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Review Article

Newborn Screening – From 'Guthrie age to Genomic age'

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Abstract

Many countries across the world have made newborn screening (NBS) mandatory. It is a laboratory test which screens the newborns for metabolic & genetic disorders, some of which can be treated or modified if detected early in life, and thereby preventing potentially disastrous consequences and saving the baby's life. The last four decades have witnessed rapid evolution in implementation & strategies used for NBS in US, Europe, Japan & other industrialised nations where NBS is well accepted public health policy. India is going through a progressive transitional phase of control over infant mortality & morbidity due to infections and emergence of genetic conditions. This is the right time to review NBS program in totality considering the global scenario of its initiation, growth, advances in technologies & its transfer from conventional to mass spectrometry techniques, as well as selection & nature of candidate NBS disorders. Nevertheless, the impact of this worldwide movement of NBS is inadequate in India; this review article discusses the various efforts required to successfully introduce this significant health service for population benefits as well as the lacunae & limitations still exist in 21st century in India. Based on the high-risk screening of congenital metabolic conditions using mass spectrometry in India, the first hand experience of more than a decade is shared here to provide better opportunities & guidelines to those who have serious urge to pursue NBS as an important preventive public health program, be it at government, public or private level for the masses of India.

Introduction

Newborn screening (NBS) popularly known as neonatal screening is testing newborn babies for serious developmental, genetic, and metabolic disorders so that important action can be taken during the critical time before symptoms such as mental and/ or motor retardation, physical disabilities or death develops. It is the process of testing newborn babies for treatable genetic, endocrinologic, metabolic, and hematologic diseases before the development of symptoms because in the newborn period, inborn errors of metabolism (IEM) can easily be misdiagnosed as sepsis or birth asphyxia. The delay in diagnosis or undiagnosed IEMs can lead to severe mental deterioration and even death. The prompt detection therefore requires vigilance and the early & presymptomatic measurement of biochemical markers. NBS thus involves a simple blood or urine screening test done in apparently healthy babies soon after birth to identify many life-threatening genetic illnesses. It has become a popular form of preventive medicine and is of increasing interest internationally. Currently, it is the best example of ongoing preventive public health

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Fig 1a : Robert Guthrie-the pioneer of newborn screening

Fig 1b: The bacterial inhibition assay plate showing the bacterial growth around the disc

policy in many countries aimed at early detection, treatment and management of the newborns who may be affected with inborn metabolic disorders, congenital endocrinopathies, hemoglobinopathies and others. These disorders may be individually rare but their collective incidence is 1 in 1,500 –3,000. Their early & presymptomatic detection is significant as timely intervention, treatment and therapy can lead to reductions of morbidity, mortality and associated disabilities in affected infants, thus giving baby the best chance of healthy life.

History of Newborn Screening

NBS began in early 1960's by pioneering work of Dr. Robert Guthrie, USA (Fig. 1-a) with discovery of detecting Phenylketonuria (PKU) from dried blood spots (DBS) on filter paper, by a simple test as bacterial inhibition assay (Fig.1-b). His research work led to the now well-known 'Guthrie' card procedure which is nothing but blood absorbed onto the special thick filter paper to screen PKU in the newborns [1]. Much of the credit of the development of screening tests goes to Bickel and co-workers[2], when they successfully made dietary control on phenylketonuria in 1954. This remarkable breakthrough in the management of PKU paved way for innovations in detection techniques. Bickel wrote, "It is reasonable to presume that the best results of dietetic treatment of phenylketonuria will be obtained if treatment is started in infancy and particularly in the neonatal period."

The study justified the need of development of screening techniques for diagnosis of PKU and other IEMs. Soon, a less sensitive single disease ferric chloride test was introduced in 1960's for PKU screening[3]. The test was utilized largely in UK and US due to its convenience, quickness, and relative reliability. However, its less sensitivity always brought scope for development of other techniques. In the early period, the development of newborn screening differed in UK and US largely due to the efforts of Robert Guthrie who developed a bacterial inhibition assay (BIA).

