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## An Approach to the Cost Evaluation of Discrete Analysers

M. J. Hablous, B.Com.

Management Services Section of the Auckland Hospital Board Received for Publication, October 1979

#### Summary

This paper suggests an approach for the cost evaluation of discrete analysers.

It is believed that technologists faced with a purchasing decision would find great benefits in performing similar evaluations. Having done so they are then in the position to decide whether the cost in each case can be supported by technical and operating capabilities.

While actual cost savings cannot be given here, as they are applicable to the laboratory studied only; the percentage reduction in costs have been included to show the extent to which savings are possible.

#### Introduction

The study arose from a request from the Clinical Biochemistry Laboratory at Green Lane Hospital.

The Lab uses a SMA 12/60 and two single channel Auto-Analyzers (Technicon Mark 1) to perform high volume tests and performs manually those tests less frequently called for (and those not conducive to performance on an Auto-Analyzer).

The system works well with high quality and timely reports being produced. Workloads are not increasing at a dramatic rate and although being below the recommended staffing establishment the Lab is able to handle all that is asked of it.

However, it was noticed that the ratio of costs to tests appeared to be proportionately higher at Green Lane Hospital than in other Laboratories, particularly those that possessed a discrete analyser. It was wondered if this was due to the greater efficiency of the discrete analyser. If it was, could the purchase of a discrete analyser for Green Lane be justified, or would the initial capital cost outweigh future savings in operating costs?

The Management Services Section of the Auckland Hospital Board was asked to investigate. Specifically, to investigate the need for the provision of a discrete analyser; to study the impact of the purchase on workloads, staffing and costs; and to recommend a course of action. **Materials and Methods** 

The first step was to identify what the discrete analyser would be used for. To this end we asked the Laboratory to identify, from the tests currently being performed, those which they would prefer to perform on a discrete analyser.

Ten tests were so identified.

We had then to decide how detailed our study would be. Would we cost in accordance with established accounting practice or would we derive an approximate costing based upon the data that was most readily available? We settled on the latter; if a discrete analyser could not-be justified using an approximate costing then it probably could not be justified using an exact costing.

Thus some of the figures and costs that were to follow were based upon averages and 'professional' estimates rather than an exact value. (For example: average labour cost, average reagent cost, average number of tests/sample). While this might mean that actual dollar amounts would differ from those of the study, the percentage differences and rankings would be about the same as all approximations were applied 'across the board' and not selectively in a manner which would advantage some areas and disadvantage others.

The study then proceeded firstly to find the annual cost of performing the 10 tests under present methods, secondly to find the annual cost using a discrete analyser. Comparisons would then be possible and a recommendation could be made.

Annual Cost of Performing Ten Tests using Present Methods

We had the following data available:

- (i) Statistics that the Laboratory keeps for returns to the Health Department.
- (ii) The methodologies employed within the laboratory including chemistries and equipment used.
- (iii) The cost of reagent kits, and the cost of chemicals used to make up reagents.
- (iv) The cost of other consumables expended in the performance of the 10 tests.
- (v) The number of staff within the department, their salaries and conditions of appointment.
- (vi) The Canadian Workload Unit values for each of the 10 tests.

The laboratory statistics enabled us to calculate the average number of tests per sample and samples per annum.

The methodologies employed, the costs of

reagent kits, and the costs of chemicals enabled us to calculate the cost of reagent per test. Some difficulty was experienced however and eventually one of three methods was used, the precise choice being determined by the data available.

Either:

- The cost of reagent per litre (say), was divided by the volume of reagent used per aspiration, or
- (ii) The amount spent on a chemical within a given period was divided by the number of tests using that chemical within the same period, or
- (iii) The cost of a reagent kit was divided by the number of tests that could, from experience, be obtained from it.

