# Point of Care Diagnosis of Anemia Using Portable Auto Analyzer

We describe a method for capillary blood sample-based point-of care testing of hemoglobin in population-based surveys using an automated analyzer system. The accuracy and precision of this method was comparable to hemoglobin estimated from venous blood sample (mean difference (SD) =0.2 (-2.77, 3.2), Pearson correlation coefficient, (0.969).

**Keywords**: *Accuracy, Hemoglobin, Venous blood.*

The Intensified National iron plus initiative (I-NIPI) guidelines of the *Anemia Mukt Bharat* aim at reducing anemia prevalence by 3% per annum [1]. One of the important strategies of the I-NIPI program includes screening and treatment of anemia among pregnant women (using invasive digital hemoglobinometers) and adolescent girls and boys (using non- invasive digital hemoglobinometers) [2]. The accuracy and reliability of hemoglobin estimates using these point of care devices have large variations depending on the collection technique, instrument used, and blood sampling methods (capillary *vs* venous) [3].

Automated hematology cell counters offer advantages like improved accuracy and precision, reduced subjective errors, efficiency with time, space and manpower, and safe handling of blood. However, the use of autoanalyzers is currently restricted to diagnostic laboratories due to relatively higher cost and need for use of venous blood samples [4].

Using capillary blood sample for hemoglobin estimation is advantageous for point of care diagnosis of anemia as it is less invasive, less painful and therefore more acceptable compared to venous blood samples. However, hemoglobin values using capillary samples are less accurate than venous samples [5]. Variability in sample quantity and quality based on skin thickness at puncture site and, the size and depth of incision make capillary sampling error prone. Moreover, variability of hemoglobin between consecutive blood drops has been demonstrated due to humidity, stability of reagents in cuvettes and inter-individual variation in the devices [6]. Standardized techniques that minimize inter-subject and inter-operator variation are required for both capillary blood sample collection and analytical measurements of hemoglobin. We, therefore, standardized a technique to collect capillary blood samples in microtainers for direct infusion in auto-analyzer for hemoglobin measurement. We also compared performance of different lancets for collection of capillary blood samples.

The present study was approved by the institutional ethics committee of our institution. Capillary blood samples were collected in 27 volunteers using one of the three finger-prick devices: i) Lancet A: traditional sterile lancet (Medipoint Blood lancet, NY, USA); ii) Lancet B: a pen needle device (Amkay blood lancet, Thane, India), and iii) Lancet C: contact activated lancet (Becton, Dickinson and company Ltd, Dublin, Ireland) (***Fig.* 1**). Venous sample was also collected in each participant following standard protocols [7].

Capillary blood samples were collected using protocol by Krleza, *et al*. [8]. The first drop was wiped off and 200 µL (about 4-6 drops) of free flowing sample was collected using $K_2$EDTA-microtainer (BD, Ltd, Dublin, Ireland) which was then mixed, and directly infused into an autoanalyzer (Horiba, Kyoto, Japan, approximate cost: Rs. 2.5 lakhs /unit) for hemoglobin measurement. Hemoglobin estimated using capillary and venous blood samples were compared using paired *t* test and Bland Altman plots.

Higher hematocrit values were observed in capillary samples compared to venous blood with all the lancets tested. The minimum difference was with lancet C (***Table* I**). The difference between capillary and venous samples in hematocrit and in hemoglobin were closely associated (r=0.9).

The touch activated lancet (lancet C) was advantageous compared to other two devices, as it reduced inter-individual variation in prick size and allowed effortless collection of 4-6 blood drops (about 200 µL in 20-25 seconds) into EDTA- microtainer tubes. This provided a capillary sample with characteristics closer to venous sample (***Table* I**). It caused less discomfort to participants, probably because the length of the incision device was not visible. The major difficulty with lancet A was related to the variable prick size which sometimes needed forced milking of the finger to collect adequate sample. The free flow of blood was achieved with lancet B; however, it took more than a minute for collecting the required volume of blood sample.

As the capillary blood sample collected with lancet C provided hemoglobin estimate closer to that using venous

**Table I Comparison Between Hemoglobin Estimated Using Capillary Blood Sample and Venous Blood Sample**

| *Lancets* | *Hemoglobin (g/dL)* | | |
|---|---|---|---|
| | *Venous* | *Capillary* | *MD (95% CI)* |
| Lancet A (*n*=9) | 13.9 (2.7) | 17.3 (2.57) | -3.5 (-12.4, 8.5) |
| Lancet B (*n*=9) | 13.9 (1.08) | 16.2 (2.94) | -2.3 (-6.5, 1.9)] |
| Lancet C(*n*=50) | 10.7 (1.9) | 10.5 (2.45) | 0.2 (-2.77, 3.2) |

**As the mean differences between capillary and venous sample were unacceptably high with lancets A and B, more samples were collected only with lancet C to assess the precision of the estimates; Pearson correlation coefficient (r) was –0.49, 0.84 and 0.97 for Lancet A, B and C, respectively; MD-Mean difference.*

blood sample, a large number of samples (*n*=50) were later collected in different age groups including young children in the age group 1-5 years (data not reported separately).

