of life

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Key messages

- Newborn blood spot screening (NBS) for cystic fibrosis has a positive effect on short-term and long-term clinical outcomes.
- Screening is a cost-effective public health strategy.
- Substantial worldwide expansion of screening for cystic fibrosis has been reported, with countries adopting different approaches depending on their target populations.
- All cystic fibrosis NBS protocols measure immunoreactive trypsinogen in the first week of life as the initial step; however, a wide range of approaches are subsequently used to improve the positive predictive value.
- Protocols that use DNA analysis for cystic fibrosis-causing mutations also detect carriers, which is considered undesirable for a NBS programme.
- DNA analysis will improve the timeliness and positive predictive value of NBS, but will increase the number of inconclusive diagnoses. For many this finding will not have any phenotypic significance.
- Population carrier screening (antenatal) might result in a decrease in the cystic fibrosis birth prevalence and affect the performance of a NBS programme.
- New emerging therapies for cystic fibrosis that correct the underlying genetic defect highlight the importance of early diagnosis through NBS.
- Cystic fibrosis NBS is now entering a new age—one of rationalisation and optimisation. A bioethical model that incorporates cost-effectiveness should be used to establish best practice.

showed that those diagnosed after the introduction of NBS have improved survival compared with those diagnosed clinically. A historical cohort study is not an ideal design to compare two public health strategies; however, the length of this study mitigates these deficiencies and the results provide good evidence that earlier diagnosis from NBS might have a notable effect on wellbeing and survival in adult life.

Cost-effectiveness studies

Beyond the existing NBS infrastructure (distribution and timely collection of screening cards), cystic fibrosis newborn screening needs additional expenditure for analysis (eg, IRT and DNA), maintenance of laboratory standards, supervision of reports, implementation of systems to ensure appropriate handling of positive results, and costs associated with the assessment and processing of false-positive results.

Whereas in the early years of cystic fibrosis NBS, decisions to screen were mainly based on clinical and social factors, subsequently more formal cost-effectiveness analysis of newborn screening for cystic fibrosis has become a key element of the assessment of this public health intervention.

A decision-tree analysis suggested that NBS was an expensive method of diagnosis, but acknowledged that it could be cost effective if it delayed the onset of symptoms.¹⁵ This suggestion is supported by results from studies showing that the cost of care of patients with cystic fibrosis was substantially lower if the disease was detected early through screening.¹⁶ In a UK Registry study that compared 184 infants diagnosed by NBS with 950 infants diagnosed clinically, substantial savings were reported in drug utilisation costs that offset the additional costs of adding cystic fibrosis screening to existing NBS

programmes.¹⁷ A Dutch study comparing different screening strategies based on a theoretical birth cohort of 200 000 infants reported savings in all models of NBS.¹⁸

The IRT assay

The IRT assay provides the framework on which all NBS for cystic fibrosis is currently based.¹⁹ Elevated IRT is thought to be related to the pancreatic damage often present in infants with cystic fibrosis, but it can also identify infants with a milder, pancreatic sufficient phenotype and some carriers.²⁰ Automated immunoassays enable large numbers of samples to be processed as part of a NBS programme.²¹ A better quality of dried blood spot sample is needed for the IRT assay compared with other analytes used for NBS. Thus, inclusion of cystic fibrosis into the panel of NBS disorders necessitates additional support and training for health-care workers who obtain the blood spot sample and, invariably, an increased number of repeat sample requests. However, in a well-organised screening laboratory and public health service, incorporation of cystic fibrosis into the NBS programme should be feasible.²¹

Other technical issues make interpretation of IRT assay results challenging, most notably variability between batches of reagents used for the assay that affect the IRT values and an apparent reduction in overall IRT values reported during hot seasons, possibly resulting from some degradation of the protein as the sample is transferred to the laboratory.²⁰ Some screening laboratories have advocated the continuous monitoring of IRT values and altering the cutoff accordingly to achieve a predetermined percentage of infants referred (a floating cutoff).²⁰

Strategies to reduce false-negative and false-positive rates

Similar to most public health interventions, a flawless cystic fibrosis NBS programme is unrealistic if not unachievable. Accurate strategic choices might optimise the effectiveness of cystic fibrosis NBS, however, some false negatives and false positives and infants in whom cystic fibrosis cannot be confirmed or excluded are inherent in screening programmes. Tolerance of a few missed cases might be instrumental in limiting unwanted effects of NBS, such as inconclusive cases. Table 1 shows how sensitivity, specificity, and positive predictive value (PPV; the ratio between true positives and all positive test results) of cystic fibrosis NBS strategies are defined.