Further allowances were made for batch sizes, the number of standards, blanks and controls per batch, reagent life and so on. Where reagents were manufactured within the department the cost of cheaper materials, such as distilled water, was ignored. So were the cost of overheads; heating, lighting, power and labour. In the end however, we obtained a reagent cost per test which appeared reasonable although it was probably understated. This was considered an advantage as it made it harder for the discrete analysers to compete.

A similar exercise was carried out to find the cost of consumables per test.

As no one person consistently performs one test, the average salary within the laboratory was used to cost labour. Adjustments were made to take into consideration annual leave, sick leave, statutory holidays and idle and personal time. No allowances were made for overtime or penal rates. Applying the labour cost so determined to the Canadian Workload Unit value for each test enabled us to obtain the cost of labour per test.

The data accumulated in this manner were then combined into cost per test, cost per sample and cost per annum.

### Annual Cost of Performing Ten Tests Using Discrete Analysers

The collection of data for this part of the study proved easier than in the earlier part. We sent questionnaires to each of the major suppliers of discrete analysers within New Zealand. In most cases the response was full and the Companies concerned went to every effort to accommodate us. The questionnaires requested costs, usage and questions of an accounting nature as well as technical questions. Replies are summarised in Table VII. (We must add that all replies were treated as fact but where two or more conflicting answers were given—i.e., between what was shown in the brochure and what was given in the reply—the less favourable data were used).

At the same time the cost of discrete analyser reagent kits, and the volume of reagent in each kit, was obtained from a number of sources including Hoechst, Smith Biolab, Roche, Syva and Beckman. Again the companies were most helpful. These costs and volumes were averaged to give an average cost of reagent per 100  $\mu$ l.

The data obtained in this manner, plus the statistical data, and the cost of labour established earlier, were then combined to derive a cost per sample for each test.

#### Replacement of Existing Equipment

It was known that some existing items of equipment would require replacement within the near future, due to the continuous use to which they were being subjected. Were, however, a discrete analyser purchased, the use of existing equipment would drop dramatically, so much so that replacement would no longer be necessary.

This being the case, and the Laboratory said it was, then the cost of a discrete analyser can be reduced by the amount saved by non-replacement of existing equipment. In effect the Hospital Board was committed to an expenditure of about \$9,000 whether it purchased a discrete analyser or not. Consequently the cost of a discrete analyser for the purposes of this study was treated as the *additional* cost over \$9,000. (Actual cost less \$9,000.)

#### Discrete Analysers

It was apparent from the replies received to the questionnaires that the cost of each discrete analyser was significantly affected by the degree of 'intelligence' that it had. For our purposes a lack of 'intelligence' was not considered to be a barrier to consideration. Consequently both analysers with microprocessors and without have been considered.

It was agreed that servicing would be done by the Laboratory and therefore the cost of service contracts was not included in the final costings.

#### Results

#### Reagents

Substantial savings in reagent costs will result from the use of a discrete analyser (Table I). The greatest cost saving would be made by the Multistat III. 
 Table 1

 Annual Reduction in Expenditure on Reagents as a Result of the Purchase of a Discrete Analyser

ABBOTT	ABBOTT	MULTISTAT	CENTRI- FICHEM
ABA 100	VP	III	300 400
72%	86%	90%	80% 80%

#### Other Consumables

Savings will also occur in the expenditure upon other consumables (Table II). There was however, one exception—the Multistat III, where expenditure on consumables would increase. This can be directly attributed to the cost of the disposable rotor.

The greatest saving in consumables was made by the Centrifichem 400.

 Table II

 Annual Reduction in Expenditure on Other Consumables as a Result of the Purchase of a Discrete Analyser

ABBOTT ABBOTT		MULTISTAT	CENTRI- FICHEM		
ABA 100 63%	VP 63%	III (1%) Increase	300 46%	400 77 <i>%</i>	

The Centrifichem 400 has a superior microprocessor to that of the Centrifichem 300 and is therefore able to produce more concise reports. This results in a lower consumption of paper. Thus, while the Centrifichem 300 can only save 46% of present 'other consumables' costs, the Centrifichem 400 is able to save 77%. *Labour* 

Savings will occur in the expenditure upon labour *directly* employed in the performance of the tests. Ultimately a reduction in staff numbers might prove possible.