Guthrie test

The name Guthrie test is to acknowledge the pioneering work of Robert Guthrie for the earliest screening for phenylketonuria in the late 1960s using blood samples on filter paper obtained by pricking a newborn baby's heel on the second day of life to get a few drops of blood (Fig 2-a & b). It is well known that Guthrie's efforts were motivated by the diagnosis of phenylketonuria in his niece affected by PKU. Furthermore, with the introduction of a system for collection and transportation of blood samples on filter paper (Fig.3-a), cost effective wide scale genetic screening became possible. The Guthrie test, also known as the Guthrie bacterial inhibition assay is a medical test performed on newborn infants to detect phenylketonuria, an inborn error of

Fig 2a : The heel of the neonate showing the pricking area for blood collection.

Fig 2b: The 'Guthrie' card showing blood soaked in the precise circles of filter paper

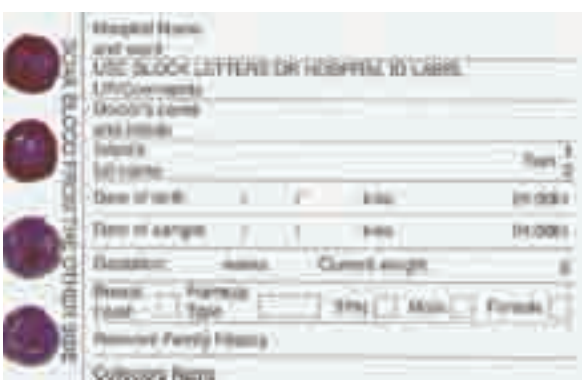

Fig 3a : The Guthrie card showing the blood spots & demographic data to be filled & sent to the LC/MS , MS/MS laboratory.

The urine soaked & air dried filter paper in a zip-lock plastic bag to be sent to the GC/MS laboratory

amino acid metabolism[4]. Although, widely accepted in the US in the late 1960's the test was not used in large parts of UK, where only ferric chloride test was used. This continued until the superiority of the inhibition test was proved & in 1970 the test was widely implemented in the UK. Since then, the test has been popular throughout North America and Europe as one of the core newborn screening tests[5].

The Guthrie test is a semi-quantitative assay designed to detect elevated blood levels of the amino acid phenylalanine, using the ability of phenylalanine to facilitate bacterial growth in a culture medium with an inhibitor. A drop of blood is usually obtained by pricking the heel of a newborn infant on the second or third day of life. The blood is collected on a piece of filter paper. A small disk of the filter paper is punched out and placed on an agar gel plate containing Bacillus subtilis and β-2-thienylalanine. The agar gel is able to support bacterial growth but the β-2-thienylalanine inhibits bacterial growth. However, in the presence of extra phenylalanine leached from the impregnated filter paper disk, the

Table. No. 1

High-Risk Screening by GC/MS

Metabolic Abnormalities in 1236/3195 Cases (38 %)

No	IEM Disorders (Total Cases)	Incidence of IEM	
		N=3195	Neonates N=827
1	Glutaric Aciduria (36)-Type-I:33 & II:3	1:88	1:276 (3)GA-II
2	Hyperglycinemia (28)	1:114	1:414 (2)
3	Maple Syrup Urine Disease(MSUD) (12)	1:266	1:118 (7)
4	Isovaleric Acidemia (4)	1:799	1:827 (1)
5	Multiple Carboxylase Def. (MCD) (13)	1:246	1:414 (2)
6	Methyl Malonic Acidemia (MMA) (43)	1:74	1:138 (6)
7	Propionic Acidemia(PA) (12)	1:266	1:138 (6)
8	Ornithine TransCarbamyl Def.(OTC) (5)	1:639	1:276 (3)
9	Tyrosinemia(4)/Hepatic dysfunction (4)	1:799	1:276 (3)
10	Urea Cycle Disorder (20)	1:160	1:83 (10)
11	Canavan Disease (10)	1:320	1:827 (1)
12	3-hydroxy-3-methyl glutaryl CoA lyase def.(6)	1:533	Not found
13	Fructose-1,6-Diphosphatase def.(9)	1:355	Not found
14	Galactosemia(3)	1:1065	1:827 (1)
15	3-ketothiolase deficiency (11)	1:290	1:827 (1)
16	Hyperphenylalaninemia (5)	1:639	1:414 (2)
17	Dihydropyrimidinase deficiency (2)	1:1598	Not found

