# **Original Research Article**

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# Study of blood sugar levels in high risk neonates using glucometer method and laboratory glucose oxidase peroxidase method

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#### **ABSTRACT**

**Background:** The glucose oxidase method used for determining the blood glucose concentration is time consuming, requires more quantity of blood and rapid glycolysis of red blood cells glucose which mandates early estimation using faster techniques. A blood glucose method requiring one drop of blood and giving reliable results within a minute would help greatly in identifying and treating the infants who require extra carbohydrate. Glucometers have the advantage of being simpler, uses less quantity of blood, quick reading and can be handled even by unskilled personnel. Hence, there is a need to know the efficacy and accuracy of glucometer used in our set up.

Methods: About 150 infants admitted in NICU and high-risk neonates in post-natal ward in department of pediatrics at our Hospital from June 2014 to June 2015 were included in the study. The blood samples were collected immediately after birth or within 5 minutes of admission. Comparison of variables representing categorical data (between two markers) were estimated by Chi Square Test. Sensitivity, Specificity and Predictive Values of Glucometer were calculated by using formulas. All values will be reported based on two-sided and all the statistical tests were interpreted at 5% level of significance level (i.e. p <0.05).

Results: After the comparison between glucometer and laboratory method, results show that blood glucose estimation using capillary blood has an excellent pick-up rate of detecting neonatal hypoglycemia. More than 93% of the proven cases of hypoglycemia proved by lab method] were diagnosed when blood glucose estimation with glucometer was done. The overall pick-up rate of hypoglycemia by glucometer is very good compared to the laboratory method.

Conclusions: Blood glucose estimation using Glucometer method is an effective method in detecting neonatal hypoglycemia.

**Keywords:** Accuracy, Capillary blood, Glucose estimation, Neonatal hypoglycemia

#### INTRODUCTION

Hypoglycemia is one of the most common metabolic problems encountered in the newborns. The term "hypoglycemia" refers to a reduction in the glucose concentration of the circulating blood. It is almost 100 years since hypoglycemia was first described in children and over 50 years since it was recognized in newborns and infants. The operational threshold for hypoglycemia is defined as that concentration of plasma or whole blood glucose at which clinicians should consider intervention, based on the evidence currently available in literature.<sup>2</sup> This threshold is currently believed to be a blood glucose value of less than 40 mg/dL.<sup>3</sup>

Variable incidence has been reported by various authors in different weight and gestational age groups.4 The overall incidence of hypoglycemia in neonates varies from 0.2 to 11.4%.<sup>5,6</sup> However, in the presence of certain risk factors i.e. small for date, large for date, infants of diabetic mothers, prematurity etc., the probability of hypoglycemia increases many folds.<sup>5</sup>

Hypoglycemia in neonates can be symptomatic and asymptomatic.<sup>7-9</sup> The most common symptoms such as jitteriness, convulsions, apathy, hypotonia, coma, refusal to feed, cyanosis, high pitched cry, hypothermia are very nonspecific and especially in small sick infants, these symptoms may be easily missed. Therefore, hypoglycemia must always be confirmed biochemically and by response to treatment. Hypoglycemia is known to be associated with brain dysfunction and neuromotor developmental retardation in both symptomatic and asymptomatic cases.<sup>1,5,10</sup>

The methods for determining blood/plasma glucose concentration include reductiometric method, glucose oxidase method and hexokinase method.<sup>1,11</sup> The glucose oxidase method used in the laboratory for determining the blood glucose concentration is precise and specific for glucose.<sup>1</sup> As it is usually performed in the main laboratory, the results are not available quickly enough for timely appropriate management.

The development of reagent strip blood glucose tests in the 1970s facilitated the practice of screening for hypoglycemia in newborns. Currently used glucometers were initially developed for glucose monitoring in adult diabetics. These glucometers are often used for blood glucose estimation in NICU.

