

Reliability tests of portable instruments
for measurement of haemoglobin
and packed cell volume

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INTRODUCTION

Simple methods for accurate and reproducible Hb-measurement for use in a PHC setting are available (1). The scope of this study is to test a number of field-use resistant techniques that could provide some extra information at an acceptable cost. This might be useful for research or referral level purposes. The techniques that were tested are two slightly more sophisticated Hb measurement techniques (the Compur 1000 mini photometer and the AO-Spencer Hb-meter) and two micro-haematocrit measurement techniques (the Compur M 1000 mini-centrifuge and a manual centrifuge)

THE TECHNIQUES

1. The Compur 1000 miniphotometer for haemoglobin
Manufactured by Compur-Electronic GmbH, Munchen
Battery or mains operated (two 1.5V batteries)
Very small portable photometer with direct Hb-scale in g/dl. Quantity of blood needed for analysis is standardized by using precalibrated capillary tubes, which have to be purchased from the manufacturer.
Quantity of diluent liquid (=modified Drabkin?) is standardized by using single-use disposable cuvettes, filled with a fixed amount of diluent, which have to be purchased from the manufacturer.
According to the manufacturer's instructions, setting the 0-value is simple.

Purchase price of the apparatus: 17.400 BF; of the capillary tubes and of the disposable cuvettes: 925/40 =29/measurement. The cuvettes should be kept at between 4-25 degrees Celcius.

2. AO Spencer Hb-meter, manufactured by American Optical,

battery or mains operated.

Small portable comparator with direct Hb-scale in g/dl. Whole, undiluted blood is placed on a cell of the comparator and haemolyzed by stirring it with a disposable saponin-coated stick supplied by the manufacturer. The comparator (continuous) scale is adjusted until its green color matches that of the sample of the cell. After use, the cell is cleaned with water and alcohol.

Purchase price of the apparatus: 14,200 BF; of the saponin-coated sticks: $780/100 = 7.8$ BF /measurement

3. Compur M 1100 mini centrifuge for haematocrit. Manufactured by Compur-Electronic GmbH, Munchen. Battery or mains operated (six 1.5 V batteries). Small portable centrifuge for micro-haematocrit measurements. The capillary tubes are calibrated for length, and are filled entirely with whole blood. The apparatus can take 6 tubes at a time. Centrifugation time is automatically fixed. Haematocrit values are read directly on a fixed scale which is incorporated in the instrument, alongside each capillary tube. A set of ordinary batteries can supply the energy for some 85 centrifugations (which makes for $85 \times 6 = 510$ blood sample measurements). The capillary tubes are supplied by the manufacturer.

Purchase price of the apparatus: 19,500 BF; of the capillary tubes: $998/500 = 2$ BF/measurement; of the transformer: 2980 BF

4. Manual haematocrit system, manufactured by Ingeniorsfirman Instrumentjanst, Sundbyberg, Sweden. A length of 18 cm of flexible polyethylene capillary tubing is sucked almost full with whole blood, doubled up in a U-shape and fitted into a groove on a flat plastic disc. The disc can take up till 8 pieces of tubing (8 grooves) and has 4 holes around its middle, through which strings

are drawn. The disc is made to rotate at high speed by pulling and releasing tension on the strings. When maximum speed is attained, the pull is repeated 50 times. The tubing is then taken out of the groove, straightened, and lengths of the total column and the packed cell column are measured. PCV is then calculated by simple division.

Purchase price of the apparatus: 3000-4000 BF
of the tubing: $1825/15m = 22$ BF/measurement.

PROCEDURE

All the measurements with the techniques under investigation were carried out by 4 observers, volunteers recruited from the second year of a school for laboratory technicians.

