Original Research Article

Comparison of automated method and photometric cyanmethemoglobin method for haemoglobin estimation

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ABSTRACT

Background: Haemoglobin is one of the commonest tests required in pathology labs. There are many methods of haemoglobin estimation. Common methods used are sahli’s acid hematin method, photometric cyanmethemoglobin method with hemoglobin estimation by colorimeter, automated method by cell counter.

Objective: To evaluate accuracy of hemoglobin estimation by manual photometric method versus automated method by hematology cell counters using the same sample at the same time.

Materials and Methods: Blood samples of 460 adult patients and 72 children (<15 Yrs), including outdoor and indoor, between May 2019 to Aug 2019, attending H.I.M.S. were collected in EDTA tubes. Samples were properly mixed on blood shaker. Hemoglobin estimation done by photometric cyanmethemoglobin method by AIMIL digital colorimeter and by Mindray (BC5150) automated cell counter.

Results: Results by photometric method showed higher mean value compared to automated method by 2.52%. Commercial control results showed 2.2% coefficient of variation by Mindray cell counter and 2.9% by photometer.

Conclusion: Both methods are accurate with 2.52% more mean value in photometric method. When Haemoglobin is the only test required, photometric method is cost effective and feasible. If sample size is large as in tertiary care hospitals and multiple parameters like complete blood count (CBC) are needed, automated method is time effective and feasible.

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1. Introduction

Hemoglobin comprises of four globin protein subunits, each having one polypeptide chain and one heme group. Oxygen binds reversibly to the ferrous iron atom in each heme group.1,2 Main function of haemoglobin is to transport oxygen from lungs to the tissues, where oxygen is utilized for metabolism,3 mainly to facilitate oxidative phosphorylation in mitochondria.4 Anaemia is very common in our country, worldwide it affects over 800 million women and children,5 also there are fair number of cases in advanced countries like US,6 so haemoglobin is advised frequently to the patients especially in antenatal patients because of increased risk of complications.7 Hb measurement is also essential for screening the eligibility of donors in blood banks.8 The reference range for normal hemoglobin (according to WHO) is 13-18 gm/dl for men, 12-16 gm/dl for women.9,10

Hemoglobin being one of the commonest investigations in laboratory, we require economical, feasible and accurate method.11 Many methods are commonly used, sahli’s acid hematin method, cyanmethemoglobin method by photometer, automated method by cell counters.3 Different labs use different methods depending on laboratory location, number of patients, availability of technical staff, problem of electricity, affordability. There is variation in hemoglobin measurement done by different instruments due to many reasons including type of sample.10

Photometric cyanmethemoglobin method is based on principle that Drabkin reagent reacts with hemoglobin...
in blood to form cyanmethemoglobin and developed
colour is measured by photometry at 550 nm. Automated
method on Mindray cell counter is based on principle of
electrical impedance for cell counting and colorimetry for
HB estimation. Automated method uses a non cyanide
hemoglobin method.

2. Aims and objective

The study were to evaluate accuracy, cost effectiveness,
suitability and feasibility of photometric versus automated
method of haemoglobin estimation.

3. Materials and Methods

We took 532 patient’s (460 adults and 72 children) Hb
estimation into consideration between May 2019 to Aug
2019 including indoor & outdoor patients attending Hind
Institute of Medical Sciences, Safedabad, Barabanki, Uttar
Pradesh. Adults were between 15 to 85yrs of age. Children
were less than 15yrs upto 03 hrs old, newly born baby.
2 ml blood samples were collected in tubes containing
K3EDTA anticoagulant. After proper mixing on blood
shaker Hb estimation was done simultaneously both by
Mindray 5 part hematology cell counter (BC-5150) and
digital colorimeter by Aimil. For manual method 5 ml
Drabkin’s reagent was taken in test tube and 20 ul of
blood was mixed and then waited for 5minutes at room
temperature before taking absorbance on colorimeter at
550 nm filter against Drabkin’s solution. A standard curve
is used to know the Hb concentration in the sample
by measuring absorbance compared to standard control.
Drabkin’s solution contains cyanide which is hazardous
during handling and disposal. Drabkin’s reagent was used
from Arkray (formerly Span Diagnostics). Standards were
also used from same company. For automated cell counter,
control blood was used from Diagon. Standards (control)
were run regularly. We did not include hemolyzed sample
in our study.

4. Results

A total of 532 samples were processed for hemoglobin
estimation. Out of these 238 were males and 294
females, 460 adults, 72 children. Mean haemoglobin
concentration on Mindray cell counter for adults and
children were 10.62±2.26 and 10.12±2.41 respectively.
Mean haemoglobin concentration by cyanmethemoglobin
method on photometer (AIMIL) for adults and children
were 10.84±2.05 and 10.42±2.32 respectively. This shows
mean by photometric method is greater by 2.07% for adults
and 2.96% for children. Accuracy of both the methods was
compared using Microsoft Excel software.

This showed 2.52 % (overall) increase in results by
manual method. Comparison of different parameters is
tabled.

5. Discussion

For a developing country like India, economically suitable
methodology is the need of the hour. At our institute cost of
the test by automated cell counter is Rs 100 whereas cost
of manual photometric method is Rs 10 per test. Cost of
Mindray Cell Counter is about Rs 4.0 lakh while AIMIL
photometer cost only for Rs 9,500. When haemoglobin is
the only test required, photometric method is cost effective
and feasible. Automated method should be used where
complete blood count (CBC), or multiple parameters are
required. International committee for standardization in
haematology (ICSH) has recommended the ‘Drabkin’ as the
method of choice and have suggested all the other method
should be adjusted to be comparable to this method because of availability of internationally accepted reference
Periodic standards (controls) must be run to maintain accuracy both in manual & automated method. Hemoglobin estimation by automated cell counters is the next best method. There is a significant and positive correlation between the manual and the automated method. For accuracy proper sample collection and proper technique is very important.
6. Conclusion

Both methods of Hb estimation are accurate and for single parameter manual method is very cost effective.

7. Abbreviations

Hb: Hemoglobin
HIMS: Hind Institute of Medical Sciences

8. Source of Funding

None.

9. Conflict of Interest

None.

References


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