The glucose oxidase method used for determining the blood glucose concentration is usually performed in the main laboratory, this procedure is time consuming, requires more quantity of blood (~2ml) and rapid glycolysis of red blood cell glucose (in case of delay glucose levels in sample may drop at a rate of 18mg/dl/hr) mandates early estimation using faster techniques.

A blood glucose method requiring one drop of blood and giving reliable results within a minute would help greatly in identifying and treating the infants who require extra carbohydrate.

Low blood glucose in newborns is difficult to detect clinically. Hence a reliable "Point of Care" device (glucometer) for early detection and treatment of low glucose is needed.

Glucometers has the advantage of being simpler, uses less quantity of blood (~1-5microL), quick reading, can be handled even by unskilled personnel (used efficiently in primary, secondary and tertiary centers).

Hence there is a need to know the efficacy and accuracy of glucometer used in our set up.

#### **METHODS**

The present study was a hospital based cross-sectional study undertaken to study reliability and sensitivity of glucometer as sole mode of monitoring blood glucose in high risk newborns.

This study was carried out on in-patient of Pediatrics Department of our Hospital, after obtaining clearance from the hospital ethical and scientific committee.

The period of the study was from June 2014 to June 2015.

All Inborn neonates admitted in neonatal intensive care unit and high-risk neonates under observation in ward.

About 150 infants admitted in Neonatal Intensive Care Unit and high-risk neonates in post-natal ward in department of pediatrics at our Hospital from June 2014 to June 2015.

Patients fulfilling the eligibility criteria were included in this study.

#### Inclusion criteria

- High risk neonates admitted in NICU and post-natal ward
- Neonate born to mother with gestational diabetes mellitus
- Neonate with IUGR
- Preterm neonate with gestational age of >28 weeks and birth weight of >1000 grams
- Neonates of either sexes.

## Exclusion criteria

- Infants >28 days
- Full term neonates with no risk factors
- Infants with a gestational age of <28 weeks
- Infants with birth weight of  $\leq 1,000$  grams
- Infants with congenital diseases
- Infants whose mothers abused drugs during pregnancy
- Newborns with sepsis or septic shock, severe anemia with hematocrit (Hct)<20% or polycythemia with hematocrit (Hct)>65%.

#### Data collection technique and tools

Glucometer

Used is Bayer Breeze 2 blood glucometer.

Test principle

Bioamperometry-Glucose oxidase in the strip converts the glucose in the blood sample to gluconolactone.

This reaction creates a harmless electrical current that the glucometer interprets for that blood glucose.

#### Procedure

The sample was collected immediately after birth or within 5 minutes of admission. Wash hands thoroughly before the procedure; pre-warm the sole to ensure good perfusion before pricking. Use a sterile lancet to prick the child. Touch a drop of blood collected to the curve edge of the test strip. Blood will be drawn into the strip automatically. Do not place the blood drop on the top of the strip. Test result will appear in 5 second.

#### Laboratory method

Enzymatic calorimetric test, GOD-POD i.e. glucose oxidase- peroxidase method.

#### **Principle**

Glucose+O2-----Gluconic acid+H2O2

The first step is catalyzed by glucose oxidase and the second by peroxidase enzyme. The reagents and standard are ready to use.

### Procedure

The sample was collected immediately after birth or within 5 minutes of admission.

Mono-reagent----- 1000microlitre.

Sample ----- 10microlitre.

Mix well; incubate for 5 minutes at 370  $^{0}$ C. Read the absorbance  $\Delta A$  against reagent blank.

#### Data analysis

#### Statistical methods

Descriptive and inferential statistical analysis has been carried out in the present study statistical software.

In this study, all the analysis was performed by using 10.0 version of statistical software SPSS.

# Descriptive analysis

Continuous variables were summarized by using summary statistics (number of observations, mean and standard deviation).

Categorical values were summarized by using frequencies and percentages.

# Tests of significance

Comparison of variables representing categorical data that is Proportion of cases with hypoglycemia between two markers were estimated by chi square Test.

