## Original Research Article

# Comparison of haemoglobin assessment by HemoCue 301 and automated haematology analyser using flowcytometry among school going children: a one year study at a tertiary care hospital

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### ABSTRACT

**Background:** Anaemia defined as reduction in the concentration in Haemoglobin is one of the key health indicators of health care system of the country. Accurate screening methods are required to estimate the levels of haemoglobin for diagnosing the cause of anaemia. Objectives of the study was to analyze and compare the results of haemoglobin concentrations estimated with automated haematology analyzer and point of care device HemoCue Hb301.
**Methods:** It is a prospective cross-sectional study was conducted for one year after ethical approval. Non fasting capillary and venous blood samples were collected from the selected cases of children and Haemoglobin concentrations were estimated by automated analyzer and HemoCue Hb301 system and the values were noted. Quality control checks were performed for both. Statistical analysis was done using IBM SPSS Version 24.0.
**Results:** Mean Hb% concentration was estimated in 108 children with 44 female and 64 males. The mean value of Automated hematology analyzer (11.965±1.012) was significantly higher when compared with the mean value of HemoCue Hb301 (11.697±1.312) (p=0.002). There was a significantly strong correlation between HemoCue Hb301and Automated hematology analyzer (r-value = 0.732, p <0.0001).
**Conclusions:** The HemoCue is useful in many different settings and remains a widely used method in field settings as it has several advantages and is relatively inexpensive compared with automated haematology analysers. Further studies are needed to better understand potential sources of error in the Hb assessment by HemoCue with the aim to better train phlebotomists and implement appropriate standardised procedures.

**Keywords:** Anemia, Automated hematology analyzer, HemoCue Hb301, Hemoglobin

## INTRODUCTION

Anaemia defined as reduction in the concentration in Haemoglobin is one of the key health indicator of health care system of the country. This clinical condition is a parameter for poor nutrition and poor health. Global prevalence of anaemia among school going children as per WHO data is 25.4%and most of the cases are observed in developing countries.[1] Effects of anaemia in children particularly among the school going are associated with impaired neuro-cognitive function and exercise intolerance and the effects continue even after successful treatment. Hence it is necessary to determine the cause of anaemia and its management at an early stage to avoid any detrimental effects. The most common cause of anaemia is under nutrition among school going children. In Indian children, high prevalence of anaemia varying from 27% to 90% has been reported in different studies. Iron deficiency due to malnutrition is claimed as one of the most common cause. Hence accurate screening

methods are required to estimate the levels of haemoglobin for diagnosing the cause of anaemia in school going children.[2] In well-resourced countries, automated hematology analyzers are used for estimation of HB concentration. This method is accurate and reliable; it requires well-equipped laboratory facilities and trained personnel, which are often unavailable in resource-poor settings, especially in rural areas. Hence in settings where automated hematology analyzers are unavailable or cannot be placed (Community surveys, small health clinics etc.) point of care devices like Haemoglobinometer (HemoCue Hb301) are essential in estimation of Hb concentrations among school children.[3] These devices are economical, provides quick easy access to lab quality results. However, the reliability of the results depends on quality of blood samples. Hence it is always essential to understand the principles and effects of different collection techniques and instruments in data analysis. Inappropriate or inaccurate point of care haemoglobin meters may result in misdiagnose of anaemia, which in turn may lead to false exclusion from, and inclusion in, clinical studies and result in compromised clinical care.[4]

Hence in the present study we analyzed the results of haemoglobin concentrations estimated with automated hematology analyzer and point of care device HemoCue Hb301. The principles of both the devices were different and the study would help in testing the accuracy of the device in estimation of Hb concentrations at community settings where automated analyzer setup is not possible.

## METHODS

The present cross-sectional observational study was conducted in Department of paediatrics of Narayana Medical College and Hospital, a tertiary care hospital of south India. The study was conducted for a period of twelve months from January 2018 to December 2018 after the ethical committee approval. The guidelines of the committee were clearly followed throughout the study period.

The study objective was to compare the haemoglobin concentrations measured by "Hemocue globinometer Hb 301 System" with the readings of automated hematology analyzer (Sysmex XN-550) which uses flow cytometric principle in calculating the Haemoglobin value among the school going children.

