

# Digital Hemoglobinometers as Point-of-Care Testing Devices for Hemoglobin Estimation: A Validation Study from India

Kapil Yadav, Shashi Kant, Gomathi Ramaswamy, Farhad Ahamed<sup>1</sup>, Kashish Vohra

National Centre of Excellence and Advanced Research on Anemia Control, Centre for Community Medicine, All India Institute of Medical Sciences, New Delhi, <sup>1</sup>Centre for Community and Family Medicine, All India Institute of Medical Sciences, Kalyani, West Bengal, India

## Abstract

**Introduction:** India has launched Anemia Mukht Bharat (AMB) strategy in 2018, to deal with the high burden of anemia in the country. Point-of-care testing (POCT) of anemia using digital hemoglobinometers and treatment is one of the primary interventions under AMB. This study aimed to determine the diagnostic validity of digital hemoglobinometers (TrueHb and HemoCue 301) for screening of anemia compared to hematology analyzer. **Methodology:** A hospital-based, cross-sectional study was conducted among pregnant women attending antenatal clinics of a selected primary health center and subdivisional hospital of Haryana, India, during January 2019. Hemoglobin (Hb) levels of the pregnant women were estimated in digital hemoglobinometers using capillary blood samples and hematology analyzer using venous blood samples. Bias, limits of agreement (LOA), and validity of digital hemoglobinometers were assessed against a hematology analyzer. **Results:** A total of 110 pregnant women were included. Bias (LOA) in Hb values estimated using digital hemoglobinometers was  $-0.09$  g/dL ( $-1.97$  to  $1.80$ ) for HemoCue301 and  $-0.04$  g/dL ( $-1.69$  to  $1.60$ ) for TrueHb compared to the hematology analyzer. HemoCue<sup>®</sup> 301 (sensitivity: 86% and specificity: 83%) had relatively higher sensitivity and specificity compared to TrueHb (sensitivity: 78.9% and specificity: 81%). **Conclusions:** Digital hemoglobinometers have high sensitivity and specificity. Thus, these can be a potential POCTs for screening of anemia in peripheral health facilities. Further studies are required to establish the validity of the digital hemoglobinometers at community settings.

**Keywords:** HemoCue, hemoglobin, pregnant women, TrueHb, validation

## INTRODUCTION

Anemia is a severe public health problem in India, with more than 40% of the population being anemic. To combat the high burden of anemia in the country, Anemia Mukht Bharat (AMB) strategy was launched in April 2018 by the Ministry of Health and Family Welfare, Government of India.<sup>[1]</sup> AMB has set a target of 3% reduction in the burden of anemia per year from 2018 to 2022. Point-of-care testing (POCT) for hemoglobin (Hb) using digital hemoglobinometers and treatment for anemia is one of the flagship interventions under this strategy. POCT provides Hb results instantly and indeed aids in appropriate and timely management of anemia. POCT also has other advantages such as requirement of less blood sample (one drop of capillary blood sample); usage at primary health-care and community settings; and nonrequirement of sophisticated laboratory or trained laboratory technicians.

A wide range of techniques and devices are available for Hb estimation. Of them, cyanmethemoglobin method is considered the gold standard and reference method for Hb estimation. However, the high complexity involved in the procedure restricts cyanmethemoglobin method for routine use in laboratories or as POCT.<sup>[2]</sup> The World Health Organization (WHO) and Indian Council of Medical Research (ICMR) recommends the use of hematology analyzer at secondary and tertiary care centers and digital hemoglobinometers at primary health centers (where

**Address for correspondence:** Dr. Kapil Yadav, Room No. 33, Centre for Community Medicine, All India Institute of Medical Sciences, New Delhi, India. E-mail: dr.kapilyadav@gmail.com

This is an open access journal, and articles are distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 4.0 License, which allows others to remix, tweak, and build upon the work non-commercially, as long as appropriate credit is given and the new creations are licensed under the identical terms.

**For reprints contact:** reprints@medknow.com

**How to cite this article:** Yadav K, Kant S, Ramaswamy G, Ahamed F, Vohra K. Digital hemoglobinometers as point-of-care testing devices for hemoglobin estimation: A validation study from India. Indian J Community Med 2020;45:506-10.