Association between weight and term with hypoglycemia was compared by Chi square test. Sensitivity, specificity and predictive positive and negative test of Glucometer were calculated by using formulas.

All values will be reported base on two-sided and all the statistical tests will be interpreted at 5% level of significance level.

#### **RESULTS**

Table 1: Profile of gravida among study cases.

Gravida	No. of cases (n=150)	%
Primigravida	72	48.0
Multigravida	78	52.0

The data reveals that 48.0% of the cases had primi gravida and 52.0% of the cases had multigravida.

Table 2: Types of delivery among study cases.

Types of delivery	No. of cases (n=150)	%
Normal	027	18.0
LSCS	115	76.7
Assisted	008	05.3

In this analysis, 18.0% of the cases had normal delivery followed by 76.7% of the cases had LSCS and 5.3% of the cases had assisted delivery.

Table 3: Distribution according to cause of admission.

No. of patients (n=150)	%
65	43.33
81	54
42	28
	No. of patients (n=150) 65 81 42

Among the Neonates admited, 65 neonates (43.33%) were admited due to infant of diabetic mother and 81 neonates (54%) were due to prematurity and 42 neonates (28%) were due to intrauterine growth retardation.

Table 4 reveals that 43.3% of the babies were Infant of diabetic mother and 56.7% of babies were born to non-diabetic mother.

Table 4: Profile of diabetes.

Category	No. of cases (n = 150)	%
Infant of diabetic mother (IDM)	65	43.3
Infant of non-diabetic mother (INDM)	85	56.7

Table 5: Profile of study group.

Variable	Mean	SD
Gestational age (weeks)	36.09	±3.34
Maternal age (years)	22.73	±4.18
Birth weight (kg)	2.53	±0.71
Blood glucose by glucometer (mg/dl)	62.87	±23.19
Blood glucose by laboratory method (mg/dl)	59.71	±25.73

Table 5 profile of study group shows that mean gestational age among neonates was  $36.09\pm3.34$  weeks; maternal age was  $22.73\pm4.18$  years; birth weight was  $2.53\pm0.71$  kg and blood glucose as  $62.87\pm23.19$  and  $59.71\pm25.73$  mg/dl by glucometer and Laboratory method respectively.

Table 6: Association between sex of baby and hypoglycemia.

Hypoglycemia (≤ 40)				
Sex	Gluce	ometer	Lab. Ox	idase method
	No.	%	No.	<b>%</b>
Male (n =81)	15	18.5	17	21.0
Female (n=69)	16	23.2	16	23.2

By chi - square test. p > 0.05, not significant

Table 6 reveals that, 18.5% of the male babies were hypoglycemic by glucometer method which was comparable to 21.0% by Lab. Oxidase method and the difference was not significant.

23.2% of the female babies each were hypoglycemic by glucometer and Lab. Oxidase method which was same and the difference was not significant.

Table 7: Association between weight of baby and hypoglycemia.

Hypoglycemia (≤ 40)				
Wajaht	Glu	cometer	Lab. oz	xidase method
Weight	No	%	No	%
<2.5 (N = 73)	19	26.0	20	27.4
>2.5 (N = 77)	12	15.6	13	16.9

By chi - square test. p >0.05, not significant

Table 7 reveals that, 26.0% of the babies with weight <2.5 were hypoglycemic by glucometer and Lab. Oxidase method which was same and the difference was not significant.

15.6% of the babies with weight >2.5 were hypoglycemic by glucometer method which was comparable to 16.9% by Lab. Oxidase method and the difference was not significant.

Table 8: Comparison of blood glucose levels among LBW and normal neonate.

Blood glucose	LBW (Mean±SD)	Normal (Mean±SD )	P value
By glucometer (mg/dl)	59.04±23.71	66.51±22.23	< 0.05
By laboratory method (mg/dl)	55.72±25.71	63.48±21.28	< 0.05

By chi - square test. p < 0.05, significant

The comparison of blood glucose levels among LBW and Normal neonate shows that mean blood glucose levels by Glucometer and laboratory method were statistically lower in LBW neonates as compared to normal weight neonates (p <0.05).