Each observer made 2 measurements of each of the 15 blood samples with every technique. They were given an instrument, and 15 random numbered blood samples. After having completed the 15 measurements, they took another instrument, and the 15 blood samples received different random numbers. After these measurements, random numbers were changed again, and each observer went back to his first instrument, and then to the second one. This procedure was repeated on the second day, where every observer used the 2 remaining techniques. Haemoglobin concentration of the 15 samples ranged from 40 g/l to about 170 g/l; they were obtained by diluting blood, collected the previous day, with its autologous plasma, obtained by centrifugation.

Reference measurements were carried out on the same day as the experimental measurements. Two reference measurements were done each day, one before the observers started their estimations (08,00 h) and one after they had finished (14,00 h). This was done for both Hb and Ht measurements. Reference values were determined from automatized

measurements of cyanmethaemoglobin by professional experienced laboratory personnel, using a haemoglobinometer manufactured by Coulter Electronics Co (Harpenden, Hertfordshire, England), and a Hawksley centrifugal apparatus for microhaematocrits, set at 5 minutes.

RESULTS

The results are presented in table 1. Mean differences and mean absolute differences between first and second measurements are a proxy for precision; mean differences and mean absolute differences with the average reference measurements are a measure of accuracy. Better information is available from the regression equation: the SEE is a measure of the width of the distribution of measurements around the average measurement of a sample, this average being given by the regression equation; it is therefore a measure of precision. The distance between the regression curve and the ideal curve (with estimated Hb = real Hb) is indirectly correlated to accuracy, and shows the variation of the accuracy in function of the haemoglobin concentration of the sample.

TABLE 1 : RESULTS

Technique	Mean difference 1st and 2nd measurements (2nd - 1st)	Mean absolute difference 1st and 2nd Measure- ments 2nd - 1st	Mean difference with average reference measu- rement	Mean absolute difference with average refe- rence measure- ment	Regression equation Ytechn = axref+b	SEE %
Reference Hb Coulter	+2.2333 g/l	2.6667 g/l				(.225 g/l)
Compur 1000 Hb	+3.4833	3.9500	+33.4583	33.4583	$y = .995x + 33.555$	3.575 g/l .993 .9965
A.O. Spencer Hb	-.3667	4.5667	+2.0333	5.2242	$y = .919x + 5.719$	5.54 g/l .980 .990
Reference Hc Hawksley	-.36667x	.46667x				(.4901)
Compur H 1100 Hc	-.3333	.8333	-.3500	.6458	$y = .992x - .155$	1.024x .993 .9965
Manual Hc	-.5917	1.4250	+5.5708	5.5708	$y = 1.19x + .662$	2.138x .979 .989

Table 2 gives the operational characteristics of each of these techniques in two typical epidemiological situations, one of pregnant women in the Comoros, with a high (2) prevalence of low Hb values, and one of adult men from Central America (3), with low prevalence of Hb values of less than 90 g/l. Sensitivity, specificity and predictive values were calculated according to the methodology described in (1).

TABLE 2 : OPERATIONAL CHARACTERISTICS

1. Applied to an existing African population of pregnant women with mean Hb at 93 g/l (S.D. 10.8 g/l) or mean Ht at 28% (S.D. 3.25%)
Cut-off point 90 g/l
After correction of measurements with regression equation

Technique	Sensitivity	Specificity	Predictive value +	Predictive value -
Compur Hb	96.6	97.9	94.9	98.7
Spencer Hb	90.0	93.7	85.1	96.0
Compur Ht	97.0	98.2	95.5	98.8
Manual Ht	90.1	93.7	85.2	96.0

2. Applied to a population of Central American males with mean Hb at 150 g/l
Cut-off point 90 g/l
After correction of measurements with regression equation

Technique	Sensitivity	Specificity	Predictive value +	Predictive value -
Compur Hb	99.9	99.9	98.3	99.9
Spencer Hb	98.9	99.9	94.8	99.9
Compur Ht	99.9	99.9	98.5	99.9
Manual Ht	98.9	99.9	94.8	99.9

3. Time per measurement (series of 15 estimations, with minimal time loss)

-Compur Hb	2.5'
-Compur Ht	2.6'
-AO Spencer	3.0'
-Manual Ht	2.3'