#### Table III

#### Annual Reduction in Expenditure Upon Direct Labour as a Result of the Purchase of a Discrete Analyser

ABBOTT	ABBOTT	MULTISTAT	CEN FICI	TRI- HEM
ABA 100	VP	III	300	400
98%	98%	66%	69%	69%

There is a lower Canadian Workload unit value

for tests performed on the Abbott equipment than on other analysers. When this is used to compute labour costs the savings produced, over present methods, is correspondingly greater. Hence a saving in labour costs on the Abbott equipment of 98 percent as opposed to 66 percent and 69 percent on the other equipment.

Total Savings in Operating Costs

The cumulative effect of the cost savings indicated above enables a substantial portion of each dollar presently spent on operating costs to be used elsewhere.

#### Value for Money

Operating Cost Savings must however be considered in conjunction with the initial capital outlay. Which discrete analyser would provide the best value for money? To determine this we used two ratios from Investment theory:

(i) PAYBACK PERIOD—the ratio of the initial fixed investment over the annual cost savings. This would indicate how long it would take to recoup our initial capital outlay in operating cost savings (Table V).

#### Table V Pay Back Period (Months) for Each Discrete Analyser

ABBOTT	ABBOTT	MULTISTAT	CEN FICI	ITRI- HEM
ABA 100	VP	III	300	400
5.9	12.7	14.8	11.6	22.9

The Abbott ABA 100 had the shortest Payback period for the non-'intelligent' analysers, and the Abbott VP had the shortest Payback period for the 'intelligent' analysers.

(ii) THE PROFITABILITY INDEX—the ratio of the present value of the future cash savings over the initial outlay. What savings will be accumulated over the life of the analyser, and how do these savings relate to the initial capital outlay? The suppliers had in most cases recommended a depreciation life of 10 years. We considered that because of obsolescence seven years would be a more reasonable figure. Thus for this comparison cost savings over the next seven years were used (Table VI).

The ABBOTT ABA 100 had the highest profitability index.

ABBOTT	ABBOTT	MULTISTAT	CEN FICI	TRI- IEM
ABA 100	VP	III	300	400
9.0	4.2	3.6	4.6	2.3

## Table IVDistribution of Each Dollar

	AT	ABBOTT	ABBOTT	MULTI-	C	ENTRIFICHEM
	PRESENT	ABA 100	VP	STAT III	300	400
REAGENTS	52%	14%	7 %	5%	10%	10%
<b>CONSUMABLES</b>	2%	1 %	1 %	2%	1.5%	0.5%
LABOUR	46%	1 %	1 %	16%	14.5%	14.5%
SAVING	NIL	84%	91%	77%	74%	75%
	1.00	1.00	1.00	1.00	1.00	1.00

# Table VII Discrete Analyser Characteristics

1.	Model	Abbott ABA Abbott VP	Multi-Stat III	Centrifichem 300 Centrifichem 400	Vitatron PA 800
2.	Supplier	Abbott Laboratories	Sci Med	Roche	Smith Biolab
3.	Cost	ABA = \$20,000 VP = \$47,000	\$46,400	300 = \$35,000 400 = \$70,000	\$42,000
4. 5. 6. 7.	Delivery Warranty Depreciation Service Contract	4-5 Weeks 12 Months 10% 10 years \$4,000	6-8 Weeks 18 Months 8 years \$4,000 or (\$18.00/hr)		
8. 9.	Training Where?	Available on site	Available on site/ Dunedin	Available on site/ Australia	
10.	No Units in NZ	10	4	7	
11.	No. of Components and size	1 Unit 3' × 3'	2 Units 4' $\times$ 2'	2 Units 3' × 2'	2 Units 5' $\times 1^{2}/3'$
			$1\frac{1}{2} \times 2$	$1\frac{1}{2} \times 2$	
12.	Mounting	Tabletop	Trolley bench	Floor bench	Bench top
13.	Special services	Power 220V 50HZ Room temp 20 °C- 35 °C Humidity 10-85%	Power 230V 50HZ Room temp 15 °C- 35 °C	Power 220V 50HZ	Power 220V Room temp 15 °C-35 °C
14.	Warm-up time	15 mins	15-30 mins	5 mins	
15.	Standby Facility		Yes	Yes	
16. 17.	Rotor/Carousel Disposable/ Re-Use	Carousel Disposable	Rotor Disposable	Rotor Re-Usable	Disc Re-Usable
	Load Method	Manual	Automatic	Automatic	Automatic

