Original Research Article

Incidence of Congenital Hypothyroidism and G6pd Deficiency in New Born

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Abstract

Congenital hypothyroidism is the most common congenital endocrine disorder in childhood and also is one of the most common preventable causes of mental retardation. The incidence in India is estimated to be 2.1 per 1000 live births.G6PD deficiency is also a common condition, affecting around 400 million people worldwide and is characterized by considerable biochemical and molecular heterogeneity. A higher incidence of G6PD deficiency is seen in tropical and subtropical zones of the world. **Aims and Objectives:** 1. To estimate the incidence of congenital hypothyroidism and G6PD Deficiency in newborn population born in a tertiary care centre. 2. To study the natural history of screen positive cases. **Materials and Methods:** The study was conducted on 1555 patients in the department of Paediatrics, Nalanda Medical College and Hospital, over a period of 18 months from Apr 2019 to Sept. 2020. Umbilical cord mixed blood samples were collected in a sterile and EDTA container, drawn from placental side of the umbilical cord incised while severing it at the time of birth. **Results:** The presenting age of the mothers ranged from 18 to 43 years with an average age of 27 years. Out of the 1027 mothers, the highest peak i.e. 430mothers (41.04%) were aged between 25 to 30 years. Mothers who where aged > 36 formed the lesser group. Of the 1555 neonates whose cord blood samples were analyzed 43 neonates had TSH values more than 20 mIU /L. On re estimation of TSH and T4 values more than 72 hrs later all cases who were found to have higher TSH values in cord blood had age appropriate TSH and T4 values. Of the 1555 cord samples 7 of them were found to be G6PD deficient (value taken as less than 6.95 mU/g of Hb). **Conclusion:**The present study adds emphasis on the need for continuing screening for the most important preventable cause of mental retardation. Similarly there were 7 cases of G6PD deficiency found on cord blood estimation. This study also showed that there is no significant difference between cord blood and venous sample values indicating that cord blood estimation suffices for screening purposes.

Keywords: G6PD Deficiency, Congenital Hypothyroidism.

Introduction

G6PD deficiency is a very common condition, affecting around 400 million people worldwide and is characterized by considerable biochemical and molecular heterogeneity. A higher incidence of G6PD deficiency is seen in tropical and subtropical zones of the world[1].During periods of oxidative stress, G6PD deficiency causes inefficient removal of free radicals from RBCs resulting in haemolysis and jaundice. G6PD deficiency detection by neonatal screening is feasible and cost effective. It also allows the early preventive measures against severe haemolysis, jaundice, kernicterus etc. to be implemented in neonatal life, as well as other preventive measures in later life. Some practical guidance to the family members of G6PD deficient babies regarding the food items, chemicals and drugs to be avoided will be beneficial, if we diagnose the condition at first opportunity.Incidence of G6PD deficiency varies in different parts of India. Data from Chandigarh suggests an incidence of 1 in 112 & data from eastern India suggests an in incidence of 1 in 15. The burden due to this disorder is likely to be nearly 3, 90,000 births per year[2]. In West Bengal there is dearth of research on G6PD deficiency. With this perspective the present study was undertaken among newborns in department of Pediatrics, NMCH, Patna. Congenital hypothyroidism is the most common congenital endocrine disorder in childhood and also is one of the most common preventable causes of mental retardation. The incidence in India is estimated to be 2.1 per 1000 live births which is at least 8 times higher than what is reported in western literature[3].The clinical features of congenital hypothyroidism are often subtle and many newborn infants remain undiagnosed at birth. This is due in part to passage of maternal thyroid hormone across the placenta providing a protective effect, especially to the fetal brain and masking the clinical signs. Also, even the most common forms of CH have some moderately functioning residual thyroid tissue making clinical diagnosis difficult. Within few weeks of birth as hypothyroxinemia progresses clinical signs and symptoms of hypothyroidism become more obvious and put neonatal brain at risk of irreversible injuryAfter making diagnosis, if the treatment is started within in a few weeks of birth, neurodevelopmental outcome is generally normal. Because of this danger, it is important to start treatment as soon as possible after birth. For all the above reasons, screening has become the best way to detect infants with CH.Pilot screening programs were first developed in Quebec, Canada and Pittsburgh, Pennsylvania in1974. In India the incidence of congenital hypothyroidism is around 1:1700 births, in one study done in Kochi the incidence was 1:1000 births. In western literature the incidence is around 1:4000 births[4,5]. Despite the overwhelming evidence of high prevalence of CH in India, there is no universal screening program for neonatal hypothyroidism. This study aims to estimate

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the prevalence in the region and highlight the need neonatal screening program.