**Received:** 31-12-19, **Accepted:** 29-08-20, **Published:** 28-10-20

### Access this article online

#### Quick Response Code:



**Website:**  
www.ijcm.org.in

**DOI:**  
10.4103/ijcm.IJCM\_558\_19

hematology analyzers are not available) and community settings. Although hematology analyzers require skilled laboratory technician, space, optimum temperature, electricity, high cost, and maintenance, they are equipped with robust internal and external quality control mechanism for estimation of Hb with minimal errors.<sup>[3]</sup> Regardless, in rural areas with insufficient infrastructure or trained workforce, POCTs are the feasible alternatives and also inevitable. However, the total quality assurance and standardization for the accurate and reliable measurement of Hb cannot be compromised for the ease of use with POCT.

POCTs with poor diagnostic accuracy or complicated procedure will lead to erroneous estimation of Hb level and eventually leads to incorrect management of anemia, affecting both the individual and the health system. The National Iron Plus Initiative (2013) recommended the use of Sahli's method as POCT. Inter-observer variability due to subjectivity in hemoglobin estimation, need for accurate manual pipetting of blood sample, chances of colour fading in the comparator might lead to poor sensitivity of hemoglobin levels estimated in Sahli's hemoglobinometer.<sup>[4,5]</sup> Digital hemoglobinometers have the following advantages compared to other POCTs: (i) no or minimal subjectivity in the estimation of Hb; (ii) rapid turnover time (within few seconds); (iii) portable and operates in a wide range of temperatures; (iv) Hb values with one decimal place will be displayed in the device monitor; (v) availability of control solution at low, normal, and high ranges; (vi) recommended by the WHO and ICMR (India) for Hb estimation at primary health-care and community settings; and (vii) features to enable tech health intervention and real-time data capture.

A wide variety of digital hemoglobinometers are available for the estimation of Hb. Identification of a device with high validity and operational feasibility for usage at the primary care or community settings is vital for testing for anemia and for further adoption in the national program. In this context, this study was conducted to assess the diagnostic validity of two digital hemoglobinometers for Hb estimation compared to hematology autoanalyzer.

## METHODOLOGY

### Subjects

A hospital-based, cross-sectional study was conducted in January 2019 at a primary health center (PHC) and a subdistrict hospital (SDH) in Haryana, India. The mean temperature and relative humidity in the study area during data collection period were 21°C and 56%, respectively. Pregnant women attending antenatal clinics of these centers were recruited for the study. Pregnant women in all trimesters of pregnancy were included for the study. Similarly, there was no restriction for gravid index of the pregnant women for inclusion in the study. The average footfall of pregnant women at the antenatal clinic was 50–60 in PHC and 100–200 in SDH. Informed written consent was obtained from all the study participants. Pregnant women with known hemoglobinopathies and chronic illnesses such as renal failure, liver failure, and cardiac diseases were excluded from the study.

### Sample size and sampling method

Considering bias (standard deviation [SD] of difference) as 0.87 (0.27), alpha error of 5%, power of 80%, and maximum allowed difference of 1.54, the minimum required sample size was 102.<sup>[6,7]</sup> A total of 110 pregnant women were recruited from the two selected sites considering 10% nonresponse rate. The pregnant women were recruited using convenient sampling till the sample size is reached and all eligible pregnant women were enrolled.

### Ethical approval

The study protocol was approved by the Institutional Ethics Committee of All India Institute of Medical Sciences, New Delhi, India.