(Source- Dave U.P., International Conf. Human Genetics, Brisbane, Australia, 2005)

inhibition is overcome and the bacteria grow[3]. Within a day, the bacterial growth surrounding the paper disk is visible to the eye (Fig.1-b). The amount of growth, measured as the diameter of the colony, is roughly proportional to the amount of phenylalanine in the serum. The result is read by comparing the diameter of each sample disk's colony to the colonies of a series of reference disks with standard phenylalanine content included on each large plate.

Population Screening

The early history of newborn screening echoes many of the similar controversies we still currently face. A population-based dried blood-spot screening (NDBS) received the general acceptance in early 1960s as an essential preventive public health activity[6]. The nickname of NBS test was a 'PKU test' in general population. Since then many feedbacks from the medical & social scientists helped in how to implement NBS as a universal screening program.

Prior to Robert Guthrie's development of the bacterial inhibition assay for PKU in 1961, no technology could reliably identify asymptomatic infants with PKU within the first week of birth, when treatment impact is greatest. In contrast, Guthrie's test was cheap, easy, and reliable. These were critical features for any test used in a mass screening program[7]. The movement from Guthrie's discovery to implementation of mass screening was swift, in part, because of previous case reports that suggested that a low phenylalanine diet could improve developmental outcomes in children with PKU2. In1962, Dr. Robert Guthrie and his co-workers in Mas-

Table No.-2 NBS Disorder Panel Recommended For Screening
(Source: http://www.mchb.hrsa.gov/screening/summary.htm.)

	Core Panel Disorders		Secondary Target Disorders
9 OA	Isovaleric acedemia (VA) Glutaric acedemia (GA 1) Hydroxymethylglutaric aciduria (HMG) Multiple Carboxylase Deficiency (MCD) Methylmalonic Acidemia Mutase Def. (MUT) 3-Methylcrotonyl –CoA Carboxylase Def.(3MCC) Methylmalonic Acidemia (Cbl A, B) Propionic Acidemia(PROP) β –ketothiolase Def.(BKT)	6 OA	Methylmalonic acidemia (Cbl C,D) Malonic Acidemia (MAL) Isobutyryl-CoA-Dehydrogenase Def. (IBG) 2-Methyl 3-HydroxyButyric Aciduria (2M3HBA) 2-Methylbutyryl-CoA-Dehydrogenase Def.(2MBG) 3-Methylglutaconic Aciduria (3 MGA)
Fatty Acid Disorders			
5 FAO	Medium-chain acyl-Coa Dehydrogenase Def.(MCAD) Very long-chain acyl-Coa Dehydrogenase Def.(VLCAD) Long-chain L-3-OH acyl-Coa Dehydrogenase Def.(LCAD) Trifunctional Protein Def.(TFP) Carnitine Uptake Defect (CUD)	8 OA	Short-chain acyl-Coa Dehydrogenase Def.(SCAD) Glutaric Acidemia Type II (GA2) Med/Short- chain L-3-OH acyl-CoA Dehydrogenase Def. (M/SCHAD) Med –chain Ketoacyl-CoA Thiolase Def.(MCKAT) Carnitine Palmitoyltransferase II Def. (CPT II) Carnitine Acylcarnitine Translocase def. (CACT) Carnitine Palmitoyltransferase I Def. (CPT IA) Dienoyl-CoA Reductase Def. (DE RED)
Amino Acids			
6 AA	Phenylketonuria (PKU) Maple Syrup Urine Disease (MSUD) Homocystinuria (HYC) Citrullinemia (CIT) Arginosuccinic Acidemia (ASA) Tyrosinemia type I (TYR I)	8 AA	Benign Hyperphenylalaninemia (HYPER-PHE) Tyrosinemia Type II (TYR II) Defects of Biopterin CofactorvBiosynthesis (BIOPT(BS)) Argininemia (ARG) Tyrosinemia Type III (TYR III) Defects Of Biopterin Cofactor Regeneration (BIOPT (REG)) Hypermethioninemia (MET) Citrullinemia Type II (CIT II)
3 Hb Pathies	Sickle Cell Anemia (Hb SS) Hb S/β –thalassemia (Hb-SβTh) Hb S/C Disease (Hb S/C)	1 Hb Pathies	Variant Hb-pathies (Var Hb)
6 Others	Congenital hypothyroidism (CH) Biotinidase Def. (BIOT) Congenital Adrenal hyperplasia (CAH) Classical Galactosemia (GALT) Hearing screening (HEAR) Cystic Fibrosis (CF)	2 Others	Galactokinase Def. (GALK) Galactose epimerase Def. (GALE)

