Regional quality control

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(On behalf of the Wessex haematologists)

SYNOPSIS Methods of quality control of some major haematological techniques are described. These methods have been applied in haematology laboratories serving a population of 2 million using existing facilities for preparation and transport of the necessary materials.

Frequent interchange of ideas between the haematologists working in the hospitals in the area formerly administered by the Wessex Regional Hospital Board, and the difficulty in maintaining acceptable standards of accuracy with increasing workload under difficult conditions, has led to the development of quality control procedures which can be applied simply to a large geographical area. An account of these procedures in blood counting, assay of vitamin B₁₂ and folate, and in blood grouping and antibody detection follows.

The Wessex Blood Standard

This preparation arose from a desire to use frequent whole blood assay as a batch control of blood counting rather than rely on the more retrospective statistical methods alone. In essence it is the issue weekly, by regular Blood Transfusion Service transport, of blood preserved in Alsever's solution. At any one time two different blood standards are in use, one of normal and one of low values. One laboratory undertakes the assay of this blood and sends its results to other participating laboratories. The technical staff of the participating laboratories discuss with the staff of the assaying laboratory any disagreement with the stated values.

The development of this standard blood has been described elsewhere (Parnell, 1972) but the following features are important. Alsever's solution is 0.8% trisodium citrate, 1.9% glucose, and 0.4% sodium chloride in water. The Blood Transfusion Centre at Southampton each Monday arranges for HAA negative blood from a donor unsuitable for transfusion purposes to be taken into Alsever's solution. The blood is then dispensed into 5 ml containers under sterile conditions. In alternate weeks the blood is partially packed before aliquoting. This provides

normal and low standards which have a life of about 14 days. It is found that values for Hb and RBC do not alter significantly in this time and the MCV and PCV increase by 4 and 0.4 respectively; the white cell count changes in an unpredictable way.

A number of aliquots suitably numbered by batch is distributed with stocks of blood for transfusion to the laboratories at Southampton, Dorchester, Salisbury, Winchester, and Basingstoke. The first and last aliquots and a selection of others are sent to Portsmouth where the blood is assayed by Coulter S Counter as well as by colorimetry against an international Hb standard, visual particle counting, and microhaematocrit. The results are sent by post to all participating laboratories (table I).

Batch	Hb (g/dl)	WBC ¹ (10 ⁹ /l)	RBC (10 ¹² /l)	MCV (fl)
243 Stated	9.5	3.5	3.16	87
Mean	9.38	3.24	3.21	84.5
SD	0.13	0.27	0.07	1.65
CV	1.4	8.5	2.3	2.0
244 Stated	10.5	4.5	3.33	93
Mean	10.72	4.88	3.54	89.33
SD	0.14	0.34	0.08	2.47
CV	1.3	7.5	2.3	2.8
245 Stated	10.5	3.5	3.27	96
Mean	10.39	3.29	3.39	90.9
SD	0.11	0.44	0.08	2.98
CV	1.1	13.4	2.2	3.3
246 Stated	12.7	5.2	4.38	91
Mean	12.82	5.02	4.48	85.4
SD	0.20	0.23	0.10	2.99
CV	1.5	4.6	0.23	3.5

Table I *Wessex Standard blood: Batches 243, 244, 245, and 246 (September 1975): Correlation of results between stated values and six other laboratories using Coulter S Counter*

¹These values are greatly affected by age of the specimen on the day of assay which will vary geographically.

The recommended use of this standard is threefold. Immediate discussion of disagreement with

Portsmouth's assay takes place each week; a fresh aliquot is used daily as a calibration check for blood counting in each laboratory; and the standard blood is used in each laboratory in the assay of a further 'standard blood' for batch control throughout the day. The last use is necessary as there is unfortunately not enough of the Wessex Standard Blood to be used throughout each working day.

Quality Control of Vitamin B₁₂ and Folate Assays

Samples of human serum, haemolysates, and solutions of vitamin B₁₂ and folic acid are distributed four times a year to the participating laboratories for assay under routine conditions. To date 15 series of assays have been performed in each of the six Wessex laboratories which perform assays, and two additional laboratories have each taken part on seven and 10 occasions. The present distribution is to nine laboratories.

Each batch of samples consists of two sera for serum vitamin B₁₂, two sera for serum folate, two haemolysates for haemolysate folate, and one solution each of vitamin B₁₂ and folic acid. Each pair of samples is selected to include one normal sample, which is expected to be read on the steepest part of the assay curve, and a low sample, within the range 50-150 pg/ml for vitamin B₁₂, 1.0-3.0 ng/ml for serum folate, and 20-200 ng/ml for haemolysate folate. Samples of the same serum are often sent in different trials.