### Methods

#### Description of devices

- TrueHb Hemometer (Wrig Nanosystems, New Delhi, India)<sup>[8]</sup> – Device A: It is a battery-operated system which works based on the principle of optical reflectance photometry. A drop of whole blood sample will be diffused over the hydrophilic mesh of the thin plastic strip which is loaded with reagents. A reaction complex will be formed between the blood sample and the reagents, and

**Table 1: Prevalence of anemia and mean (standard deviation) of hemoglobin determined by the Device A, Device B, and hematology analyzer among the pregnant women in North India (n=110)**

| Hemoglobin concentration, g/dL | Hematology analyzer (n=110) | Device A        |                                      | Device B        |                                      |
|--------------------------------|-----------------------------|-----------------|--------------------------------------|-----------------|--------------------------------------|
|                                |                             | Overall (n=110) | P (device A vs. hematology analyzer) | Overall (n=110) | P (device B vs. hematology analyzer) |
| Mean±SD                        | 10.9±1.6                    | 10.9±1.8        | 0.612                                | 10.8±1.8        | 0.334                                |
| Range                          | 5.8-15.1                    | 5.7-16.0        |                                      | 6.3-15.9        |                                      |
| Prevalence of anemia, n (%)    | 53 (48.2)                   | 55 (50.0)       | 0.787                                | 52 (47.3)       | 0.893                                |
| No anemia, n (%)               | 57 (51.8)                   | 55 (50.0)       | 0.863                                | 58 (52.7)       | 0.687                                |
| Mild anemia, n (%)             | 29 (26.4)                   | 28 (25.4)       |                                      | 26 (23.6)       |                                      |
| Moderate anemia, n (%)         | 22 (20.0)                   | 23 (20.9)       |                                      | 21 (19.1)       |                                      |
| Severe anemia, n (%)           | 2 (1.8)                     | 4 (3.6)         |                                      | 5 (4.5)         |                                      |

SD: Standard deviation

- Hb will be estimated by optical reflectance photometry technique
- ii. HemoCue® Hb 301 System (HemoCue AB, Angelholm, Sweden) – Device B:<sup>[9]</sup> The HemoCue 301 is a battery-operated system which estimates Hb by measuring the absorbance of whole blood at the Hb/HbO<sub>2</sub> isosbestic points at the wavelength of 506 nm and 880 nm for compensation of turbidity
  - iii. Gold standard (hematology analyzer)
  - iv. Hematology analyzer (Sysmex XS 1000i, Kobe, Japan): This system is an automated blood cell counter that utilizes noncyanide method (sodium lauryl sulfate method) for the measurement of Hb.

### Data collection

Data on sociodemographic and obstetric details were collected using a pretested semi-structured interview schedule. A trained laboratory technician collected the capillary and venous blood samples from all the pregnant women at both the sites.

Second and third drops of capillary blood were taken for the estimation of Hb in Device A and Device B, alternatively. The sequence of digital hemoglobinometers (Device A and B) was alternated to overcome any bias that may arise due to the difference in the second and third drops of blood.

Similarly, 2 mL of venous blood sample was drawn from the antecubital fossa for the estimation of Hb in an autoanalyzer. The labeled venous samples from PHC were kept in a cold box at 2°–8°C and transported to the laboratory at SDH, and Hb values were estimated within 4 h of collection.

### Quality control

All the three levels of internal quality control (IQC) samples (normal, low, and high) were checked on a daily basis and plotted in Levy Jennings plot for both digital hemoglobinometers and Sysmex autoanalyzer. All values were found to be within the 2 SD range without any pattern suggestive of IQC error.

### Statistical analysis

The mean of difference (SD of difference) between the digital hemoglobinometers and hematology analyzer and limits of agreement (LOA) were calculated based on Bland–Atman plot. Lin's concordance correlation coefficient (CCC) and intraclass correlation coefficient were calculated with 95% confidence interval (CI). Receiver operating characteristic curve was plotted for both digital hemoglobinometers, to compare the area under the curve (AUC) for diagnosis of anemia. Weighted kappa was calculated to assess the agreement of digital hemoglobinometer for different grades of anemia compared to hematology analyzer.<sup>[10]</sup> Passing-Bablok regression was done to check the linear modal validity (MedCalc Software, Trail version 19.5.1, Ostend, Belgium). Sensitivity, specificity, and positive and negative predictive values were calculated to assess the validity of the digital hemoglobinometers. Data were entered in Microsoft Excel (version 2013) and analyzed using Stata 12 software (Stata Corp LP. 2011, College Station, TX, USA).

