Establishment of reference interval for cord blood TSH

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Abstract

Introduction: TSH values in the cord blood is useful for neonatal screening of thyroid disorders. Screening is essential to rule out congenital hypothyroidism in neonates without any physical signs. In South Indian population, the reference interval for cord blood TSH has not been established.

Aim: To establish the reference interval of cord blood TSH in South Indian population.

Methods and Material: This study was conducted at Dr. SMCSI Medical College, Karakonam, Trivandrum. 200 neonates were selected as reference individuals for cord blood collection. The rejection criteria were complications in the delivery or hypothyroidism in the mother or the neonate. TSH was estimated in the cord blood by chemiluminescence method in VITROS ECi.

Statistical Analysis: The data obtained was analyzed using SPSS version 16. Paired T test was used for statistical analysis. The reference interval was determined by nonparametric method by taking middle 95 percentile. Correlation analysis was done using Pearson correlation coefficient.

Results: The reference interval for cord blood TSH is 2.2 to 18 mIU/L. The results showed that there is no significant difference between male and female neonates as well as the normal and the Caesarean section. Correlation study showed that cord blood TSH level was independent of the gestational age. Hence subgroup reference interval was not required.

Conclusions: The reference interval for cord blood TSH is from 2.2 to 18 mIU/L. Establishment of subgroup reference interval is not required since there is no significant variation between male and female neonates as well as between normal and caesarean section.

Keywords: Cord Blood, TSH, Reference Interval

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Introduction

The nervous system development and maturation depends on the vital role played by the thyroid hormones.(1) The development of nervous system of the fetus in the first and second trimester depends on the maternal thyroid levels. The maternal iodine levels also influence the fetal and neonatal thyroid hormones.(2,3) The neuronal development and the functioning capability of the fetus is affected by iodine deficiency in the mother.(2,3) Reduced thyroid hormones supply to the fetus and the neonate are indicated by raised level of TSH.(4)

Congenital Hypothyroidism may occur in the neonate due to defective thyroid gland, improper TSH secretion from the pituitary gland, iodine deficiency or inborn error of thyroid metabolism. It is considered as one of most common disorder associated with both mental and growth retardation.(5) This poses a major socioeconomic burden to the family and the society.

Since treatment and prevention of mental retardation is possible in congenital hypothyroidism, neonatal screening plays a vital role. Blood samples obtained after 24 hours of age by heel prick is used for screening.(6,7) However difficulty in obtaining samples because of early discharge from hospital, there is a higher probability that some neonates are not screened. Cord blood samples are at more ease of collection and all the neonates can be screened. The Indian Academy of Pediatrics also recommends cord blood as the sample of choice for screening.(8) Very few studies are done to establish the cord blood TSH levels. Hence we aimed at determining the reference interval of cord blood TSH in South Indian population.

Materials and Method

The study was conducted at Dr. SMCSI Medical College, Karakonam, Trivandrum after obtaining the ethical clearance. Individuals taken for the study were neonates from 200 sequential deliveries. Mother's history and neonatal data were collected. The following inclusion and exclusion criteria were applied for the study group

Inclusion criteria

- Gestational age of 38 – 42 weeks
- Normal vaginal delivery and caesarean section
- Babies with normal physical and mental development (assessed in the well-baby clinic at the time of vaccination – Oral Polio Vaccine).

Exclusion criteria

- History of hypothyroidism in mother
- Drug intake
- Gestational diabetes mellitus
- Pregnancy induced hypertension

- Physiological jaundice
- Preterm delivery

Cord blood samples were collected for the estimation of TSH after obtaining informed consent from the mother. Yellow topped gel vacutainer was used for cord blood collection. The serum was separated after centrifugation and stored at - 20 C until analysis. TSH was estimated in batches by chemiluminescence method in VITROS ECi.

Statistical Analysis: Data analysis was carried out using statistical package for social science (SPSS vs 16). The mean, median, standard deviation and 0.25 to 99.75 percentiles were determined.

T-tests was used to compare the means of TSH values of male and female neonates and in normal vaginal delivery and caesarean section. In the above test 'p' value less than 0.05 was accepted as statistically significant.

Pearson Correlation was done to analyze the differences in TSH values according to the variable (total number of gestational age). The 'p' value less than 0.05 is accepted as statistically significant.

Results

The median TSH level in the sera for all the neonates was 7mIU/L. Standard deviation is 5.1mIU/L and the mean value 8.1mIU/L. The percentile cut-offs of the TSH levels in the cord sera of all the neonates are presented in the Table: 1 the lower limit (2.5th) and upper limit (97.50th) of the TSH percentile cut-off levels in the cord serum samples for all the neonates were 2.2 and mIU/L and 18mIU/L.

Fig. 1 shows the type of distribution of values determined by plotting Histogram and skewed normal distribution was obtained. The degree of skew is the difference between the mean and median is less than 1/3 of standard deviation. So, the distribution doesn't require a logarithmic transformation. The sample size of 200 is not too high to take it as normal distribution. So, non-parametric method for reference interval determination would be followed. The 2.5 percentile to 97.5 percentile interval would be the reference interval.