All the school going children between 6-15 years of age residing in tribal welfare schools and hostels were included in the study. The study was clearly explained, and informed consent was obtained from all the Parents/ Guardians of the children who participated in the study. The socio demographic data including age, sex, and history of any illness in the past, history of medications etc. was noted and entered into Microsoft excel spread sheet for corrections before analysis.

Non fasting capillary and venous blood samples were collected from the selected cases of children by trained phlebotomists with the child in seating position. In capillary blood, the tip of the left middle finger was pricked with a lancet and first drop of blood was wiped with cotton. A standard Hb 301 microcuvette was used to collect the blood and immediately inserted in the HemoCue Hb 301 device for analysis. The Hb concentration was determined by measuring the absorbance of whole blood at an Hb/HbO2 isosbestic point. The results are displayed numerically in g/dL at the time of the measurement and noted manually in the data sheet.

Venous blood about 3ml was collected from the ante cubital vein and transferred to EDTA containing vacutainers by following standard guidelines. The tubes were gently inverted 4-5 times and transferred into a cold box maintained at 4-8ºC. The samples were transferred to the laboratory on the same day and processed. The Automated haematology analyser (Sysmex XN-550) was used and the complete blood picture of the specimen was performed and Haemoglobin concentration was noted.

The quality control checks for HemoCue Hb 301 system was performed weekly by using Eurotrol Hb301 Control. For Sysmex XN-550 automated hematology analyser Quality checks were performed daily as per manufacture instructions

### *Statistical analysis*

Data has been entered into MS-Excel and statistical analysis was done using IBM SPSS Version 24.0. For continuous variables, the data value is expressed as Mean±SD (Standard Deviation). To test the mean difference between two groups, student's t-test (Paired sample t-test) was used and to test linear relationship between the groups, Pearson's correlation coefficient (r-value) was used. To estimate the bias and limits of agreement between the hemoCueHb301 and flow cytometry, Bland-Altman plot was used. All the p values having less than 0.05 were considered as statistically significant.

## RESULTS

A total of 108 children with 64 male (59.26%) and 44 female children (40.74%) were included in the study. 53.7% of the children were in the age range of 6-10 years and 46.3% in group of 11-15 years. The mean age of the study group was 7.12±1.5 years. The mean Hb% concentration by HemoCue Hb 301was 11.697 (±1.312 Standard deviation) and with Sysmex XN-550 automated hematology analyzer was 11.965 (±1.012 standard deviation) (Table 1).

The mean value of Sysmex XN-550 automated hematology analyzer (11.965±1.012) was significantly higher when compared with the mean value of HemoCue

Hb301 (11.697±1.312) [p=0.002]. There was a significantly strong correlation between HemoCue Hb301and Sysmex XN-550 analyzer (r-value = 0.732, p <0.0001) which is shown in (Table 1, Figure 1 and 2).

**Table 1: Comparison of mean values between HemoCue Hb301 and Automated hematology analyzer methods.**

|  | N | Mean | Std. Deviation | Mean Difference | t-value | p value | r-value (p-value) |
|---|---|---|---|---|---|---|---|
| HemoCue Hb301 | 108 | 11.697 | 1.312 | -0.26 | -3.108 | 0.002* | 0.732 (<0.0001**) |
| Automated analyzer | 108 | 11.965 | 1.012 | | | | |

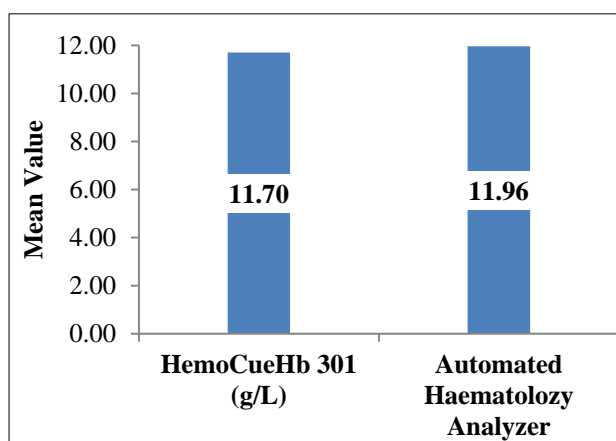*p<0.05 - Significant, **p<0.0001 - Very High Significant (VHS)