## RESULTS

In total, 110 pregnant women were included in this study. The mean age of the pregnant women was 23.3 ± 3.4 years. The overall prevalence of anemia was 48.2%; 95% CI: 38.9%–57.6% (mild anemia –26.4%, moderate anemia –20.0%, and severe anemia –1.8%) using the hematology analyzer [Table 1]. Prevalence of anemia using Device A was 50% (95% CI: 40.6%–59.4%) and Device B was 47.3% (95% CI: 38.0%–56.7%). There was no significant difference in the prevalence of anemia between Device A ( $P=0.787$ ) and Device B ( $P=0.893$ ) compared to hematology analyzer. Similar finding was observed in the subgroup analysis on the prevalence of anemia compared between devices based on the sequence of drop with hematology analyzer.

### Bland–Altman and Passing-Bablok regression analysis

The mean (SD) of Hb (g/dL) values using Device A was 10.9 (0.2), Device B was 10.8 (0.2), and hematology analyzer was 10.9 (0.1) ( $P=0.903$ ). Compared to hematology analyzer, bias and LOA for Device A were –0.04 g/dL (–1.69 to 1.60) and Device B was –0.09 g/dL (–1.97 to 1.80) [Figure 1 and Table 2]. Figure 2 depicts the linear model regression equation for Hb estimation using Passing-Bablok regression analysis. The intercept for device A was –1.34 with a slope of 1.11 (95% CI: 1.00 to 1.22), which reports no significant deviation from linearity compared to autoanalyzer ( $P=0.740$ ). Similarly, the intercept for Device B was –1.38 with slope 1.12 (95% CI: 1.00 to 1.25), which also shows linearity with autoanalyzer ( $P=0.970$ ).

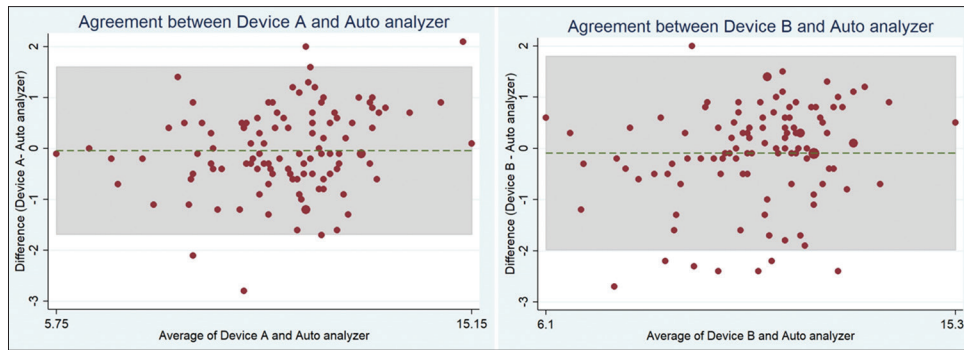
### Lin's concordance and agreement between digital hemoglobinometers and autoanalyzer

Lin's concordance coefficient for Device A (Lin's CCC = 0.88;  $P < 0.001$ ) and Device B (Lin's CCC = 0.84;  $P < 0.001$ ) was almost similar. The AUC for Device A was 0.90 and Device B was and 0.89 ( $P = 0.707$ ) [Figure 3]. The weighted Cohen's Kappa coefficient for different grades (no anemia/mild/moderate/severe anemia) for Device A was 0.67 and Device B was 0.70, indicating good degree of agreement compared to the hematology analyzer.