Improving specificity

Measurement of IRT during the first week of life (IRT-1) provides a sensitive test to identify infants with cystic fibrosis; however, the test is not specific. To limit the number of false positives and achieve an acceptable combination of sensitivity and specificity, a range of different second-tier and sometimes third-tier tests are used in infants with a raised IRT-1 (table 2).

	Affected	Not affected
NBS positive	A: true positive	B: false positive
NBS negative	C: false negative	D: true negative

Sensitivity=A / (A + C). Specificity=D / (B + D). Positive predictive value=A / (A + B).
NBS=newborn blood spot screening.

Table 1: Definitions of sensitivity, specificity, and positive predictive value of NBS strategies for cystic fibrosis

	Strategy 1	Strategy 2	Strategy 3	Strategy 4	Strategy 5	Strategy 6	Strategy 7
1 st tier	IRT1	IRT1	IRT1	IRT1	IRT1	IRT1	IRT1
2 nd tier	IRT2	DNA	DNA	IRT2	DNA	PAP	PAP
3 rd tier	EGA	DNA	IRT2	DNA	DNA
4 th tier	EGA

IRT1=immunoreactive trypsinogen assay at birth. IRT2=immunoreactive trypsinogen assay at day 21. DNA=panel of cystic fibrosis transmembrane conductance regulator (CFTR) mutations. EGA=extended genetic analysis by sequencing. PAP=pancreatitis associated protein. NBS=newborn blood spot screening. All strategies select NBS-positive infants. Confirmation or exclusion of cystic fibrosis diagnosis is obtained by sweat test.

Table 2: Most frequently used NBS strategies for cystic fibrosis

The second-tier test used in early NBS programmes was a repeat IRT measurement from a second sample taken at day 10–21 of life (IRT-2). IRT values decrease in infants without cystic fibrosis over the first 4 weeks of life, but remain high in those with cystic fibrosis.²² The IRT/IRT protocol improves PPV by reducing the number of infants that are referred for a sweat test. This strategy depends on health resources being available to reliably obtain a second sample for IRT measurement.

Identification of the cystic fibrosis transmembrane conductance regulator (*CFTR*) gene provided an alternative second-tier strategy. The same dried blood spot sample obtained for IRT-1 measurement could be analysed for *CFTR* mutations. Initially, analysis was only for phe508del (c.1521_1523delCTT), the most common mutation associated with cystic fibrosis; however, larger panels incorporating more mutations have been used as the technology has advanced.^{20,23,24} An IRT/DNA protocol has the advantage of removing the need for a second sample, improving the timeliness of the diagnosis. For infants carrying two mutations included in the panel, the use of an IRT/DNA protocol can reduce time to diagnosis substantially, in some cases to less than 14 days of age.²⁵

Improving sensitivity

Although the IRT assay is a valid instrument for NBS, even the most effective programmes will miss cases. By far the most common cause of an affected but undetected case is that the IRT-1 value falls below the cutoff. Reducing the IRT-1 cutoff is the most straightforward way of improving sensitivity.²¹ However, even a small reduction in the IRT-1 cutoff will have a substantial effect on PPV, with an increase in the number of infants requiring referral for diagnostic assessment and a sweat test.

Furthermore, sensitivity might be affected by the large number of *CFTR* mutations that exist (often at very low population frequencies), with the possibility of a missed mutation being a feature of IRT/DNA protocols. To address this issue, genetic analysis included in the NBS protocol can be adapted in two ways: first, DNA panels could be used to identify larger numbers of mutations; and second, large areas of the *CFTR* gene could be sequenced. Using a DNA panel that resembles the DNA of the population being screened is good practice but, irrespective of size, the panel will always miss rare mutations. Sequencing the *CFTR* gene, which is faster and less expensive owing to the introduction of next-generation sequencing technologies,²⁶ could help eliminate this issue. However, sequencing can lead to increased diagnostic uncertainty with the identification of mutations with unclear phenotypic outcomes.