Aims And Objectives

1. To estimate the incidence of congenital hypothyroidism and G6PD Deficiency in newborn population born in a tertiary care centre.
2. To study the natural history of s screen positive cases.

Materials and Methods

The study was conducted on 1555 patients in the department of Paediatrics, Nalanda Medical College and Hospital, over a period of 18 months from Apr 2019 to Sept 2020. Umbilical cord mixed blood samples were collected in a sterile and EDTA container, drawn from placental side of the umbilical cord incised while severing it at the time of birth.

Inclusion criteria

All babies born in Nalanda Medical College and Hospital, Patna was included for the study and continued for 18 consecutive months.

Exclusion Criteria

Prematurity less than 32 weeks and severe perinatal asphyxia requiring extensive resuscitation.

The mother's age, parity, comorbid conditions like diabetes, PIH, hypothyroidism, any previous history of child /themselves with G6PD deficiency, unexplained anemia, jaundice requiring exchange transfusion was recorded. The type of medications given to the mother till birth of the baby was recorded. At birth, the babies weight, gender, apgar, congenital abnormalities, were noted.

G6PD was estimated within 48 hours by quantitative method using ILab 650 fully automated analyzer. All babies wherein the cord blood G6PD levels were less than 6.95 mU/g of Hb was taken as deficient. Such babies were followed up and repeat G6PD estimation was done at 3 months of life.

TSH was estimated within 24 hours by electrochemiluminescence immunoassay 'ECLIA' on elecsys 2010 analyser. All babies wherein the cord TSH was found to be over 20mIU/L were intimated within 24hrs of the test. A second venous blood sample from these babies for serum T4 and TSH estimation was collected between 2- 4 day of life.

Quantitative method of G6PD estimation: The estimation of G6PD levels was a quantitative method based on UV method.

Statistical Analysis

All the data was entered into Microsoft Excel 2012spreadsheet and analysed. The various clinical parameters like gestational age and birth weight were correlated with the cord blood TSH levels using the Contingency coefficient analysis (Cross tabs procedure). Chi square test and student 't' test were used to test the nominal significance at the p value < 0.05 level, for high significance at the p value was < 0.01 and for not significant at the p value > 0.05. Descriptive statistics of the various clinical and laboratory parameters and measures of central tendency using the mean and median with standard deviation have been performed.

Results

The results obtained are as follows.

The presenting age of the mothers ranged from 18 to 43 years with an average age of 27 years. Out of the 1027 mothers, the highest peak i.e. 430 mothers (41.04%) were aged between 25 to 30 years. Mothers who were aged > 36 formed the lesser group. GDM was the most common maternal co morbidity noted followed by PIH and Hypothyroidism. Of the hypothyroid mothers two were found to have anti- TPO antibodies. Other common comorbidites like anemia and UTI were not taken into account. The average gestational age was 38.2 weeks. The longest POG was 41 weeks 1 day. 172(16.5%) of cases were between 32 to 37 weeks of gestation, and the rest were more than 37 weeks of gestation. The average birth weight was 2.93 kgs. The highest recorded was 4.22 kgs and lowest was 1.7kgs. Descriptive Statistics of TSH levels and G6PD levels are mentioned in Table 1 and 2

Table 1:Descriptive Statistics of TSH levels

Parameter		No of Patients N=1037	Cord Blood TSH		P Value
			TSH <20	TSH>20	
Gestational Week	32-37 weeks	216 (13.8)	210	6	0.26
	>37 weeks	1312(84.3)	1270	42	
Birth weight	1.5-2.49 kgs	223(14.3)	217	6	0.482
	> 2.5 kgs	1332(85.6)	1290	42	
Maternal age	<20 yrs	79(5.11)	73	6	0.868
	21-25 yrs	531(34.1)	515	16	
	26-30 yrs	639(41.0)	624	15	
	31-35 yrs	253(16.29)	244	9	
	>36 yrs	52(3.3)	50	2	
Maternal comorbidites	GDM	249(16.0)	244	5	0.98
	Hypothyroid	97(6.2)	92	5	
	GDM with hypothyroidism	39(2.5)	37	2	
	Others	111(7.1)	108	3	
	Non diseased	1098(70.5)	1062	36	
Neonatal Comorbidites	NNJ	97(15.9)	89	8	0.9847
	Mild Birth Asphyxia	22(1.4)	20	2	
	TTNB/RDS	28(1.8)	25	3	
	Others	45(2.8)	43	2	
	No illness	1212(77.9)	1176	36	