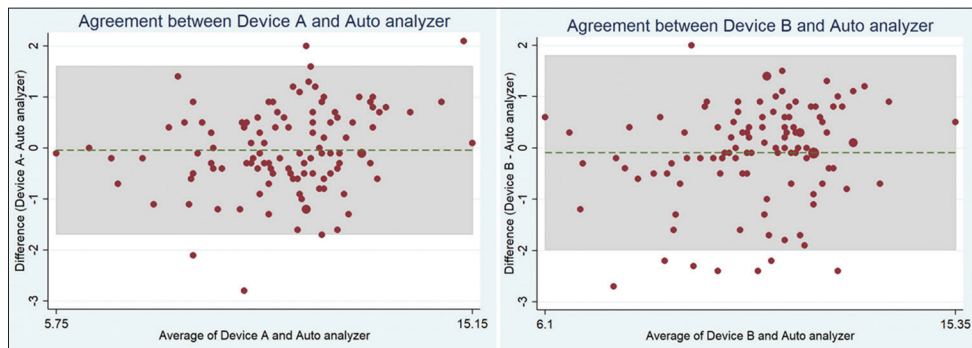
**Table 2: Bias (standard deviation) of difference, limits of agreement, and concordance determined by Device A, Device B, and hematology analyzer among the pregnant women in North India (n=110)**

|                               | Device A<br>(n=110) | Device B<br>(n=110) |
|-------------------------------|---------------------|---------------------|
| Bias (SD) of difference       | –0.04±0.8           | –0.09±0.9           |
| 95% limits of agreement       | –1.69, 1.60         | –1.97, 1.80         |
| Sensitivity (%)               | 81                  | 83                  |
| Specificity (%)               | 79                  | 86                  |
| Positive predictive value (%) | 78                  | 85                  |
| Negative predictive value (%) | 82                  | 84                  |
| Lin's CCC (ρ)                 | 0.88                | 0.84                |
| Cohen's Kappa coefficient     | 0.67                | 0.70                |

SD: Standard deviation, CCC: Concordance correlation coefficient



**Figure 1:** Bland–Altman plot showing agreement in hemoglobin concentration assessed by Devices A and B and hematology analyzer



**Figure 2:** Passing-Bablok regression depicting the linear equation of hemoglobin level assessed by Devices A and B compared to hematology analyzer

### Sensitivity and specificity

Device A had 81% sensitivity, 79% specificity, 78% positive predictive value, and 82% negative predictive value. The sensitivity, specificity, positive predictive value, and negative predictive value were 83%, 86%, 85%, and 84%, respectively, for Device B for the detection of anemia as compared to the hematology analyzer.

In terms of feasibility and convenience, Devices A and B were easy and convenient to use at the hospital settings. The test results were provided within 60 s for Device A and within 3 s for Device B from a single drop of the whole capillary blood sample.

### DISCUSSION

This study assessed the diagnostic accuracy of two digital hemoglobinometers compared to autoanalyzer for estimation of Hb among pregnant women. The mean difference (LOA) in the estimation of Hb compared to autoanalyzer by Device A was  $-0.04$  ( $-1.69$ – $1.60$ ) g/dL and Device B was  $-0.09$  ( $-1.97$ – $1.80$ ) g/dL. Both the devices showed good degree of agreement, linearity in Passing-Bablok regression with hematology analyzer, and similar concordance and AUCs. However, the Device B showed slightly higher sensitivity, specificity, and predictive values compared to that of Device A.

The mean difference in the Hb values measured using both the digital hemoglobinometers was  $<0.1$  g/dL as compared to the gold standard. Two hospital-based studies from India reported similar bias using Device A as compared to hematology analyzer.<sup>[11,12]</sup> In contrast to our study, studies conducted in other parts of the world have documented a slightly higher and

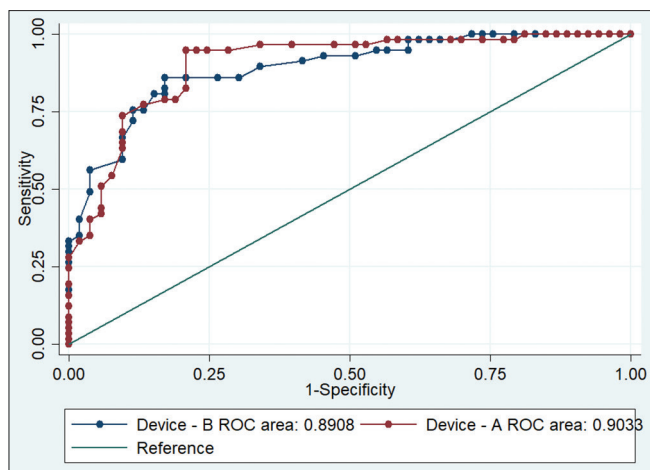
variable range of bias from 0.43 g/dL<sup>[13]</sup> (adult blood donors), 0.56 g/dL<sup>[14]</sup> (adult blood donors), and 0.61 g/dL<sup>[6]</sup> (children 6–23 months age) using Device B.