A widely applied strategy to avoid missed cases is to further test very high IRT-1 results, irrespective of the result of the DNA testing. This strategy is often referred to as safety net, failsafe, or ultra-high IRT. Several different protocols exist, including referral straight to

sweat testing or measurement of a second IRT at day 21 of age.^{21,24,27–29} The advantage of incorporating such a strategy is that it compensates for discrimination against ethnic populations with rarer *CFTR* mutations not included on a typical screening panel. One concern is that the strategy generally involves a substantial number of diagnostic assessments for recognition of a small number of infants with cystic fibrosis. This situation is exacerbated by raised IRT levels in infants born prematurely, neonates that are unwell, and neonates with a chromosomal abnormality.^{30,31} The use of a safety net strategy is debated and programmes should carefully monitor its performance.

When mutation frequencies in the screened population are unknown or the use of genetic analysis is restricted, measuring pancreatitis associated protein (PAP) in addition to IRT^{32,33} might be an option. IRT/PAP can achieve acceptable sensitivity, but the PPV is reduced compared with IRT/DNA.^{34,35} This reduction in PPV could be nullified by a multi-tier protocol that uses a combination of IRT, PAP, and selected plus expanded genetic analysis. In the CHOPIN study,³² this combination resulted in improved PPV compared with a strategy in which only IRT/PAP was used.

Irrespective of the protocol used, a positive NBS test for cystic fibrosis needs careful clinical evaluation including a sweat test to provide a definitive diagnosis or, occasionally, to refute it. Although outside the scope of this Review, the importance of standardisation and quality monitoring of sweat chloride testing cannot be overemphasised. Maintaining a high-quality sweat test service is a key factor affecting the effectiveness of NBS for cystic fibrosis.

Byproducts of cystic fibrosis NBS

Carriers

Genetic analysis is considered inappropriate in children, unless it has health advantages. This is the case for cystic fibrosis NBS, in which the inclusion of mutation analysis usually improves the performance of the protocol. Inevitably, some infants who have a normal sweat test will have one *CFTR* mutation detected. These children are carriers, a widely recognised incidental finding for programmes that include mutation analysis. Reports suggest that the rate of carrier recognition is higher than expected in view of the frequency of *CFTR* mutations in the population screened. For example, in New South Wales, Australia,³⁶ the rate of carriers was 1.8 times the expected frequency. Similar values were reported in Victoria (Australia), Wisconsin (USA), and Italy.^{36–39} The cause of the high carrier rate has never been clearly explained, but is probably related to the effect of the *CFTR* mutation on pancreatic electrolyte secretion. This alteration might affect trypsinogen release and skew the distribution of IRT in carriers above the cutoff used for the whole population.⁴⁰ Evidence that the *CFTR* mutation affects pancreatic