Table 9: Association between term/preterm and hypoglycemia.

Hypoglycemia (≤40)				
Term/Preterm	Glucometer		Lab. o	oxidase od
	No.	<b>%</b>	No.	%
Term $(n = 69)$	09	13.0	10	14.5
Preterm $(n = 81)$	22	27.2	23	28.4

By chi - square test. p >0.05, not significant

In this study, 13.0% of the term babies were hypoglycemic by glucometer method which was comparable to 14.5% by Lab. Oxidase method and the difference was not significant.

27.2% of the preterm babies each were hypoglycemic by glucometer and Lab. Oxidase method which was same and the difference was not-significant.

Table 10: Validity of glucometer.

Parameter	(%)
Sensitivity	96.8
Specificity	98.3
Positive predictive value (+ve)	93.9
Negative predictive Value (-ve)	99.1

Table 11: Correlation between glucometer and hypoglycemia.

Parameter	Mean $(\bar{x} \pm SD)$
Glucometer	62.87±23.19
Lab method	59.71±25.73
R	0.952

By pearson correlation coefficient. \*significant (p <0.05)

According to Table 11 there is a significant correlation between Glucometer and Laboratory method.

Table 12: Correlation between glucometer and lab method value (<40).

Parameter	Mean $(\bar{x} \pm SD)$
Glucometer (n=31)	31.31±04.45
Lab method (n=33)	27.73±07.92
R	0.979

By pearson correlation coefficient. \*significant (p < 0.05)

According to above Table 12 there is a significant correlation between Glucometer and Laboratory method.

Table 13: Correlation between glucometer and lab method value (>40).

Parameter	Mean $(\overline{x} \pm SD)$
Glucometer (n=119)	71.43±18.26
Lab method (n=117)	68.73±21.45
R	0.989

By pearson correlation coefficient. \*significant (p < 0.05)

According to Table 13 there is a significant correlation between Glucometer and Laboratory method.

Table 14: Correlation between glucometer and lab value with lab value >40mg% and <40mg% groups.

Methods	(n=150)	Pearson correlation	P value
Glucometer vs lab (<40mg %)	33	0.979	*0.001
Glucometer vs lab (>40mg %)	117	0.989	*0.001

By pearson correlation coefficient. \*significant (p < 0.05)

Table 15: Overall incidence of hypoglycemia among study cases.

Methods	(n=150)	%
Glucometer	31	20.5
Lab method	33	22.0

Table 15 reveals that, 20.5% of the cases were hypoglycemic by glucometer method which was comparable to 22.0% by Laboratory (Lab.) Oxidase method.

#### **DISCUSSION**

The present cross sectional study was undertaken to evaluate the efficacy of glucometers in estimating the blood glucose levels in newborns, in comparison with laboratory glucose oxidase method. The study was conducted during the period of June 2014 to June 2015 in Department of Pediatrics at our Hospital.

Main stress was laid on detection of hypoglycemia by both the methods and to know if glucometer is a good screening tool to detect hypoglycemia. Laboratory glucose oxidase method of blood glucose estimation was taken as gold standard.

In this study, 150 neonates who were admitted in NICU and postnatal ward with varied symptomatology were enrolled. This is due to the fact that majority of the clinical conditions associated with hypoglycemia such as birth asphyxia, prematurity, respiratory distress etc. manifest in the early neonatal period and seek admission.

The study was conducted after obtaining clearance from the ethical committee of the institute. The data collection was done by using predesigned pretested questionnaire. All the subjects included in the study volunteered after proper consent from the parents.