The present study reported high sensitivity for both digital hemoglobinometers (Device A – 81% and Device B 80%–83%) as compared with the gold standard hematology analyzer. In contrast, studies have reported high sensitivity of 82.1%<sup>[15]</sup> and 89.4%<sup>[11]</sup> for Device A, but varied sensitivity for device B (23.1%<sup>[13]</sup> 68.7%<sup>[7]</sup> 72%<sup>[14]</sup> and 90%<sup>[16]</sup>). However, similar to sensitivity, Device B (86%) had relatively higher sensitivity compared to Device A (79%) in the current study. Comparable findings were also reported by other studies.<sup>[13–15]</sup> The reported specificity for Device A was 63.6%<sup>[11]</sup> and 77.9%<sup>[15]</sup> and Device B was 72.1%–100%.<sup>[6,13,14,16,17]</sup>

Several factors contribute to the observed variations between the studies. Geographical variations (altitude and temperature), procedural differences in capillary blood sample, collection and Hb estimation, study participant-related factors (age groups, smoking, and biological determinants including Fahraeus effect-lower hematocrit in capillary blood vessels compared to venous vessels) might have contributed to variation across studies.<sup>[13–15,18]</sup>

The present study reiterates that both digital hemoglobinometers can be used for the screening of anemia at individual and population level. However, confirmation of anemia should be done using the gold standard devices or hematology analyzers with strict adherence to the quality control protocols.

There is a need to assess the cost-effectiveness of these devices, durability, ease of use, chances of procedural errors,



**Figure 3:** Receiver operating characteristic of hemoglobin values measured using digital hemoglobinometers compared to hematology analyzers

and ease of cleaning of the photometry/absorbance lens, which assess the Hb level, to understand the long-term functionality, maintenance charges, and use by frontline workers.

One strength of the study is adherence to quality control process for both the autoanalyzer and digital hemoglobinometers throughout the study. Single trained laboratory technician collected the capillary and venous samples, and the procedure was closely monitored. Our study has few limitations also. The accuracy of digital hemoglobinometers using the venous blood was not assessed to rule out the difference in capillary and venous blood Hb level. In addition, the study results cannot be extrapolated to all health-care workers as a trained laboratory technician collected both the capillary and venous blood samples at the health facility. We have also not assessed the validity of the devices in the subgroups, such as based on trimester and obstetric index due to the small sample size in the subgroups.

## CONCLUSIONS

Both Devices A and B have good diagnostic validity for Hb estimation. Device B fared relatively better than Device A in terms of sensitivity, specificity, and positive and negative predictive values. Increased acceptability and usage of digital hemoglobinometers in government public health systems will help in effective screening and management of anemia. Further research is required for the use of the digital hemoglobinometers at community settings by frontline health-care workers.

## Acknowledgment

We acknowledge the staff nurses, auxiliary nurse midwives, and the laboratory technicians of PHC, Chainsa, and SDH, Ballabgarh, Faridabad, Haryana, for their support in conducting the study.

## Financial support and sponsorship

Nil.

## Conflicts of interest

There are no conflicts of interest.

