

Letters to the Editor

Rapid cyanmethaemoglobin estimation

In order to obtain a more rapid estimation of whole blood haemoglobin as cyanmethaemoglobin, for use in antenatal and other outpatient clinics, we have studied the conversion rates achieved by two commercially available neutral cyanide-ferricyanide solutions which are based on the Van Kampen and Zijlstra (1961) formula. Two commercially available alkaline solutions (Drabkin and Austin, 1932) were also studied by way of comparison.

Haemoglobin readings were made at minute intervals following dilution of 0.02 ml samples of EDTA (sequestrene) venous blood in 4 ml volumes of the appropriate cyanide-ferricyanide solution (reconstituted in glass-distilled water of pH 5.8). The readings were made to the nearest 0.05 g/100 ml using a simple colorimeter (EEL haemoglobin meter, Evans Electroselenium Ltd) and the method gave a coefficient of variation of 1.3% for 12 replicate estimations. The commercial reagents studied are listed in the table and their respective mean conversion rates to a stable haemoglobin value, for 10 venous specimens are shown in the figure.

Each of the alkaline solutions gave a virtually stable haemoglobin value at 15 minutes, and this conversion time was not shortened by the addition of a detergent to one of them to promote red cell lysis (Fielding, 1959). Only one of the two 'neutral' solutions conformed to the recommendations of the International Committee for Standardization in Haematology (ICSH) (1967) that the working pH be in the range 7.0-7.4. Reagent 4, which is marketed in a dry form, gave a working solution of pH 7.7; while further samples from the same batch, but reconstituted 12 weeks later, gave a working solution of pH 8.2. A fresh batch obtained from the manufacturer gave a working solution of pH 7.3, and this failure of the buffer to maintain the correct pH after storage is presumably due to absorption of moisture by the dry potassium cyanide (Matsubara and Shibata, 1969). All samples of reagent 5, which is marketed as a concentrated solution, gave a working pH of

Reagent	Manufacturer	Chemical Composition (per Litre of Distilled Water)	pH Obtained
1 Aculte	Ortho Diagnostics Ltd	K ₃ Fe (CN) ₆ 200 mg KCN 50 mg NaHCO ₃ 1000 mg	8.6
2 Diagen	Diagnostic Reagents Ltd	K ₃ Fe (CN) ₆ 200 mg KCN 50 mg NaHCO ₃ 1000 mg	8.6
3 Diagen	Diagnostic Reagents Ltd (with added detergent)	K ₃ Fe (CN) ₆ 200 mg KCN 50 mg NaHCO ₃ 1000 mg Nonidet P40 1 ml	8.6
4 Buffered diluent for haemoglobin determination (stable dry reagent)	British Drug Houses Ltd	K ₃ Fe (CN) ₆ 200 mg KCN 50 mg KH ₂ PO ₄ 140 mg Nonidet P40 1 ml	7.7
5 Concentrated neutral Drabkin solution	Diagnostic Reagents Ltd	K ₃ Fe (CN) ₆ 200 mg KCN 50 mg KH ₂ PO ₄ 140 mg Nonidet P40 1 ml	7.0

Table Key to the five preparations used in the study. (Preparation 3 is not commercially available)

7.0. This is critical for rapid conversion (see Figure).

An additional 75 venous specimens (Hb range 3.10-17.80 g/100 ml) were studied using reagent 5. Stable haemoglobin values were again obtained within five minutes. When the haemoglobin value was read at one minute, compared with five minutes, a false low mean error of only 0.05 g/100 ml (SD \pm 0.07, range 0.0-2 g/100 ml) was obtained. Strict precautions were taken to exclude light

from the working solution. Ten of the specimens gave white cell counts within the range 12,000-54,600/cmm and two specimens contained paraproteins. Neither the leucocytosis nor the presence of abnormal proteins had any detectable effect on the conversion rate.

Reagent 5 has therefore been found to conform to the ICSH (1967) recommendations and, provided that light is excluded from the working solution, the conversion rate is sufficiently rapid for haemoglobin readings to be made at one minute. The reagent can therefore be recommended for outpatient and emergency use. Cyanide-ferricyanide solutions reconstituted from dry products are not recommended unless the final pH is checked on each occasion and adjusted where necessary.

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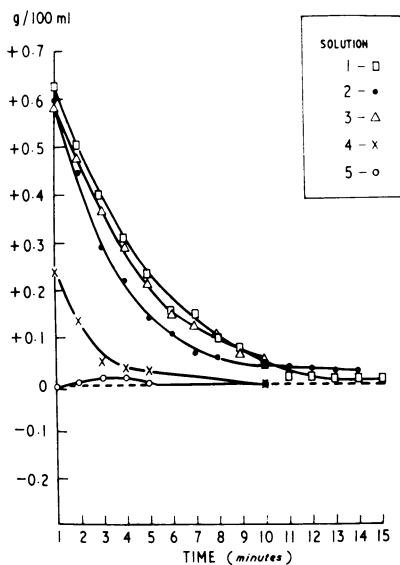


Fig. Rate of conversion to cyanmethaemoglobin of 10 venous blood specimens using 5 cyanide-ferricyanide solutions (net change of mean haemoglobin values recorded at minute intervals).

References

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