**Figure 1: Comparison of mean values between HemoCue Hb301 and automated hematology analyzer methods.**
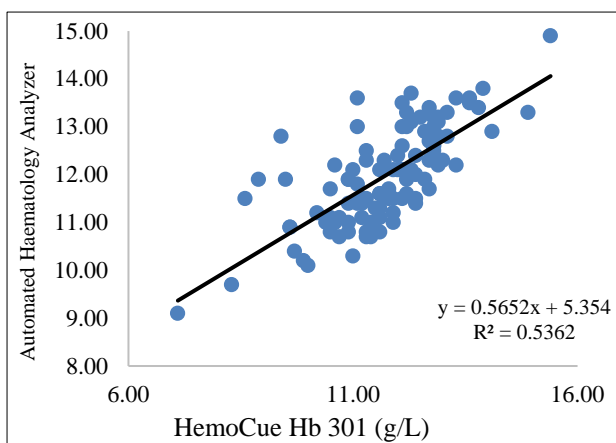


**Figure 2: Scatter plot for testing the correlation between HemoCueHb 301 (g/L) and automated hematology analyzer.**

Based on the Bland-Altman plot, the assessment of agreement was good between the HemoCue Hb301 and Automated hematology analyzer with a bias with 95% limits of agreement was -0.3 (-2.0 to 1.5) g/L and the within-instrument variation (coefficient of variation (CV)) for HemoCueHb301and Sysmex XN-550 analyzer was 11.21% and 8.46% respectively (Figure 3).

## DISCUSSION

Anaemia is a correctable clinical condition and a major health problem in developing countries. Accurate diagnosis and preventive measures and a correct health policy measures are essential in prevention of anaemia and related morbidity and mortality. Studies have documented that rapid physical and physiological development makes school-age children more vulnerable for anaemia, especially for Iron Deficiency anaemia.[5] Consequences of anaemia in school children are poor psychomotor development, poor IQ, poor school performance, reduced work capacity and poor quality of life. Socio economic factors including poverty, poor sanitation, community factors, parent's level of education and income also play an important role in development of anaemia among school children.

Estimation of Haemoglobin concentration among the school children is one of the main screening methods in diagnosing anaemia. However, the cause may be further evaluated by various other parameters like serum ferritin estimation, iron binding capacity etc. To prevent false positive cases of anaemia, an accurate and reliable method with high sensitivity and specificity is required at a hospital setting and at community level where facilities are poor and meagre in establishing a high-end laboratory setup. In this context, availability of point of care devices like HemoCue Hb301 system and their validity and accuracy in determining the haemoglobin concentration need to be evaluated which was done in the present study by comparing with automated analyzer.

The haemoglobin concentrations of 108 children have been evaluated whose mean age was 7.12±1.5 years. The hemoglobin concentration of these study participants was estimated by automated hematology analyzer and HemoCue Hb 301 point of care device. The mean Hb% concentration by HemoCue Hb 301was 11.697 (±1.312 Standard deviation) and with automated analyzer was 11.965 (±1.012 standard deviation). Finding of our study revealed that the mean Hb obtained by analyzer was higher than that obtained by HemoCue 301 device in our age group of participants. Findings of our study concurred with the findings of Hinnouho et al, who

reported that Hemocue 301 system was less sensitive and detected lesser concentrations when compared with analyzer.[6] There was a significantly strong correlation between HemoCue Hb301 and automated analyzer (r-value = 0.732, p <0.0001). However, the difference was small but clinically significant. However direct comparison of our findings with reports of previous studies was limited by several factors like, using different point of care devices; some studies have used Hemocue Hb201 system which employs different biochemical methods for estimation of Hb concentrations. Other influencing factors in comparing the study results was age of the study group, ethnicity and whether Hb was assessed on fasting or non-fasting blood samples. In a study of Paiva AA et al, venous Hb levels by automated hematology analyzer {were consistently higher than capillary levels by HemoCue when the Hb concentrations were in the lower part of the normal non-anaemic range.[7] This is in contrast to our study, which found that HemoCue Hb301 overestimated capillary Hb levels at lower Hb concentrations, resulting in a lower prevalence of anaemia. Hence our study clearly highlights that HemoCue Hb 301 point of care device estimated capillary blood levels of Hb and provided comparable results with automated analyzer. It has been suggested that the HemoCue device may be more accurate in Hb concentration determination than automated analysers given that the latter requires a sample dilution, while HemoCue assesses Hb directly and is not affected by changes in turbidity. However, in the study of Chen PP et al, and Rappaport AI et al, they reported within-subject variability of capillary blood haemoglobin values that might explain the unreliability of the method, and it has been shown that two capillary samples taken from different fingers of the same subjects had haemoglobin concentrations differing by/ more than two g/dL using the HemoCue.[8,9] Rechner IJ et al, and Adam et al, investigating a group of forty (40) health adults, observed a wide variation between paired Hb measurements determined in capillary samples by the HemoCue®. The differences were higher than 10.0 g/L (1.0 mg/dL), comparing the measurements conducted by health visitors with the ones by biomedical scientists, who received identical training on the use of the HemoCue.[10,11]