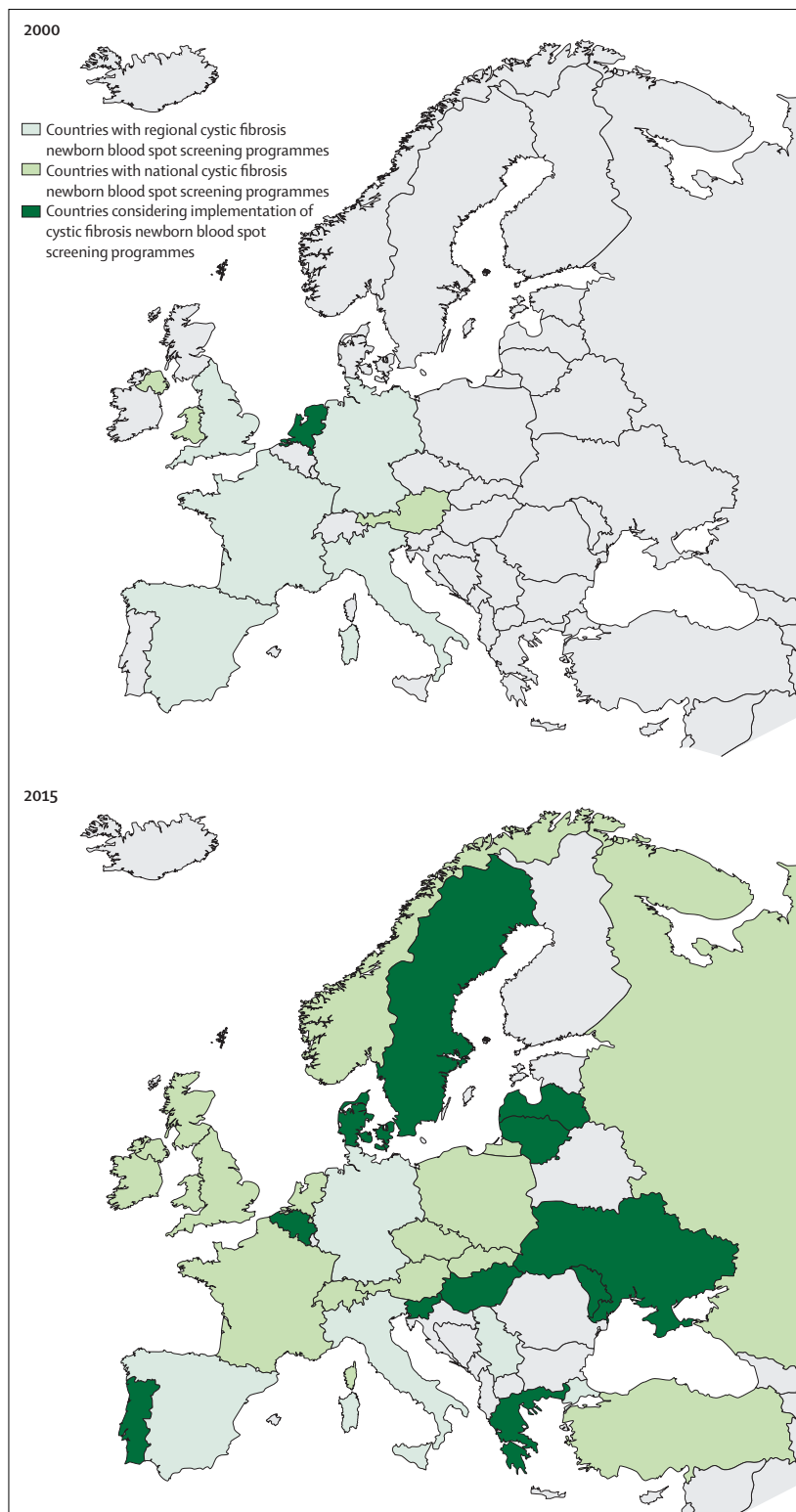


Figure 1: Expansion of cystic fibrosis NBS programmes in Europe

function is indirectly supported by the increased frequency of carriers in individuals with idiopathic chronic pancreatitis.⁴¹

Carrier detection is generally considered to be an unwanted byproduct of screening. Thus, antenatal information to parents should clearly state that NBS will not only detect infants with cystic fibrosis, but also some carriers. Most centres advocate disclosure of carriers to help with genetic counselling and provide couples with information on the potential risk of cystic fibrosis for future pregnancies, and likewise the risk for pregnancies of the index cases when they are older.⁴² Furthermore, genetic counselling can result in cascade family testing that, in turn, gives relatives who are at increased risk of being carriers a choice of whether to be tested or not.⁴³

Discovering their carrier status before their legal majority does not seem to negatively affect children. Furthermore, no substantial issues with the perception of vulnerability of carriers have been identified. However, one study identified that up to 18% of parents of carriers detected by NBS had some level of anxiety about the health of their child up to 4 years after screening.^{44,45} Solutions to the dilemma of unwanted carrier detection include using alternative screening strategies that avoid or minimise *CFTR* mutation analysis (table 2), but this usually results in reduced sensitivity and specificity.

Inconclusive diagnosis

Although infants with cystic fibrosis benefit from early diagnosis by NBS, the identification of newborn babies who have a positive screening result but an inconclusive diagnosis is a drawback of NBS. For most of these infants, the positive result has no long-term clinical significance. These infants might have ambiguous sweat chloride levels or normal sweat test values and genotypes including mutations with unclear clinical liability. With the development of the CFTR2 project,⁴⁶ an increasing number of such mutations might be reclassified as cystic fibrosis causing or non-cystic fibrosis causing, but arguably there will always be mutations that do not have a clear phenotypic characterisation.