4. Minimal significant difference ($p = 0.05$): $2s\sqrt{2}$

-Compur Hb	10.1 g/l
-AO Spencer Hb	15.7 g/l
-Compur Ht	2.9 %
-Manual Ht	6.0 %

DISCUSSION

The COMPUR 1000 haemoglobinometer is nearly as precise as the reference technique. There are some inter-reader differences in precision: the average absolute differences between measurements range between 0.0 and 0.0 g/l, the latter being due to a systematic error made by observer 3.

Accuracy, however, is not satisfactory. A systematic error of + 33.5 g/l was recorded, even after adjusting the zero level according to the manufacturer's instructions. It is therefore necessary to use a haemoglobin standard and calculate necessary corrections; this correction is simple in as much as the regression curve is parallel to the reference value curve, and a simple addition-subtraction is sufficient.

It remains nevertheless a serious handicap for this technique. Moreover, the COMPUR is relatively costly: cost is of about 20 B.F. (*) per measurement. The high cost, need for constant supply of the special dilution materials and systematic errors in accuracy do not make it a useful alternative to simpler and cheaper methods (Lovibond undiluted) that are not significantly less precise and are more accurate. In high prevalence situations one may nevertheless obtain an important gain in positive predictive value, after the correction for the systematic error has been made.

The AO (Spencer) Haemoglobinometer has a precision and accuracy comparable to that of the Lovibond-undiluted (1). It is more expensive than the latter (about 14200 BF investment and 8 BF per measurement running costs), more cumbersome in use and requires supplies of saponin sticks and batteries. It offers no particular advantages over the

*: 1 US\$ = 55 BF

Lovibond-undiluted for survey purposes, and has some operational disadvantages for routine use in a PHC-setting.

Precision and accuracy of the Handpowered Microcentrifuge are satisfactory; there is nevertheless a systematic error at higher values. This confirms observations by Mahin et al(4) on cattle and equidae. Used as a screening technique, sensitivity, specificity and predictive values, after correction for the systematic errors, compare well with the Lovibond undiluted and Spencer techniques. The cost is nevertheless high: 22 BF per measurement, without counting investment. The technique is cumbersome and tiring and requires a supply of special tubing; on the other hand the apparatus seems almost indestructible. Its only advantage is that no power supply is needed. Although it may offer a useful complement to information on Hb, the Compur microcentrifuge seems to be a better choice.

The COMPUR M1100 Minicentrifuge has an excellent precision and accuracy, without systematic errors. Robustness could not be tested, the cost is of about 19500 BF investment with 2 BF per measurement running costs, and a supply of batteries is required. Nevertheless this technique seems a useful addition to haemoglobin measurement, at least at referral level and for survey purposes. It can be used as an excellent screening tool in a PHC-setting if the price is not considered prohibitive.

REFERENCES

1. W. Van Luchburgh, G. Rogels, G. Cornelis, C. Ancona, E. Mangelmohr & H. Van Halbeek. Haemoglobin measurement: Reliability of some simple techniques for use in a primary health care setting. Bull WHO 61(6) 1983.
2. G. Cornelis. Unpublished data.
3. F. Viteri & M.A. Guzman. Haematological status of the Central American population: prevalence of individuals with haemoglobin levels below 'normal'. British Journal of haematology, 23: 725-735 (1974).
4. Mahin, Kindermann & Verhulst: Ann.Soc.Belge Med.Trop.62:259-260 1980.

ANNEX

Fig. 1 (p.10 - 11)

Regression line (full line) and 95 % confidence belt (broken lines) of all measurements made with each of the techniques described above, on the reference value. The diagonal (broken-dotted line) is the line of perfect agreement.