sachusetts tested every newborn for PKU. The concept of NBS developed rapidly & soon included other inborn errors of metabolism (IEM), thereby a list of candidate disorders went on increasing in US. Each state had their own program with different disorders & methodology used. However, there were disparities in newborn screening services available across the country & acute need was felt for the uniformity.

In 1975, the World Health Organization and National Academy of Sciences described the principles underlying the effective newborn screening. American Associations Paediatrics Newborn Screening Task Force in 1999 recommended[8] that "Maternal and Child Health Bureau (MCHB) of Health Resource and Services Administration (HRSA) should engage in a national process involving government, professionals, and consumers to advance the recommendations of this Task Force and assist in the development and implementation of nationally recognized newborn screening system standards and policies." The Maternal and Child Health Bureau (MCHB) of Health Resource and Services Administration commissioned the American College of Medical Genetics (ACMG) & outlined a process of standardization, of outcomes and guidelines for State Newborn Screening Programs, defined responsibilities for collecting and evaluating outcome data, recommended uniform panel of conditions to include in State Newborn Screening Programs[9].

WHO Guidelines for NBS Program

NBS is not simply a test for disorders considered in the program, but it consists of other aspects too as education, follow up, diagnosis, management of the positive case and evaluation of program. World Health Organization (WHO) has issued guidelines and criteria for selecting disorders in NBS program of a particular nation which also have important ethical and legal implications. Wilson and Jungner in 1968 outlined the criteria for selection of NBS candidates – viz. the metabolic disorder must be common in a given population, must be an important health problem, suitable tests must be available, there should be facilities for diagnosis and treatment, & it must be a cost effective strategy[10]. Thus, before starting the regional NBS program in any country, the initiator must consider the points depicted below-

1. Recognize and understand the role of newborn screening in metabolic/ genetic disorder
2. Review past, present and future of newborn screening
3. Discuss clinical approach to metabolic disorders and identify management strategies
4. Understand the role of provider and caregiver in the management of common childhood illnesses in children with specific metabolic/genetic disorders.

Criteria for disease selection: -

1. Treatable disease.
2. Affordable screening test & not difficult to diagnose.
3. Requires immediate therapy to prevent disability and mortality.
4. Reasonably frequent in population

As a result of WHO interception, the NBS movement spread across the Asian countries, though the problems faced & difficulties encountered in implementation differed significantly from the Western countries11, 12. In 1983, sickle cell disease and in1994, HIV infection was added to the mandatory screening of all newborns in US. In 1990, the newborn hearing screening (NHS) was accepted in medical practice though its possibility was suggested way back by audiometry identification in 1960s [13].

A current scenario of NBS is well depicted by Therrell & his group in 2006, emphasising still a lack of US national NBS policy and each state having its own law for the required NB screening[14]. It is suggested that the program infrastructure should be contracted out to the private laboratories, partly or completely rather than provided within the existing public health system. The screening, patient tracking & follow-up should be contracted to academic & speciality genetic centres whereby more defined roles & responsibilities of every NBS system components such as education, screening, follow-up, diagnosis, management & evaluation can be effectively measured.