The diagnosis of some carriers detected by NBS might be inconclusive when subsequent genetic testing identifies a second mutation with unknown or variable phenotypic outcomes.^{47,48}

New diagnostic designations for infants with an inconclusive diagnosis after NBS

Two independent recommendations were proposed for the classification and management of infants with an unclear diagnosis after cystic fibrosis NBS. These two exercises were undertaken in parallel and advocate a similar management approach for these infants; however, two different designation terms were suggested: CF transmembrane conductance regulator-related metabolic syndrome (CRMS) in the USA and cystic fibrosis screen-positive, inconclusive diagnosis (CFSPID)

in Europe. CRMS/CFSPID infants are defined by high IRT at birth plus an equivocal sweat chloride concentration and fewer than two mutations of the *CFTR* gene that are known to be associated with cystic fibrosis. Some infants might have two *CFTR* mutations, one of which might be of variable clinical significance, with a sweat chloride concentration <30 mmol/L.^{46,49,50}

Varying frequency of CRMS/CFSPID and screening algorithms

All NBS programmes for cystic fibrosis can result in the identification of infants with CRMS/CFSPID, including IRT/IRT, but the frequency has increased as protocols incorporate more detailed DNA analysis and sequencing. Some programmes with an IRT/DNA algorithm have reported the identification of one newborn baby with unclear diagnosis for every two with cystic fibrosis.^{51,52} The first algorithm to use extended gene sequencing was an attempt to improve PPV in the ethnically diverse population of California.⁵³ Samples from infants with one mutation on a limited DNA panel were sent for extended gene sequencing. This algorithm resulted in the identification of two mutations known to be disease causing on the first mutation panel in 52% (138 of 266) of all cystic fibrosis infants diagnosed, and an additional 41% (110 of 266) after the sequencing step. The algorithm identified CRMS in 279 infants, which is more than those diagnosed with cystic fibrosis by NBS.⁵³

Clinical outcome of newborn babies with an inconclusive diagnosis

The outcome for infants with CRMS/CFSPID is not well characterised, particularly for those with no apparent clinical sequelae. Long-term figures on CRMS/CFSPID infants might be biased towards symptomatic children and prone to loss to follow up, since families with a well infant can disengage from clinical contact. Data available

at present suggest that children with CRMS/CFSPID might have higher than expected frequencies of respiratory pathogens associated with cystic fibrosis, such as *Pseudomonas aeruginosa* or *Staphylococcus aureus*.^{40,52,54} It remains unclear if this is the result of more frequent culturing, or an indication of the underlying prevalence of these bacteria in the non-cystic fibrosis population, particularly in the case of *Staphylococcus aureus* infection.

The number of CRMS/CFSPID infants that are subsequently diagnosed with cystic fibrosis is unclear; currently available evidence suggests that up to 3 years of age, roughly 1 in 10 infants is diagnosed with cystic fibrosis.^{51,54} A substantial number of the mutations recognised in CRMS/CFSPID infants are well characterised in adults, often in the context of late diagnosis or milder disease presentations. Some adults have a condition called CFTR-related disorder, which is characterised by single organ disease (generally congenital bilateral absence of the vas deferens) and a normal sweat test.⁵⁵ Infants with CRMS/CFSPID might have an increased risk of a CFTR-related disorder in adult life. Comprehensive genotyping and long-term data collection are important, particularly in cystic fibrosis registries as a subgroup undergoing separate analyses. Although predicting individual correlations between genotype and phenotype evolution might prove unrealistic, long-term reviews will improve our understanding of the effect of inconclusive NBS diagnoses on public health.

Clinical guidelines for the care of CRMS/CFSPID infants recommend regular although infrequent clinic visits and repeating the sweat test at 6 months and 2 years of age.⁵⁰ Families should be aware of potential health problems and the importance of maintaining a healthy lifestyle, even if they are discharged from the cystic fibrosis clinic.⁵⁶