Capillary method of collection can overestimate hemoglobin due to higher hematocrit [4], which can be minimized by the collection method used in the present study. The (per unit) cost of lancet C was substantially higher (Rs.32) than the lancet A (Rs. 2) and lancet B (Rs. 3.50). Overall cost of analysis (about Rs 75 per sample) including, lancet C, microtainer and reagents was substantially higher than that with other commonly used PoC method like digital hemoglobinometer. However, considering the important advantages of the autoanalyzer and potential economy of scale when used in population based surveys and in screening programs for treating anemia, the benefits are likely to outweigh the cost.

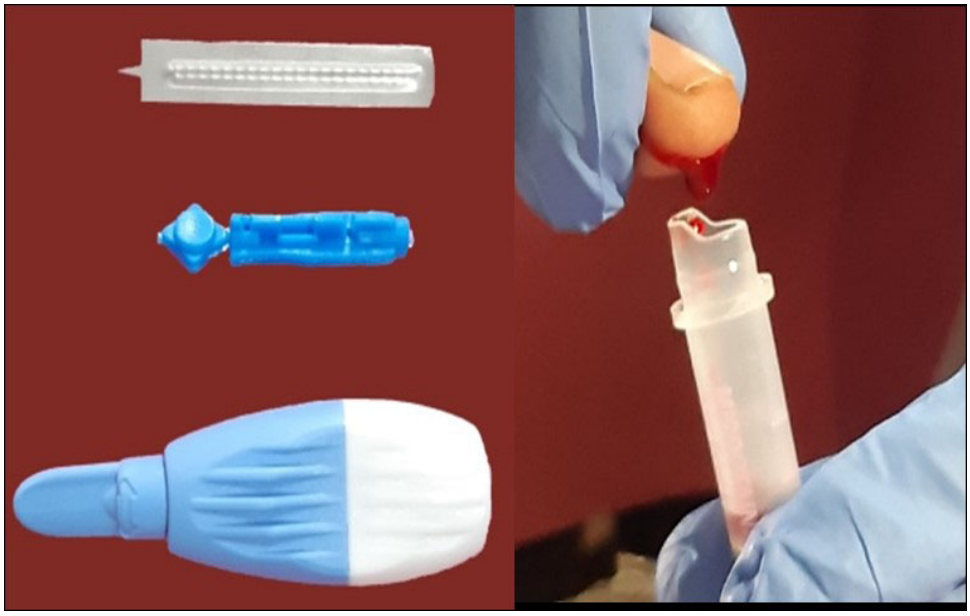
*Contributions*: LA: analyzed and interpreted data, drafted the work; TR, RNP: designed the work, acquired data, revised the manuscript; RP, BK: conceptualized and designed the work, revised the manuscript critically for important intellectual content. All authors approved for the version to be published and agree to be accountable for all aspects of the work.

*Funding*: Indian Council of Medical Research;
*Competing interests*: None stated

**LITTLE FLOWER AUGUSTINE, TEENA DASI, RAVINDRANADH PALIKA, RAGHU PULLAKHANDAM AND BHARATI KULKARNI***
*National Institute of Nutrition, Jamai-Osmania PO, Tarnaka, India.*
**dr.bharatikulkarni@gmail.com*

## REFERENCES

1. Guidelines for control of iron deficiency anemia: National iron plus initiative. Ministry of Health and Family Welfare, Government of India 2013. Available from: *https://www.nhm.gov.in/images/pdf/programmes/child-health/guidelines/Control-of-Iron-Deficiency-Anaemia.pdf. Accessed October 15, 2019.*
2. Intensified National Iron Plus Initiative (I-NIPI): Operational guidelines for programme managers. Ministry of Health and Family Welfare, Government of India 2018. Available from: *https://www.fitterfly.com/site/pdf/anemia-mukt-bharat.pdf.* Accessed September 30, 2019.
3. Neufeld LM, Larson LM, Kurpad A, Mburu S, Martorell R, Brown KH. Hemoglobin concentration and anemia diagnosis in venous and capillary blood: biological basis and policy implications. Ann NY Acad Sci. 2019;1450: 172-89.
4. 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.
5. Tang R, Yang H, Choi JR, Gong Y, You M, Wen T, *et al*. Capillary blood for point-of-care testing. Crit Rev Cl Lab Sci. 2017;54:294-308.
6. Whitehead Jr RD, Mei Z, Mapango C, Jefferds ME. Methods and analyzers for hemoglobin measurement in clinical laboratories and field settings. Ann NY Acad Sci. 2019;1450:147-71.
7. World Health Organization. WHO guidelines on drawing blood: best practices in phlebotomy 2010. Available from: *https://www.who.int/infection-prevention/publications/drawing_blood_best/en/.* Accessed October 15, 2019.
8. Krleza LJ, Dorotic A, Grzunov A, Maradin M. Capillary blood sampling: National recommendations on behalf of the Croatian society of medical biochemistry and laboratory medicine. Biochemia Medica. 2015;25:335-58.



**Web Fig**. **1** *Lancet devices and microtainer; Lancet a: traditional; Lancet b: pen needle device; and lancet c: contact activated.*