## REFERENCES

1. Ministry of Health and Family Welfare, Government of India. Anemia Mukh Bharat-Operational Guideline for Intensified National Iron Plus Initiative (I-NIPI) For Programme Managers; 2018.
2. UNICEF/UNU/WHO. Iron Deficiency Anaemia Assessment, Prevention, and Control A Guide for Programme Managers; 2001.
3. Oshiro I, Takenaka T, Maeda J. New method for hemoglobin determination by using sodium lauryl sulfate (SLS). *Clin Biochem* 1982;15:83-8.
4. Ahankari AS, Dixit J V, Fogarty AW, Tata LJ, Myles PR. Comparison of the NBM 200 non-invasive haemoglobin sensor with Sahli's haemometer among adolescent girls in rural India. *BMJ Innov*. 2016;2:144-8.
5. Patil P, Thakare G, Patil S. Variability and Accuracy of Sahli's Method in Estimation of Haemoglobin Concentration. *Natl J Integr Res Med* 2013;4:38-44.
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 Clin Pathol* 2018;71:532-8.
7. MedCalc Statistical Software version 16.4.3 (MedCalc Software bvba, Ostend, Belgium). Sample Size Calculation: Bland-Altman Plot. Available from: [https://www.medcalc.org/manual/sampling\\_blandaltman.php](https://www.medcalc.org/manual/sampling_blandaltman.php). [Last accessed on 2018 Aug 20].
8. Srivastava A, Koul V, Dwivedi SN, Upadhyaya AD, Ahuja A, Saxena R. Performance analysis of newly developed point-of-care hemoglobinometer (TrueHb) against an automated hematology analyzer (Sysmex XT 1800i) in terms of precision in hemoglobin measurement. *Int J Lab Hematol* 2015;37:483-5.
9. HemoCue. Point-of-care Anemia Screening-HemoCue® Hb 301 System-HemoCue. Available from: <https://www.hemocue.in/en-in/solutions/hematology/hemocue-hb-301-system>. [Last accessed on 2018 Jul 17].
10. University of York Department of Health Sciences. Measurement in Health and Disease - Cohen's Kappa [Internet]. 2008 Available from: [https://www-users.york.ac.uk/~mb55/msc/clinimet/week4/kappa\\_text.pdf](https://www-users.york.ac.uk/~mb55/msc/clinimet/week4/kappa_text.pdf). [Last accessed on 2019 Aug 02].
11. Toppo M, Pal DK, Gour D, Melwani V, Dubey M, Mishra A. Comparison of performance of digital hemoglobinometer over automated hematology analyzer for hemoglobin estimation and its user friendliness among the pregnant women in selected district hospitals of Madhya Pradesh. *Indian J Community Med*. 2019;44:31-4.
12. Dubey A, Murti S. Validation of a hemoglobinometer for use in outdoor blood donation camps. *Asian J Transfus Sci* 2016;10:164-5.
13. Sümniç A, Hron G, Westphal A, Petersmann A, Kohlmann T, Greinacher A, *et al.* The impact of noninvasive, capillary, and venous hemoglobin screening on donor deferrals and the hemoglobin content of red blood cells concentrates: A prospective study. *Transfusion* 2015;55:2847-54.
14. Baart AM, de Kort WL, van den Hurk K, Pasker-de Jong PC. Hemoglobin assessment: Precision and practicability evaluated in the Netherlands-the HAPPEN study. *Transfusion* 2016;56:1984-93.
15. Neogi SB, Negandhi H, Kar R, Bhattacharya M, Sen R, Varma N, *et al.* Diagnostic accuracy of haemoglobin colour strip (HCS-HLL), a digital haemoglobinometer (TrueHb) and a non-invasive device (TouchHb) for screening patients with anaemia. *J Clin Pathol* 2016;69:164-70.
16. Yadav K, Kant S, Ramaswamy G, Ahamed F, Jacob OM, Vyas H, *et al.* Validation of Point of Care Hemoglobin Estimation Among Pregnant Women Using Digital Hemoglobinometers (HemoCue 301 and HemoCue 201+) as Compared with Auto-Analyzer. *Indian J Hematol Blood Transfus* [Internet]. 2019;1-9. Available from: <http://link.springer.com/10.1007/s12288-019-01196-5>. [Last accessed on 2020 Jan 07].
17. Tayou Tagny C, Kouam L, Mbanya D. The new HemoCue system Hb 301 for the haemoglobin measurement in pregnant women. *Ann Biol Clin (Paris)* 2008;66:90-4.
18. Chaudhary R, Dubey A, Sonker A. Techniques used for the screening of hemoglobin levels in blood donors: Current insights and future directions. *J Blood Med* 2017;8:75-88.