Research needed

How many, and which, infants with inconclusive diagnoses detected through cystic fibrosis NBS will eventually develop clinically relevant manifestations related to CFTR?	Long-term follow-up studies in CRMS/CFSPID children
Is the integration of the PAP assay into cystic fibrosis NBS programmes compatible with a reduction of some unwanted effects (eg, carrier detection and inconclusive diagnoses) while maintaining acceptable performance of the programme?	Comparative longitudinal studies
Can the sensitivity (inclusion of cystic fibrosis-causing mutations only) and specificity (exclusion of non-cystic fibrosis-causing mutations) of genetic analysis used in cystic fibrosis NBS be improved?	Expansion of functional and clinical studies assessing disease liability of <i>CFTR</i> mutations, notably the CFTR2 project
Can the performance of cystic fibrosis NBS be improved in subpopulations with different ethnic backgrounds and mutation distribution?	Further assessment of the effectiveness of programmes not using genetic analysis (PAP), a wide range of studies on <i>CFTR</i> allelic heterogeneity, and identification of more cystic fibrosis-causing mutations (CFTR2 project)
How are the positive benefits of NBS for cystic fibrosis (which can take decades to materialise) balanced against the negative effect of poor positive predictive value and the recognition of carriers and CFSPID/CRMS	Development of a bioethical model that balances positive outcome and cost-effectiveness with the effect of carrier and CFSPID/CRMS recognition on families
Can cystic fibrosis NBS be offered to newborn babies and families in countries where this health policy has not yet been implemented?	Audit of barriers to implementation and development of strategies to overcome them

NBS=newborn blood spot screening. CFTR=cystic fibrosis transmembrane conductance regulator. CRMS=conductance regulator-related metabolic syndrome. CFSPID=cystic fibrosis screen-positive, inconclusive diagnosis. PAP=pancreatitis associated protein.

Table 3: Unanswered questions and future research directions

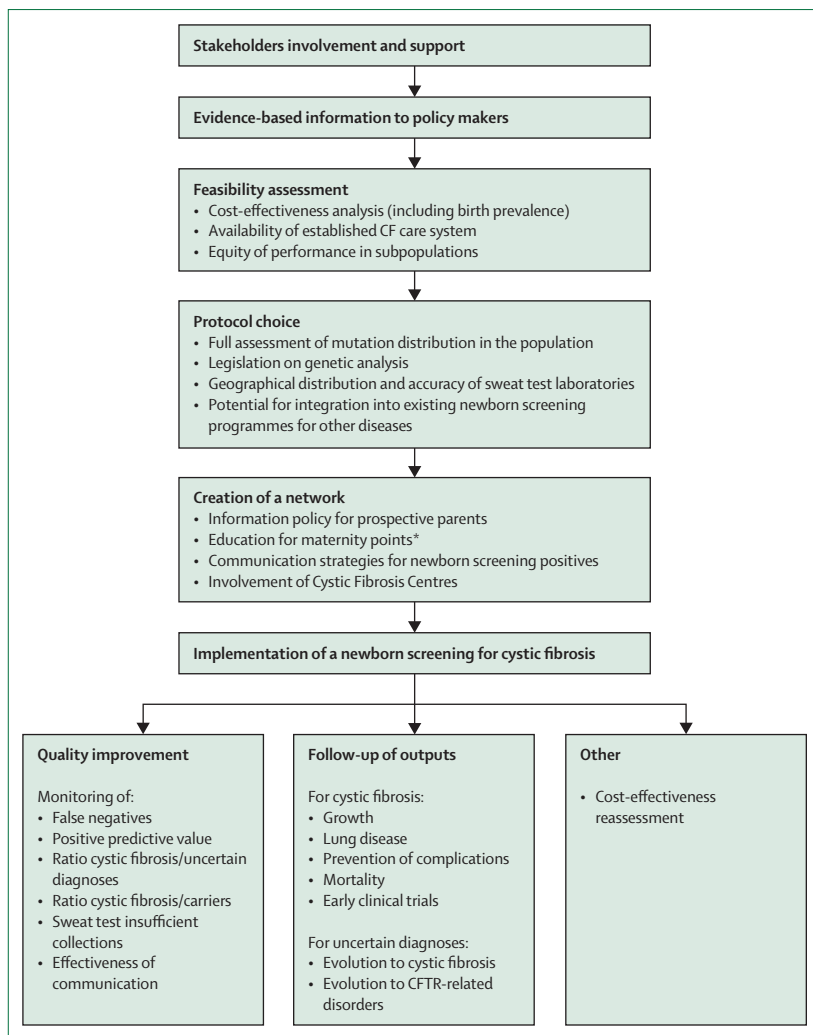


Figure 2: Analyses and actions before and after implementation of cystic fibrosis newborn blood spot screening programmes

CFTR=cystic fibrosis transmembrane conductance regulator. CF=cystic fibrosis. *Staff in peripheral maternity points to be made aware of the screening procedure and able to provide correct information to parents.