Among the Neonates admited, 65 neonates (43.33%) were admited due to infant of diabetic mother and 81 neonates (54%) were due to prematurity and 42 neonates (28%) were due to intrauterine growth retardation

Out of 150 neonates 72 (48%) were born to primigravida mothers, and 78 (52%) were born to multigravida mothers. 115 (76.7) babies born via LSCS, 27 (18%) born normal vaginal route and 8 (5.3%) babies born with assisted delivery. 65 (43.3%) infants born to diabetic mother [IDM (gestational or type 2)].

In the present study, profile of study group shows that mean gestational age among neonates was  $36.09\pm3.34$  weeks; maternal age was  $22.73\pm4.18$  years; birth weight was  $2.53\pm0.71$  kg and blood glucose as  $62.87\pm23.19$  and  $59.71\pm25.73$  mg/dl by glucometer and Laboratory method respectively.

Hypoglycemia was defined as blood glucose level less than 40mg% as defined by the study. 3,12

21% prevalence of hypoglycemia observed in male babies and 23.2% prevalence of hypoglycemia observed in female babies.

In the present study it reveals that, 18.5% of the male babies were hypoglycemic by glucometer method which was comparable to 21.0% by Lab. Oxidase method and the difference was not significant. 23.2% of the female babies each were hypoglycemic by glucometer and Lab. Oxidase method which was same and the difference was not significant

The prevalence of hypoglycemia observed in LBW neonates was 27.4%. The prevalence of hypoglycemia observed in normal birth weight neonates in this study was 16.9%. The higher prevalence of hypoglycemia observed in LBW neonate relative to normal weight neonates (27.4% versus 16.9%).

Our study reveals that, 26.0% of the babies with weight <2.5 were hypoglycemic by glucometer and Lab. Oxidase method which was same and the difference was not significant. 15.6% of the babies with weight >2.5 were hypoglycemic by glucometer method which was comparable to 16.9% by Lab. Oxidase method and the difference was not significant.

The prevalence of hypoglycemia observed in this study for full term neonates at our Hospital was 14.5%. The prevalence of hypoglycemia observed in preterm neonates in this study was 28.4%.

The comparison of blood glucose levels among LBW and Normal neonate shows that mean blood glucose levels by Glucometer and laboratory method were statistically lower in LBW neonates as compared to normal weight neonates. (p <0.05).

The higher prevalence of hypoglycemia observed in preterm neonates relative to full term neonates (28.4% versus 14.5%) could be attributed to lower energy reserves in hepatic and muscle glycogen and fats, higher insulin levels which matches the higher protein intake which is an insulinomic stimulus, less developed gluconeogenic pathways to synthesize glucose from glucogenic amino acids, high brain to body mass ratio in small for gestational age neonates resulting in high glucose consumption, reduced fat stores and failure of counter regulatory hormones in low birth weight neonates. <sup>13-16</sup>

The findings of present study were in contrast to study done by Harish J et al who enrolled 250 neonates of which 63.2% were males and 36.8% were females.

103 cases (41.2%) were found to be hypoglycemic by lab values while 52 (20.8%) were found to be hypoglycemic by glucometer.<sup>17</sup>

A total of 150 samples were taken from neonates, Hypoglycemia was detected in 33 samples (22%).

The instrument used showed a sensitivity of 96.8% and specificity of 98.3% to detect neonatal hypoglycemia (< 40 mg/dl) with a positive predictive value of 93.9% and negative predictive value 99.1%. 22% were found to be hypoglycemic by laboratory glucose oxidase method whereas by glucometer 20.50% were found to be hypoglycemic.

Thus, the incidence of hypoglycemia as detected by the gold standard method i.e. laboratory glucose oxidase method in our study is 22 %, which was higher than Singhal PK study (4.8%) and Mishra PK study (4.8%).<sup>5,4</sup>

This wide variation in the incidence of hypoglycemia could be attributed to the lack of uniform definition of hypoglycemia, varied sample size and risk factors.

Table 16: Comparison of overall incidence of hypoglycemia with other studies.