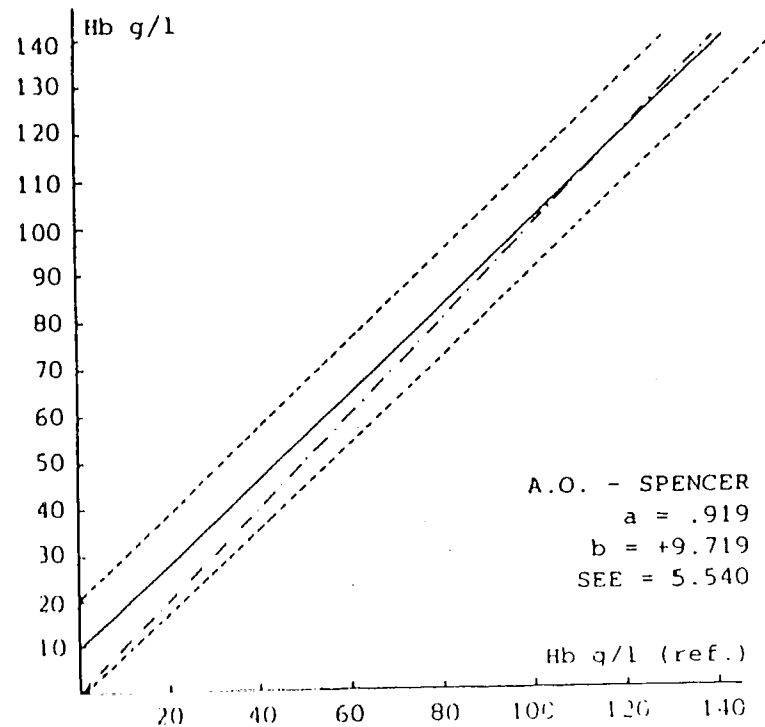
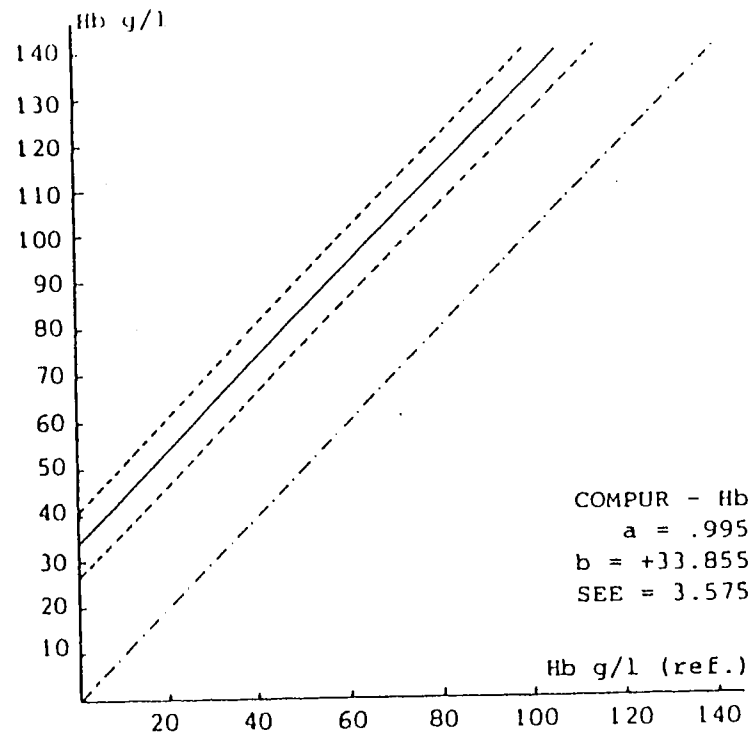
The constants a and b refer to the regression equation $y_{\text{techn.}} = ax + b$ (see also table 1)