Among the Asian countries, Japan is on the frontier of adapting the NBS program. In 1977, Ministry of Welfare, Japan initiated screening for 5 IEMs (PKU, MSUD, Histidinemia, Homocystinuria & Galectosemia) in all neonates in Japan. Cretinism in 1979 & Neuroblastoma in 1984 were added to the list for all infants. As a developed nation, the advanced technology for better, faster & more sensitive tests like mass spectrometry are developed[15] & are rapidly replacing the

conventional method[16]. Today, China, Thailand, Pakistan, Singapore & Philippines are some of the Asian countries which have made efforts in starting NBS at the regional levels with the help of NGOs or external funding.

The Wilson-Jungner criteria10 for appraising the validity of a screening program are as follows:

1. The condition being screened for should be an important health problem
2. The natural history of the condition should be well understood
3. There should be a detectable early stage
4. Treatment at an early stage should be of more benefit than at a later stage
5. A suitable test should be devised for the early stage
6. The test should be acceptable
7. Intervals for repeating the test should be determined
8. Adequate health service provision should be made for the extra clinical workload resulting from screening
9. The risks, both physical and psychological, should be less than the benefits
10. The costs should be balanced against the benefits

Advances in Technology of NBS

During its 45 years of evolution, NBS has come a long way with tremendous increase in the number of metabolic/genetic disorders screened, type of sample used from invasive blood spots to non-invasive urine and improvements in the technology. Initially, the Bacterial Inhibition Assay: Guthrie's method, Beutler's method, enzyme-linked immuno assays (ELISA) & radioimmunoassays (RIA) used were a single assay method for screening each disorder. These tests consumed more time and had several disadvantages as:

- Low accuracy and sensitivity
- More frequency of false positive and false negative results
- One-test One-disorder – unsuitable for mass screening

Various methods were developed for a high-risk screening of diagnosing organic & amino acid disorders in the last decade & it has been found that simultaneous analyses of blood carnitine & amino acids by tandem mass spectrometry (MS/MS) is more rapid and multicomponent technique which can be used for NBS[17, 18]. From initial list of about 15-20 metabolic- amino, organic & fatty acid disorders, the number has increased to about 40 conditions which can be simultaneously screened from a single blood spot by LC/MS/MS. As capital cost is very high, one disease-one-test approach has also been used by the resource meagre countries using conventional RIA, ELISA or fluoroimmunoassay methods.

Since its first application by Tanaka in 1966, GC/MS has been used worldwide in diagnosis of IEM because of its high accuracy, sensitivity and power of analyzing multiple compounds simultaneously[19]. During the same period, to overcome the problem of invasiveness & to include more no. of metabolites-'a new chemical diagnostic method for Inborn Errors of Metabolism by mass spectrometry was developed by Matsumoto & his research group as a rapid, practical simultaneous urinary metabolite analysis in Japan20. More than 100 metabolic conditions can be simultaneously screened using gas chromatography/ mass spectrometry (GC/MS) for abnormal markers from the air dried urinary filter paper The same GC/MS method[20] was first time used by the author (U.P. Dave) to screen high-risk cases of IEM using urine filter paper (Fig. 3-b) in Indian patients[21]. This was the first study to indicate the common metabolic conditions in Indian population using GC/MS (Table 1).

Target NBS Disorders

Based on the earlier regional studies, NBS expert group of ACMG recommended a panel of NBS conditions as 'Core Panel' & 'Secondary Targets' (Table 2) which forms the guidelines for selection criteria. The following are some of the core panel diseases that have been routinely screened in the developed countries & incidence are made available by epidemiological studies in various countries. These are briefly described here, however the detailed are available on the Websites of HRSA[22, 23].

Congenital Hypothyroidism (CH) – Congenital hypothyroidism was the second disease after PKU widely added in the 1970s. Because of the early response to the treatment soon after the presymptomatic detection, CH was the ideal NBS candidate disorder all over the world. Affected babies don't have enough thyroid hormone and impair growth and brain development if goes unnoticed. The thyroid gland dysfunction leads to deficiency of hormone- TSH & /T4 that control metabolism and growth. The ELISA or radioimmunoassay methods

are used. If the disorder is detected early, a baby can be treated with oral doses of thyroid hormone to permit normal development. Incidence: 1 in 4,000, but is higher in India ranging from 1800- 280024 and is predicted still higher in iodine deficient endemic areas.