18. 19. 20. 21. 22	Cost Capacity Operating Speed Max Speed Washable?	\$40.00/Box × 60 32 NA NA Sometimes	\$1.00 ea 20 1000 RPM 4000 RPM Sometimes	NA 30 1000 RPM 1000 RPM Ves	18 Ves
<ol> <li>23.</li> <li>24.</li> <li>25.</li> </ol>	Auto/Manual Wash time Wash amount	Manual	Accessory under development	Automatic 3 mins 70 mls	Automatic
26.	Wash type	Distilled Water	Distilled Water	Distilled Water and Methanol	Distilled Water
27.	Priming Req'd	Yes	No	No	
28. 29.	Auto/Manual Time	Automatic 30 Seconds	NA NA	NA NA	
30.	Rotor Load Time	NA	5 mins/rotor	4 mins/rotor	
a. b.	Sample Volume Reagent Volume	$2.5-25.0\mu$ l ABA = 500 VP = 250	3.0-20.0μl 150-250μl	5.0-50.0µl 250 or 350µl	5.0-100.0μl 300-800μl
c.	System Thru-put	ABA = 100-150/hr VP = 232-465/hr	120-250/hr	120-360/hr	100-180/hr
d.	Samples/rotor	28-31 samples	16-17 samples	28 samples	NA
31.	Pipettes?	No	Yes	Yes	
32.	Disposable/	NA	Re-Usable	Disposable	
33.	Re-Use Cost?	NA	(2-3 years) \$10.00	\$7.12/box 200 (Use 1 per rotor)	
34.	Cuvettes?	No	Yes	No	Yes
35.	Disposable/	NA	Re-Use	NA	Re-Usable
	Re-Use		(1 per month)		
36.	Cost?	NA	$12 \times $1.00$ pa	NA	
37.	Wash needed?	NA	Yes	NA	Yes
30.	The Control		Automatic		Automatic
39. 40	Temp Control	waterbath + 0.1°C	Intra-Red	Air-bath	+ 0.00 %
40. 41	Temp Range	I U.I C	$\pm 0.1^{\circ}C$	±0.1°C	
42.	Temp settings	25 °C, 30 °C, 37 °C	2 settings your choice	25 ℃, 30 ℃, 37 ℃	25 °C, 30 °C, 37 °C
43.	Lamp Type	Tungsten Halogen	Quartz Halogen	Tungsten Iodide	Quartz-Iodine
44.	Lamp Life	6 months	2-3 years	5 years	
45.	Lamp Cost	\$4.20	\$68.00	NA	
46.	System Used	Bichromatic	Bichromatic	0 Eiltore	
47	v Range	330-750 nm	320_690 nm	290-720 nm	330-650 nm
48.	Bandwidth	5 nm	10 nm	12 nm	550-050 mm
49.	Data Points	330	12		
	Readings	_	32 per data point	8 per data point	
50.	Printout	Yes	Yes	Yes	Yes
51.	Paper Type	Plain	Plain	Plain	
52.	Paper Cost	\$0.80	\$7.00	\$2.50	
53.	Paper Usage	9 inch/test	9 inch/test	-	
54.	Paper Length	200 It	Nono	roll every two days	
55.	Other Displays?	ABA = None	None	8 × 8" screen	
56.	Computer Interface?	VP = Yes and Built in	Yes	Yes	