Studies	Incidence
Singhal PK et al	4.8%
Mishra PK et al	9.7%
Our study	22%

In the Singhal PK et al study hypoglycemia was defined as blood glucose level <30 mg% while in the Mishra PK et al and study it was taken as 20mg%.<sup>5,4</sup>

In our study, the cut-off value for hypoglycemia was taken as 40mg%.

In our study, the results depict the good pick-up rate of hypoglycemia by glucometer in comparison with the laboratory values.

It shows that nearly all of the cases of proven hypoglycemia (by laboratory method) were detected by glucometer, indicating that glucometers are good screening tool for detecting hypoglycemia in newborns.

Table 17: Comparison of overall incidence of hypoglycemia with other studies.

Studies	Sensitivity	Specificity
Dahlberg et al	100%	84%
Mehta et al	86%	89%
HO HT et al	92.3%	91%
Hamid MH et al	98%	93%
Our Study	96.8%	98.30%

The present study shows that glucometer have a sensitivity of 96.8% to detect hypoglycemia and specificity of 98.3%.

The findings of the present study were in accordance with study done by Dahlberg et al where the glucometer had sensitivity of 100%.<sup>18</sup>

Similarly, findings were seen in study by Hamid MH et al where glucometer had sensitivity of 98% and specificity of 93%. 12

The findings were in contrast to study done by Mehta et al where the glucometer had sensitivity of 86% and specificity of 89%.<sup>19</sup> Our study shows that glucometer have a sensitivity of 96.80% to detect hypoglycemia which is in comparable with Ho HT et al (92.3%).<sup>20</sup> Our study also showed that glucometers had a positive predictive value of 93.9% and a negative predictive value of 99.1%, which is high when correlate with the results obtained by Ho HT et al and Dahlberg et al.<sup>20,18</sup>

The overall accuracy of glucometers to detect hypoglycemia in newborns in comparison with the gold standard i.e. laboratory value is good (93%).

Table 18: Correlation between glucometer and laboratory oxidase method.

Studies	Correlation coefficient
Hamid MH et al	0.976
Harish JP et al	0.771
Our study	0.952

Our study shows a strong correlation between glucometer and laboratory values when all blood glucose values were considered irrespective of hypoglycemia. The Mean  $(\overline{x}\pm SD)$  of glucometer was  $62.87\pm23.19$  and of laboratory value was  $59.71\pm25.73$  and coefficient of correlation was r=0.952 (p-value <0.001). There is a strong correlation between Glucometer and Lab method for value (<40).

The Mean ( $\overline{x} \pm SD$ ) of glucometer was 31.31±04.45 and of laboratory value was 27.73±07.92 and coefficient of correlation was r=0.979 (p-value <0.001). There is a strong correlation between Glucometer and Lab method for value (>40). The Mean ( $\overline{x} \pm SD$ ) of glucometer was 71.43±18.26 and of laboratory value was 68.73±21.45 and coefficient of correlation was r=0.989.

Our study showed a strong correlation between glucometer and laboratory values when all blood glucose values were considered irrespective of hypoglycemia. That is r=0.952.

The finding of present study was in accordance with Hamid M H et al study (0.976). 12

Correlations of glucometer values with laboratory values of blood glucose levels were strong throughout the range when the gold standard value is >40mg% coefficient of correlation (r) was 0.989 (p-value < 0.001). And when the value is <40mg%, there was also strong correlation throughout the range with coefficient of correlation (r) 0.979 (p-value <0.001). This further stress the excellent efficacy of glucometers in detecting hypoglycemia in newborns.

#### CONCLUSION

Overall, after the comparison between glucometer and laboratory method, results show that blood glucose estimation using capillary blood has an excellent pick-up rate of detecting neonatal hypoglycemia. More than 93% of the proven cases of hypoglycemia (proved by lab method) were diagnosed when blood glucose estimation with glucometer was done. The overall pick-up rate of hypoglycemia by glucometer is very good compared to the laboratory method.

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