Congenital Adrenal Hyperplasia (CAH) - This is a group of disorders involving a deficiency of certain hormones produced by the adrenal gland; ambiguous genitalia, hirsuitism being clinically highly suspected features. It can affect the development of the genitals and severe form may cause death due to loss of salt from the kidneys. Lifelong treatment through supplementation of the missing hormones manages the condition. Incidence: 1 in 12,000. A sample survey from South India revealed incidence to be 1: 2575 25. It is an under-diagnosed condition in India with frequency ranging from 1.4 % to 38%.

Phenylketonuria (PKU) – As described above, mental retardation can be prevented when detected early, by feeding an infant a special formula- low in phenylalanine. The condition occurs due to a deficiency of the enzyme phenylalanine hydroxylase that converts phenylalanine into tyrosine. A low-phenylalanine diet will need to be followed throughout childhood and adolescence, and perhaps into adult life. This diet cuts out all high-protein foods, so people with PKU often need to take a special artificial formula as a nutritional substitute easily provided in US , Europe, Japan & other developed countries (not available in India.). Incidence is 1 in 10,000 to 25,000. The PKU was not found in more than three thousand high-risk cases screened for amino acid disorder (Table 1) indicating the low incidence in India & probably not the ideal candidate for NBS as in the Western countries.

Galactosemia (GA): This is a disorder of galactose metabolism, lacking the enzyme, galactose-1-phosphate uridyl transferase (GALT) that converts galactose (one of two sugars found in lactose) into glucose. Variants of GALT deficiency are milder forms. As a result, milk (including breast milk) and other dairy products must be eliminated from the diet. Otherwise, galactose can build up in the system and damage the body's cells and organs, leading to cataract, blindness, severe mental retardation, growth deficiency, and even death. Several less severe forms of galactosemia are detected by newborn screening. Incidence: 1 in 60,000 to 80,000. In India we yet do not know the incidence, but above typical clinical signs help the clinicians to diagnose galectosemia in infants. As seen in Table no.1, only 3 classical forms were found.

Biotinidase Deficiency (Multiple Carboxylase Deficiency or MCD)-

The deficiency of this enzyme indicates early symptoms like hypotonia, alopecia, seizures, skin rash, ataxia, conjunctivitis & hearing impairment. The other severe symptoms are mental retardation, coma, and even death. Biotinidase recycles biotin in the body. If the deficiency is detected in time, however, problems can be prevented by giving the baby extra dose of biotin & carnitine. The important point is the screening test is not performed by MS/MS, but by a colorimetric method. Incidence: 1 in 72,000 to 126,000. The patients with partial deficiency have enzyme activity between 10-30% & severe with, 10 %. It is relatively higher in our population as 13 cases were diagnosed (Table. 1) and were further confirmed by the enzyme assay.

Maple Syrup Urine Disease (MSUD) – It is caused due to a deficiency of important branched chain keto-acid dehydrogenase enzyme resulting into elevation of three amino acids- leucine, isoleucine & valine. When not metabolized properly, the accumulated substrates become toxic, excreted in the urine; smell like maple syrup or sweet, burnt sugar. Clinically babies usually have little appetite, extreme irritability, seizures, metabolic acidosis, mental retardation, physical disability, & even death if not detected and treated early. A carefully controlled diet that cuts out certain high-protein foods containing those amino acids can prevent the episode. Like PKU, those with MSUD are often given a special formula that supplies the necessary nutrients but low in proteins/ amino acids. The patients must follow special diet. Incidence: 1 in 250,000. The high-risk cases in our population showed MSUD as a common neonatal form resulting into death within a week with metabolic crisis (Table 1).

Cystic Fibrosis (CF) : Cystic fibrosis is a genetic disorder, considered as rare in India, that particularly affects the lungs and digestive system and makes children more vulnerable to repeated lung infections. There is no known cure and treatment involves trying to prevent serious lung infections and providing adequate nutrition. Early diagnosis by mutation study of F508del may help doctors reduce the problems associated with CF and identifying the carrier parents. But the real impact of newborn screening has yet to be determined. Inci-

dence: 1 in 2,000 Caucasian babies; less common in African-Americans, Hispanics, and Asians. Though considered rare in India, possibly due to under-diagnosis, the tertiary care hospitals have started reporting CF cases.