## CONCLUSION

To conclude, this study is first of the study to compare the Haemoglobin concentrations in school going children with HemoCue 301 system which is a point of care device and an automated analyzer used in laboratory setup. The HemoCue is useful in many different settings and remains a widely used method in field settings as it has several advantages and is relatively inexpensive compared with automated hematology analyzers. Further studies are needed to better understand potential sources of error in the Hb assessment by HemoCue with the aim to better train phlebotomists and implement appropriate standardized procedures. The Hemocue 301 showed poor agreement compared with automated analyzer in haemoglobin estimation.

## REFERENCES

1. Bruno de Benoist, Erin McLean, Ines Egli and Mary Cogswell. Worldwide prevalence of anaemia 1993-2005. WHO Global Database on Anaemia Geneva, World Health Organization, 2008. Available at: https://www.who.int/nutrition/publications/micronutrients/anaemia_iron_deficiency/9789241596657. Accessed on 14 November 2019.
2. McLean E, Cogswell M, Egli I, Wojdyla D, De Benoist B. Worldwide prevalence of anaemia, WHO vitamin and mineral nutrition information system, 1993-2005. Public Health Nutr. 2009 Apr;12(4):444-54.
3. Rippmann CE, Nett PC, Popovic D, Seifert B, Pasch T, Spahn DR. HemoCue®, an accurate bedside method of hemoglobin measurement?. J Clini Monitor. 1997 Nov 1;13(6):373-7.
4. Sari M, Pee SD, Martini E, Herman S, Bloem MW, Yip R. Estimating the prevalence of anaemia: a comparison of three methods. Bulletin World Health Org. 2001;79:506-11.
5. Tesfaye M, Yemane T, Adisu W, Asres Y, Gedefaw L. Anemia and iron deficiency among school adolescents: burden, severity, and determinant factors in southwest Ethiopia. Adolescent Health, Med Therap. 2015;6:189-96.
6. Hinnouho GM, Barffour MA, Wessells KR, Brown KH, Kounnavong S, Chanhthavong B, et al. Comparison of haemoglobin assessments by HemoCue and two automated haematology analysers in young Laotian children. J Clini Pathol. 2018 Jun 1;71(6):532-8.
7. Paiva AD, Rondó PH, Silva SS, Latorre MD. Comparison between the HemoCue® and an automated counter for measuring hemoglobin. Revista Saude Publica. 2004;38:585-7.
8. Chen PP, Short TG, Leung DH, Oh TE. A clinical evaluation of the Hemocue haemoglobinometer using capillary, venous and arterial samples. Anaesthesia Inten Care. 1992 Nov;20(4):497-500.
9. Rappaport AI, Barr SI, Green TJ, Karakochuk CD. Variation in haemoglobin measurement across different HemoCue devices and device operators in rural Cambodia. J Clini Pathol. 2017 Jul 1;70(7):615-8.
10. Rechner IJ, Twigg A, Davies AF, Imong S. Evaluation of the HemoCue compared with the Coulter STKS for measurement of neonatal haemoglobin. Arch Dis Childhood-Fetal Neonatal Edition. 2002 May 1;86(3):F188-9.

11. Adam I, Ahmed S, Mahmoud MH, Yassin MI. Comparison of HemoCue® hemoglobin-meter and automated hematology analyzer in measurement of hemoglobin levels in pregnant women at Khartoum hospital, Sudan. Diagnostic Pathol. 2012 Dec;7(1):30.