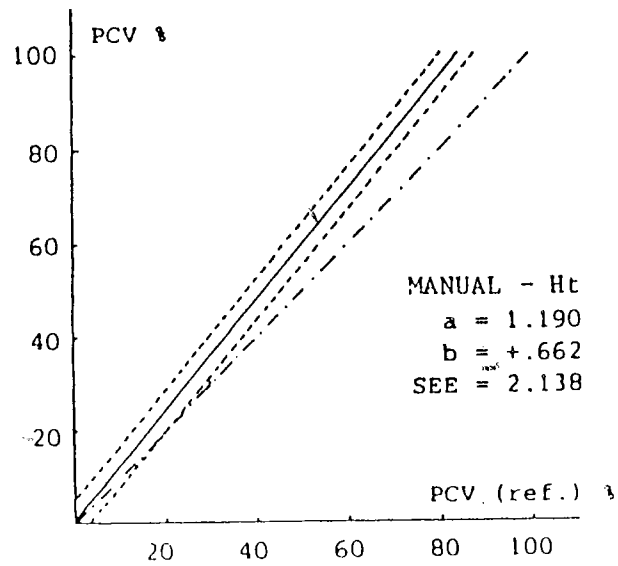
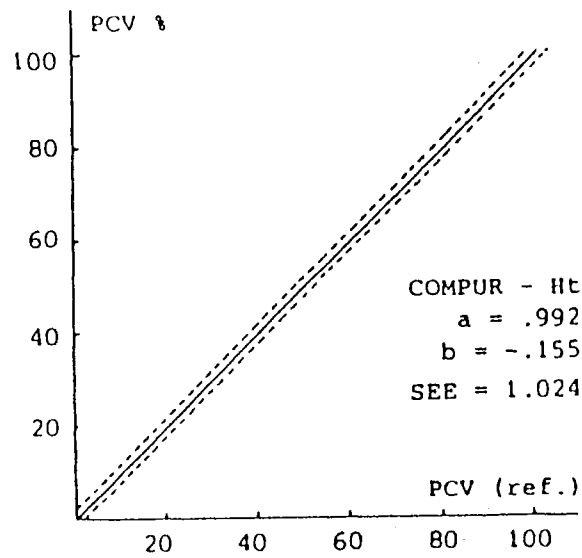
Tables 3 - 4

Expected proportions of false positives and false negatives within each haemoglobin or P.C.V. class when screening a population at a cut-off point of 90 g/l or 27 % P.C.V..

The same proportions can be used with other cut-off points, provided the class width remains the same.

These proportions should be applied after correction of measurement results with the regression equation.





Hb g/l	Compur 1000 Hb	A.O. Spencer Hb	Lovib. Und. Hb
40 - 45			
45 - 50			
50 - 55			
55 - 60			
60 - 65			.000009316
65 - 70		.0000168	.00018037
70 - 75		.0004519	.00215
75 - 80	.00002	.0064184	.01616
80 - 85	.00269	.048571	.076804
85 - 90	.0820206	.201471	.237776
90 - 95	.0820206	.201471	.237776
95 - 100	.00269	.048571	.076804
100 - 105	.00002	.0064184	.01616
105 - 110		.0004519	.00215
110 - 115		.0000168	.00018037
115 - 120			.000009316
120 - 125			
125 - 130			
130 - 135			
135 - 140			
140 - 145			

Table 3. Expected proportions of false positives (above the line) and false negatives (below the line) within each haemoglobin class when screening for anaemia with a cut-off point of 90 g/l

PCV %	Compu M1100 Hc	Manual Hc
12-13.5		
13.5-15		
15-16.5		
16.5-18		
18-19.5		
19.5-21		.000015
21-22.5		.004196
22.5-24	.000007	.006128
24-25.5	.00183	.04748
25.5-27	.07309	.2018898
27-28.5	.07309	.2018898
28.5-30	.00183	.04748
30-31.5	.000007	.006128
31.5-33		.004196
33-34.5		.000015
34.5-36		
36-37.5		
37.5-39		
39-40.5		
40.5-42		

Table 4. Expected proportions of false positives (above the line) and false negatives (below the line) within each PCV-class when screening for anaemia with a cut-off point of 27 %