MCAD deficiency (Medium Chain Acyl CoA Dehydrogenase deficiency) : Children with this fatty acid metabolism disorder are prone to repeated episodes of hypoglycemia, which can cause seizures and interfere with normal growth and development. Treatment makes sure that patients don't fast (skip meals) and supplies extra nutrition (usually by intravenous nutrients) when they're ill. Early detection and treatment can help affected children live normal lives. MCAD is most common in comparison with LCAD(Long Chain AD) & VLCAD (Very Long Chain AD).

As seen in Table 2, there are group of Fatty Acid Disorders (FAD) that involve enzymes along the fatty acid oxidation pathway. These errors present wide variety of clinical spectrum as patients are healthy until a catabolic or fasting state when they experience Reye-like syndrome, viz. lethargy, nausea, vomiting & metabolic crisis, often with non-ketotic hypoglycemia. Neonatal onset has varied picture of failure to thrive, developmental delay, seizures & myopathy. Ongoing treatment includes supplementation of carnitine & a low fat diet with avoidance of fasting.

Sickle Cell Disease : Sickle cell disease is inherited blood disease in which red blood cells mutate into abnormal "sickle" shapes and can cause episodes of pain, damage to vital organs such as the lungs and kidneys, and even death. Young children with sickle cell disease are especially prone to certain bacterial infections, such as pneumonia and meningitis.

Studies suggest that newborn screening can alert doctors to begin antibiotic treatment before infections occur and to monitor symptoms from worsening more closely. The screening test can also detect other disorders affecting hemoglobin. Incidence: about 1 in every 500 African-American births and 1 in every 1,000 to 1,400 Hispanic-American births; also occurs frequently among people of Mediterranean, Middle Eastern, and South Asian descent. Sickle cell disease is one of the most common hemoglobinopathies in India, especially in tribal community. The gene frequency in different population may indicate the need of this & other hemoglobin disorders to be considered for NBS in different geographic & racial population.

Hearing Screening : Hearing test for newborns is very necessary. This is conducted before they're discharged from the hospital. It is important for the parents to ensure that neonate gets screened within the first 3 weeks of life. The children develop critical speaking and language skills in their first few years. If the hearing aid is provided at the right time, the speech & language acquisition can be restored which otherwise could have occurred due to sensorineural hearing loss[13].

Progress in basic sciences and advances in technology, we have gained insight in the biochemical and molecular basis of hereditary metabolic diseases[26]. The application of tandem mass spectrometry (TMS) to newborn screening for inborn errors of metabolism offers the potential of substantially altering the natural history of these diseases by reducing morbidity and mortality[27]. Many fatty & amino acid disorders can be successfully diagnosed by use of acylcarnitine profiles generated by TMS. However, the number of screen positive tests need to be diagnosed by confirmed tests such as actual enzyme assays for deficiency, GC/MS analysis or by DNA testing. The back-up support of genetic laboratory, experts & genetic counselors under one roof at the Central Laboratory is the key for a correct diagnosis and successful NBS.

Indian Scenario

The lack of health insurance facility for NBS is one of the major hurdles giving this test less priority in India as well as other national health priorities. Indian population has already crossed the mark of one billion. Our annual birth rate is 21.76/1000 population. Out of the approximately 25 million new births in India, there are an estimated 1.6 Million babies born with birth defects including about 620,000 with genetic disorders. With no mandatory newborn screening at national/ government levels, the countrywide burden of genetic disorders especially the congenital conditions is huge and still unknown[28]. We simply follow the Western data for incidence of common genetic disorders[21]. However, the importance of it has been recognized by ICMR. After 2 years of brain storming sessions with experts from all over India, ICMR has undertaken a pilot project (2007) at national level for 2 disorders, viz. CH & CAH screening of 100,000 newborns free of cost from 4 zones (East, West, North & South) of India to study their prevalence. The author -U. P. Dave was a member

of this Task Force Committee of ICMR for Newborn Screening. As the NBS awareness is taking momentum, many private players have come forward using technologies as immunoassays, RIA, & Tandem MS. The health economists in other countries have already proved the cost effectiveness of newborn screening & hence the services are well accepted. Although screening at present in India seems to be cost-intensive, the costs far exceed the benefits obtained and the amount spent in the long run considering prevention and rehabilitation cost of the disabled child!