Population carrier screening and NBS

Early diagnosis might be possible through antenatal cystic fibrosis carrier screening. Couples are offered testing for common *CFTR* mutations and those identified to be carriers might choose prenatal diagnosis. If parents opt not to terminate an affected fetus, the opportunity for very early care is available. Although formal cystic fibrosis carrier screening programmes are less widely available than NBS, many couples of reproductive age are offered the carrier test in the USA and in some areas of Europe and Australia.^{57–59}

Population carrier screening has been shown to affect the performance of NBS programmes in regions who offer it. After several years, carrier screening might lead to a decrease in cystic fibrosis birth prevalence, a trend inversely correlated with the number of carrier tests performed and of carrier couples detected.⁶⁰ The high

costs, organisational burden, and byproducts associated with NBS will be scrutinised further should the number of infants born with cystic fibrosis decrease. Indeed, the European Cystic Fibrosis Society suggests a careful evaluation of the validity of NBS when the cystic fibrosis incidence is less than 1 in 7000.^{21,61} Moreover, the incidence decline connected with carrier screening might generate an unfavourable ratio between cystic fibrosis cases and unwanted effects, such as false positives and inconclusive diagnoses.⁶²

NBS in the context of *CFTR* modulator therapies

The development of therapeutic options that target the underlying defect in cystic fibrosis has put the potential for early disease control into sharp perspective. These therapies, which target the basic pathophysiology of cystic fibrosis, could substantially reduce morbidity and mortality in individuals with specific mutations. The use of ivacaftor in patients carrying class III mutations has become the model of a personalised medicine approach to cystic fibrosis care, and more compounds addressing other defects of protein synthesis or function are being investigated.⁶³

Since ivacaftor is only licensed for use in infants over 2 years of age, evidence examining the potential of ivacaftor in newborn infants with susceptible *CFTR* mutations is non-existent. Exploring the efficacy of modulators in very young children for whom a full clinical picture of cystic fibrosis is not yet complete might prove technically difficult. Furthermore, selecting disease markers and endpoints will be challenging. If studies can prove that ivacaftor and other modulators that might be available in the future are beneficial and safe from birth, their impact on lung disease and survival rates might be strengthened. Moreover, preliminary reports of discontinuation of pancreatic enzyme therapy and partial recovery of exocrine pancreatic function in some patients given ivacaftor raises the prospect of therapies that might halt or postpone the loss of residual pancreatic function, which is often seen in the first months of life.⁶⁴ The early and possibly presymptomatic use of these drugs would epitomise the notion of pre-emptive treatment, of which NBS is the archetypal example, and emphasises the significance of early diagnosis.

The future: consolidation of a complex system

Cystic fibrosis NBS has been through three phases: discovery, assessment, and expansion. After a slow start, cystic fibrosis NBS has expanded rapidly over the past decade. Data from a European Cystic Fibrosis Society Neonatal Screening Working Group survey showed that over 5 million European newborn babies and 4 million US newborn babies were screened for CF in 2012.¹¹ Furthermore, it has been estimated that worldwide over 12 million newborn infants were screened in 2014.¹¹ The use of cystic fibrosis NBS increased rapidly inside Europe from 2000 to 2015 (figure 1). This expansion has made

early diagnosis and care possible for an unprecedented number of infants with cystic fibrosis.

Rationalisation and optimisation of cystic fibrosis NBS are now the focus of this public health strategy (table 3) as it enters a fourth phase, one of consolidation. In the future, programmes will need to focus both on further expansion in areas where cystic fibrosis NBS has not yet been implemented and monitoring, and if necessary enhancement, of existing structures. Strong NBS structures will be in the best position to appraise and possibly integrate upcoming opportunities, such as next generation sequencing, PAP, and new therapies.

Introduction in non-screened populations

The European Cystic Fibrosis Society Neonatal Screening Working Group estimates that more than 2 million neonates born in Europe each year are not screened for cystic fibrosis.¹¹ Many more children are born in countries where cystic fibrosis might be underdiagnosed and would benefit from a NBS programme. Providing all infants with the same opportunities for cystic fibrosis NBS is good medical practice and appeals to an ethical principle of justice.