Suitable serum is collected from patients known to have normal or low blood levels, usually being serum remaining after laboratory testing. Sera are stored at -30°C and, once thawed, are not refrozen. Haemolysates are prepared from normal human red cells and are suitably diluted. Solutions of cyanocobalamin in cyanide-acetate buffer and pteroylglutamic acid in ascorbate-phosphate buffer are prepared in dilutions suitable for direct addition to an equal volume of assay medium; these solutions assess the assay procedure alone by excluding errors arising from dilution, and multiple pipetting. These batches of samples are despatched by post for inclusion in the next routine assay.

Individual laboratories return their results within three weeks and a summary of all reports is distributed quarterly.

The mean, SD, and CV for replicate assays performed in 10 consecutive weeks at the organizing laboratory are used for assessing the Regional results (table II). When assessing results returned from Regional laboratories owing to the small number of results the calculated median value is taken after excluding any values which are outside 3 SD of the crude median. The reported results from each

laboratory are charted and the ratio of values from paired samples is also reported.

	Mean	SD	CV%
Serum vitamin B ₁₂	303	51	17
Serum folate	2.8	0.9	31
Red cell folate	42	11.7	28

Table II *Analysis of a typical series of replicate assays performed in the organizing laboratory on samples to be sent to regional laboratories*

Quality Control in Blood Transfusion Serology

This takes the form of proficiency testing of cross-matching procedures. At approximately three-monthly intervals the 11 hospital blood banks in the Wessex Region receive five different samples of serum to be tested against four different red cell samples. The techniques used are saline at room temperature and 37°C, albumin displacement, and the indirect antiglobulin test at 37°C. The antibodies used are selected from anti-A, -B, -D, -C + D, -c, -K, -M, and anti-P₁. Usually it is arranged that one cell sample is compatible with one serum sample by all methods.

The cells and sera are also tested by the Blood Group Reference Laboratory, London and the scheme closely follows the one originally devised by Dr J. Darnborough of the Cambridge Regional Blood Transfusion Centre.

After the results have been analysed they are sent to the hospital haematologist and the senior blood bank technician within 10 days, together with any recommendations which might be suggested by the findings.

Discussion

We feel that the Wessex Blood Standard has proved its value during the four years that it has been in use. It is cheaper to use than commercial whole blood standards and it has the advantage that we have to determine our own standards of accuracy. Our blood standards are complementary to those originated by Dr S. M. Lewis at the Royal Postgraduate Medical School at Hammersmith, and both are an essential and integral part of our overall quality control system for blood counting. The main disadvantage is the variable instability of the white cell count but it is hoped that at some stage nucleated erythrocytes might be added in place of the white cells. The participating laboratories tackle quality control of the white cell count by other methods although major alterations in counting would be detected by the present standard.

With vitamin B₁₂ and folate assays improvement in the quality of work at some laboratories has resulted from awareness of the results from other laboratories and subsequent discussion of methodology and technique. Distribution of aliquots of the same serum at intervals has shown that serum folate stored at - 30°C is stable for 12 months provided that it is not refrozen after thawing. Ascorbic acid is not required for the preservation of folate under these conditions and this allows the same serum to be used for both folate and vitamin B₁₂ assays. Dilute solutions of vitamin B₁₂ and folate are stable for three weeks if frozen.

It is considered that the organization of proficiency testing in compatibility procedures is best handled at the Regional level where the technical and medical staff of the Regional Transfusion Centre are in daily contact with their colleagues in the hospital laboratories and familiar with their problems. Generally all staff have welcomed and fully cooperated in the proficiency testing scheme concerning cross-matching procedures, and often several technicians in each hospital blood bank have completed the tests. Two of the smaller hospital laboratories found the scheme time-consuming as presently organized, and it was suggested that three cells were tested against four serum samples in their instance.

These approaches to quality control have been initiated by haematologists in an attempt to solve the problems of the maintenance of accuracy in their laboratories. They have helped a great deal in this and have, by stimulating discussion between the staffs of the participating laboratories, encouraged interchange of ideas and methods.

The consultant haematologists of the Region who have participated in the schemes described are: Dr Aronstam (Basingstoke), Dr Gilliver (Dorchester), Dr Lee Potter and Dr Hamblin (Bournemouth, Poole), Dr Green and Dr O'Brien (Portsmouth), Dr Pinkerton (Salisbury), Dr Hyde, Dr Leslie, Dr Guy, and Dr Chisholm (Southampton), and Dr Mavor (Winchester).

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Reference

- Parnell, D. H. (1972). A quality control system for the Coulter Counter Model 'S'. *Med. Lab. Technol.*, **29**, 401-404.