A pilot newborn screening program using dried blood spots from heel prick was initiated by CDFD at Hyderabad in 1988 & 12,500 newborns were screened for aminoacidopathies, Congenital hypothyroidism (CH), Congenital Adrenal Hyperplasia (CAH), Glucose-6-phospahte dehydrogenase deficiency (G-6-PD), etc. CH (1 in 1700) followed by CAH (1 in 2575) emerged as most common disorders. But this was only for the state of Andhrapradesh[24, 25]. In January 2007, the Union Territory (UT) of Chandigarh was declared the first state or UT in India to fund mass genetic screening (NBS and prenatal diagnosis), with a reduced fee for all and free access for the poor[29] In February the following year, Goa was said to be the first state to introduce mandatory NBS[30]. Finally, in February 2009, the union cabinet was reported to have approved a proposal to establish the institute in Kalyani (West Bengal) to launch a large scale programme for NBS[31]. The most common IEMs revealed by high-risk screening are MMA, PA, MSUD, glutaric aciduria type-I, hyperglycinemia, urea cycle disorders, FDPD & 3-ketothiolase deficiency (Table 1).

In our experience, it is not the technology that is preventing Indian babies from getting screened, be it newborn or high-risk screening. But lack of awareness about genetic screening and knowledge about the latest technologies among the medical professionals, as well as our different national health priorities are the main contributing factors for the delay in implementing NBS program. As in the case of introducing iodinated salt or compulsory polio vaccination, the support & advocacy by the Govt. of India is a prime factor at the population level. Government support will go a long way in establishing national level newborn screening programs in collaboration with private laboratories as a countrywide network system similar to that going on in China. The support should also be of huge aid in creating awareness among the clinicians and the layman which is an absolute must for the success of any newborn screening program. The key point here is that genetic/ metabolic disorders can happen to anybody with/ without family history of it and a simple test is available to detect them, but the timing of the test is of essence. Such programmes should be analogous to the immunisation programme that has been successfully implemented all over India, as our babies need better and our future deserves better.

Nevertheless, it is also the moral responsibility of those professionals caring for the neonates to inform & educate the parents about the newborn screening & explain them its long-term benefits & cost-effective approach of prevention of disabilities. The primary care physician & paramedical staff like nurses, midwives need to be educated. In the absence of NBS, the screening of high-risk (NICU & PICU babies) cases viz. neonates and infants is the need of today[21]. This will not only help in reducing the mortality and morbidity but will produce important genetic epidemiological data which is currently lacking in India.

Conclusion

Important questions about the NBS process remain unanswered. For an infant with a positive screening result, what barriers does a primary care physician face in coordinating a medical evaluation and communicating with the family? What obstacles do families confront in the time after a newborn screening result returns positive? When a positive newborn screen result is confirmed, how can coordination of follow-up care be optimized? These fundamental questions must be addressed to optimize collaboration between primary care and specialty care physicians by public private partnership, and to ensure the continued success of newborn screening in the 21st century.

Benefits of NBS do not end only with saving life of the diagnosed case but they extend up to prenatal period of diagnosis and family genetic counseling, thus reducing the national burden of genetic disorders. It is also true that that the fruits of genomic science should not remain a luxury available only to the developed nations. The next technological advance, tandem mass spectrometry, DNA microarrays is on the horizon and fast approaching [32, 33]. Yet, it should be noted that technology is only one facet of a well-functioning newborn screening program, which must have both excellent detection and follow-up services. The ethical, social & legal implications should not be overlooked[34]. The challenge fi-

nally in India is the ultimate coverage of 100 % screening of neonates & infants which can only be achieved with a political will & financial commitment considering our socioeconomic infrastructure.

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