Several aspects of organisation and management need to be carefully considered when planning the introduction of cystic fibrosis NBS in a non-screened population. Past experiences have shown that protocol faults, which can greatly affect the performance of the system, are often connected with inadequate planning. Preparations for the programme should carefully consider a series of analyses and actions (figure 2), including: the assessment of disease incidence, which should take into account resident subpopulations from different ethnic backgrounds; the briefing of policy makers with precise information on the performance of screening strategies in their population, improvements in outcomes and containment of harm to families and society, and a clear evidence base on the effect on cost-effectiveness; and a model for language-appropriate information resources for parents.

Monitoring and improvement

The practice of NBS for cystic fibrosis is very varied and dependent on local facilities and preferences. Such heterogeneity is not necessarily a weakness since NBS protocols have to be adapted to local circumstances, such as the ethnic composition of the population, the prevalence of *CFTR* mutations, the practice of neonatal screening for other diseases already in place, the discharge policy of maternity points (ie, when newborn babies are discharged), and the accessibility of skilled sweat test facilities. Conversely, the absence of a unique model implies that NBS programmes might occasionally be inadequately planned and underperforming compared with worldwide acknowledged standards.^{19,21,61} Strategies for quality improvement are therefore essential. Crucial areas that need to be periodically assessed and if necessary improved

Search strategy and selection criteria

We searched PubMed for reports published in English between Jan 1, 1979, and July 31, 2015, with the search terms “newborn screening” AND “cystic fibrosis”. We also identified relevant trials and studies through hand searches of relevant journals (from 2000 to 2015) and abstract books of conference proceedings (from 1990 to 2015), including the European Cystic Fibrosis Conference and the North American Cystic Fibrosis Conference between 1990 and 2015. Selection for inclusion was on the basis of our skills and our perception of the relevance and effect of the report on the subject of newborn screening for cystic fibrosis.

are: validity and effectiveness of the system; potential inequities in access to NBS and discrimination of ethnic subpopulations not properly covered; efficiency of communication to parents from before the initial sample is taken through to receiving the sweat test results; results in terms of not only affected infants, but also false positives, carriers, and infants with an unclear diagnosis; and middle-term and long-term clinical outcomes (figure 2).

The ethical perspective of universal cystic fibrosis NBS

NBS for cystic fibrosis is clearly a valid public health strategy in populations in which cystic fibrosis is common. Whether NBS can be considered a basic human right with a corresponding duty to provide it is contested at the fundamental level of whether health care is a right.⁶⁵ Leaving this theoretical consideration aside, cystic fibrosis NBS can be examined in ethical terms and this analysis used to solve some of the unanswered questions.⁵⁶

Newborn screening for cystic fibrosis follows the original and revised guidelines for disease screening proposed by Wilson and Jungner.⁶⁶⁻⁶⁸ These guidelines consider the four key principles of biomedical ethics: respect for autonomy, beneficence, non-maleficence, and justice. Examples of these principles from NBS for cystic fibrosis include clearly established health benefits of screened infants, minimising harm by attempting to limit the number of false positives and carriers detected, genetic counselling for carrier families, and cost-effectiveness, which is important when health-care resources are limited. Further research is needed to minimise the number of infants with uncertain diagnosis after NBS. Ethical analysis could be used to help resolve issues related to emerging technologies (eg, the role of whole *CFTR* genome sequencing in screening), introduction of cystic fibrosis NBS in non-screened populations, and discrimination of ethnic subpopulations not properly covered by NBS.

Cystic fibrosis NBS should not be considered merely in terms of sensitivity and specificity, nor exclusively from a cost-effectiveness viewpoint, but rather as a bioethical model embracing all these factors and its performance judged in the context of its effect on society as a whole.

Contributors

CC selected the areas of interest included in the manuscript, contributed to the writing of the manuscript, supervised the production of the manuscript, and edited the final version. JM contributed to the planning, writing, and revision of the manuscript. MS contributed to the literature search and manuscript writing. KWS contributed to the literature search and manuscript writing and revision.

Declaration of interests

We declare no competing interests.

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