Table 1: Percentile Values of Upper Seven and Lower Seven Rank Numbers

Rank No.	TSH Values	Percentile Values
1	1.3	0.25
2	1.7	0.75
3	1.8	1.25
4	2.1	1.75
5	2.1	2.25
6	2.2	2.75
7	2.4	3.25
→→	→	→
194	17.4	96.75
195	18	97.25
196	20.4	97.75
197	21.5	98.25
198	25.6	98.75
199	38	99.25
200	43.8	99.75

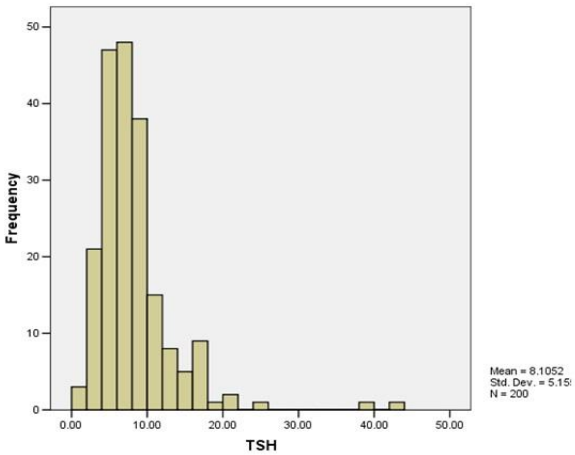

Fig. 1: The Histogram shows the skewed distribution of TSH levels of Reference Individuals

To analyze the correlation between the gestational age at the time of delivery and the cord blood TSH level, Pearson correlation coefficient was determined (Table 2) and the scatter plot of these two variables (Fig. 2) was examined. The Pearson correlation coefficient was -0.41 and there is no significant correlation p = 0.567. The scatter plot also showed no distinct pattern.

Table 2: Correlation between TSH and the Gestational Age

		TSH	Total Days
TSH	Pearson Correlation	1	-.041
	Sig. (2-tailed)		.567
	N	200	200
Total Days	Pearson Correlation	-.041	1
	Sig. (2-tailed)	.567	
	N	200	200

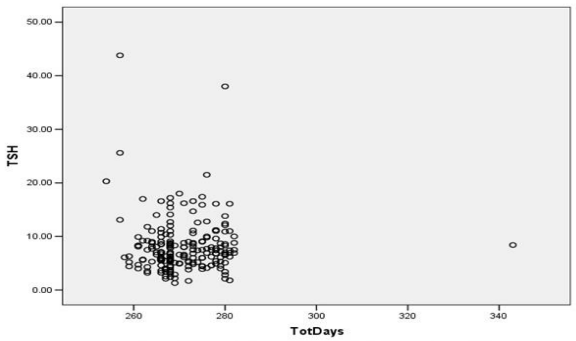

Fig. 2: The graph- (Scatter Plot) represents the insignificant variation with respective of the Gestational Age

Discussion

The results demonstrated that there were three neonates with greater than 20 mIU/l TSH values in the study population. Some studies have shown that the threshold value for a significant TSH elevation to be 20-25 mIU/l for blood samples taken from heel pricks. In a study conducted at North America by Klein et al the TSH level in cord blood samples was less than 20μU/ml in 85% and higher than 60 in 0.04% of the cases.[12] In Ethiopia, Feleke and co-workers measured TSH from heel pricks in neonates from six hours of life up to seven day sold infants. They determined the cut off point for TSH to be 29.4 mIU/l. Since there are numerous iodine deficient areas, it may be reasonable to take a lower cut off value for TSH.[13] In the present work the 97.8th percentile of the TSH value was 18.0 mIU/l, which is closely comparable to a study conducted by J. G. Henry et al in Kingdom of Saudi Arabia the reference ranges for cord blood TSH is 2.38 -19.06mIU/L. Reference ranges for cord blood TSH based on 2.5 and 97.5th percentiles is 2.4 - 20.6mIU/L.[14]

At a population level, neonatal TSH has the major advantage of being a single indicator that allows prediction of possible impairment of mental development. Primary TSH screening is the most cost-effective program which provides the highest detection rate and a very acceptable low recall rate. The screening test for primary hypothyroidism should be done either on a cord specimen or a specimen collected after 24 to 48 hours of life. The maternal wards of the hospitals in the present study give services to hundreds of women daily and the mothers are usually discharged in about 24 hrs. It is apparently difficult to get heel prick samples from new-borns at a later date. However, getting the cord blood was found to be practical.

The results demonstrated that cord blood samples were easy to collect and applicable for TSH assay, which can be used in routine screening.

Conclusion

The present study established the following in 200 reference individuals:

- The reference value for cord blood TSH is 2.2-18mIU/L.
- The female and male cord blood TSH level variation is insignificant.
- No significant variation in cord blood TSH levels observed with respective of the gestational age
- No significant variation in Cord blood TSH levels between normally delivered and caesarean groups

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