Table 2:Descriptive Statistics of G6PD levels

Parameter		No of Patients N=1555 (%)	G6PD values		P Value
			G6P <6.95	G6PD > 6.95	
Sample type	Umbilical	1523 (98)	7(0.9)	1519(99.5)	0.02
	Venous	47 (3)	0	47	
Gender	Male	761 (49)	5(0.7)	756 (99.3)	0.30
	Female	794 (52)	2(0.1)	792(99.9)	
Birth Weight	1.5-2.49 kgs	234(15)	1	233(14.09)	0.91
	> 2.5 kgs	1321(84)	2	1319 (81.9)	

Hyperbilirubinemia	Require phototherapy	249 (16)	3 (0.18)	246 (15.4)	0.002
	No Jaundice/not requiring phototherapy	1506(85)	3 (0.28)	1503(4.9)	

Discussion

Thyroid hormones are necessary for normal development of the human fetal brain and the maturation of other organs. Insufficient production of thyroid hormones during the fetal and neonatal period may result in serious complications and the central nervous system is affected most. It plays an important role for myelination and for normal neuronal connections.[6] Cord blood TSH screening for CH is a simple and accessible procedure. Previous studies have shown a transient TSH surge in the first 24–48 hours of life.[7] However, the measurement of cord serum TSH for CH screening is well established.[8] Walfish*et al*. suggested that cord TSH had a better specificity and sensitivity as compared with cord or filter paper T4 at 3–5 days of age.[9] Fuse *et al*. showed that cord serum is a good sampling technique for screening CH.[10] Mahachoklertwattana*et al*. showed that, if TSH is measured for screening CH, samples should be obtained from the umbilical cord of infants.[11] In India, Singapore, Japan and Ethiopia, cord serum TSH levels have been used for neonatal screening for CH because of the difficulty of calling neonates back.[12,13,14] Newer TSH assay techniques, such as the enzyme-linked immunoassays, chemiluminescent assays and fluoroimmunoassays offer the advantages of using non-radioactive labels and greater sensitivity with the potential for better separation between normal and abnormal TSH concentrations. Thus, many screening programs are considering switching to a primary TSH approach. A majority of European and Japanese programs favor screening by means of primary TSH measurements, supplemented by T4 determinations for those The neonatal serum FT4 levels rapidly increase after delivery to the maximum level at 1 day of age. Thereafter, they decline to a steady state level within 2–4 weeks. After a transient TSH surge in the first 24–48 hrs of life, neonatal serum TSH levels decline and the level at 1–3 days of age is similar to that of the cord serum.[15] It changes little after 3 days of age. Therefore, for those infants with initial cord blood TSH > 20mIU/L a repeat blood sample should be obtained after >48 hours. However, the trend towards early discharge of infants and mothers presents problems with this approach. This would result in an unacceptably high recall rate for this group of infants unless the TSH cut off was adjusted for age. Experience using newer assays in a primary TSH screening approach, in a population of infants discharged early, is necessary to determine the effects on recall rates and the possibility of any false-negative test results. infants with elevated TSH values. [16] Incidence rates vary by race or ethnicity. Among Asian Indians, 1:1,200; Hispanic, 1:1,600; Asian (Chinese and Vietnamese), 1:2,380; non-Hispanic White, 1:3,533; and non-Hispanic Black, 1:11,000.[17] Incidence was higher in preterm, low birth weight babies: more than 2500 grams, 1:1843; 1500–2500g, 1:851; and less than 1500 grams, 1:396. Harris and Pass [18] reported a 23% increase in babies born weighing less than 1500 grams.[19] In this study there were no confirmed cases of CH when screened by cord blood TSH estimation. This may be due to a low prevalence in the population and a relatively smaller sample size compared to other studies.

In the United States, the recall rate after primary TSH screening is approximately 0.05%.[20] In the study conducted by Azizi and colleagues in Tehran and Damavand using cord blood samples for screening of CH, a recall rate of 1.06% was obtained with a TSH cut off level of more than 20mIU/L,[21] whereas in Esfahan, the recall rate was approximately 2.2% after primary screening for serum TSH levels using the same Cut off limit of 20mIU/L.[22] These varying recall rates for different TSH cut off levels may be because of several factors, such as the use of T4 or TSH level or both for screening, differences in sample-collection methods and analysis procedures in different laboratories, and differences in recall criteria, which are related to the cultural, regional, and social factors of a country.[23] The recall rates in other countries, after primary TSH level assessment in neonates aged 3–5 days, may vary from 0.2% to 3.3%. The recall rates with TSH cut off of more than 20mIU/L were 0.16% in the Philippines, 0.35% in Austria, 0.3% in Greece, 0.28–0.29% in Hungary, 2.3% in Turkey, and 3.3% in Estonia.

In contrast, studies conducted in Italy, the recall rate measured on the basis of T4 levels was 2.5%, while that measured on the basis of both T4 and TSH levels was 0.11%.[24] In this study, using only primary cord blood TSH for screening congenital hypothyroidism we had a recall rate of 6.23%. If the cut off is raised to 40 mIU/L we would have a recall rate of 0.6%. Therefore, the laboratory screening methods and TSH cut off level need to be revised to ensure more specific and sensitive CH screening. Recent studies have shown a high prevalence of CH and high patient recall rate after primary screening, which was in line with the results of previous studies in Iran.Although environmental and genetic variations in addition to the low cut off TSH level may be responsible for the high recall rate, a nationwide study is necessary to clarify the reasons for the high incidence of CH. Future studies should also be able to clarify why small changes in TSH cut off levels during screening lead to substantial changes in the number of neonates with undetected CH. The downside to lowering TSH cut offs is an increase in recall of infants with false-positive tests. Each screening program needs to work out its own test cut off, weighing increased detection of mild cases *vs.* harm from recall of normal infants. In our opinion, until there is good evidence of no intellectual impairment, we can come down on the side of detection and treatment of these milder cases.[25] The study of a birth cohort in Southern Spain revealed an impaired mental development at 4 years of age in children with higher neonatal TSH levels compared with children with lower neonatal TSH levels within the normal reference range. These findings indicate that a more thorough screening for neonatal thyroid deficiency is required to prevent long-term developmental effects. Further research is warranted into the influence on neurodevelopment of marginally altered TSH concentrations in newborns.[26] The high recall rate in this study can be attributed to the low cut off for cord blood TSH.

CH was diagnosed if cord blood TSH > 20mIU/L and repeat serum TSH was above the age appropriate cut off. In this study, of the total 1555 deliveries, cord blood was available for 1512 infants. The missed cord blood was attributed to emergency deliveries and lost to follow up cases. Of the 1512 infants, 127 (4%) infants had cord blood TSH > 20mIU/L. Repeat TSH levels were done on all infants after 72hr of delivery, which were found to be normal. Neonates in whom cord blood could not be taken, a venous sample was taken after 72 hrs for TSH measurement. 123 venous samples of such neonates were estimated for TSH and were found to be normal. In a study done in Mumbai, all cases of confirmed congenital hypothyroidism had a cord blood TSH level of > 30 mIU/L, [26]. The maximum value obtained here was 63.15 which on repeat sampling was found to be normal.In our study G6PDdeficiency was found in 7 cases, 5 in males (0.7%) and 2 in females (0.1%). Saifalsaif et al studies found gender specific incidence of G6PD deficiency of 3.06% in males and 0.85% in females. This difference could be because of small sample size of our study.

Conclusion

The present study adds emphasis on the need for continuing screening for the most important preventable cause of mental retardation. There were no positive cases found in this study, probably due to decreased sample size. In India and other developing nations primary cord blood TSH followed by serum TSH and fT4 if cord blood TSH is above cut-off levels, seems to be the best cost

effective and sensitive screening tool. This study showed a high recall rate of 3.04% when compared to other studies. The reason for the high recall rate was possibly a low cord blood cut-off level. Screening for congenital hypothyroidism was taken up for the first time in our hospital. Hence a safe cut-off of level in order to prevent false negative cases was implemented. However from this study, it is evident that both the truly positive cases had a Cord TSH which was >20mIU/L. Similarly there were 7 cases of G6PD found on cord blood estimation. This study also showed that there is no significant difference between cord blood and venous sample values indicating that cord blood estimation suffices for screening purposes. A higher average obtained for G6PD values from venous samples indicate that cord blood estimation has a higher sensitivity. There was no statistically significant difference when G6PD levels of normal neonates were compared with those neonates who had physiological jaundice requiring phototherapy.

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