57.	Built in/Extra	ABA = No lo	Extra	Extra	
58. 59.	No. of Reagents Add after premix	2	2 1	2 None	
60.	Methodology	End Point Kinetic Rate Trigger reaction	End Point Kinetic EMIT	End Point Kinetic Recall	End Point Kinetic EMIT
61.	Accessories	Nephelometry	Fluorometry	Computer for Patient Reports	
70.	Other Features	<ol> <li>Abbott ABA</li> <li>Has no micro- processor. Must therefore be set up for each test.</li> <li>Performs simple calculations only</li> <li>Further calcula- tions required after results are printed.</li> </ol>	<ol> <li>Multi-Stat III</li> <li>a. Programmed procedures</li> <li>contained on cassette.</li> <li>b. Test parameters may also be entered manually.</li> <li>c. Can make up own procedures and store on cassette.</li> <li>d. Red flags abnormal results.</li> </ol>	(1) Centrifichem 300 a. Test parameters must be entered manually each time.	<ul> <li>Vitatron PA 800</li> <li>a. Uses two disposable re- agent containers.</li> <li>b. Uses dispos- able sample vials.</li> <li>c. Contains pro- grammable calculator.</li> <li>d. 15 pre- programmed</li> </ul>
		<ul> <li>(2) Abbott VP</li> <li>a. Micro-processor goes through self checking routines.</li> <li>b. Gives error, fault messages.</li> <li>c. Contains program- med procedures called up by code.</li> <li>(2) Abbott VP</li> <li>d. Allows test parameters to be entered manually, although there is no means of storing them.</li> </ul>		<ul> <li>(2) Centrifichem 400</li> <li>a. Pre-programmed test parameters for 23 tests. Called up by code.</li> <li>b. Data can be stored and recalled until start of next test.</li> </ul>	standard appli- cation cards supplied with unit.

#### Discussion

Having completed a cost evaluation the laboratory's technical evaluation becomes a lot easier. The technical features of each machine can now be compared against the capital and operating costs and the question asked: Are these features worth the additional cost? The final decision must be the laboratory's but having made their choice they should be able to justify it on both technical and cost grounds. Management can then be approached with a proposition: that in return for a discrete analyser the laboratory can accept a cut in their operating

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#### budget of X dollars per annum.

I believe that not only will Management look favourably upon such a suggestion but funds will be available for the purpose.

#### Acknowledgment

I would like to thank John Powell and Eric Johnston for their assistance and also all those people and companies that provided information.

#### An Evaluation of the Chemlab Microprocessor-Controlled 8-Channel Continuous Flow Analyser

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Laboratory Services Department, Dunedin Hospital.

Received for Publication, October 1979

#### Introduction

Experience of the Chemlab 8-channel analytical system (Chemlab Instruments Ltd, 129 Upminster Road, Hornchurch, Essex, UK) which has been in routine use for one year is described. Evaluation procedures followed that recommended by the Laboratory Equipment and Method Advisory Group (1969).<sup>1</sup>

The Chemical Pathology Laboratory in late 1978 required the replacement of an ageing Auto-Analyzer I system used for a 5-channel liver function screen, calcium and phosphate, uric acid, iron and TIBC.

The Chemlab system was chosen for the following reasons.

- Use of modular components to allow flexibility in methodology.
- Use of widely used and evaluated continuous flow methods.
- Analytical rate in excess of 60 samples/hour elimination of sequential channel phasing.
- Automatic standardisation, calibration, and drift correction.

Automatic error detection.

Printout of results by channel and grouped for each patient.

Ease of operation.

Low operating cost.

Low capital cost.

The last item determined the choice, the cost of Chemlab analytical system was approximately one half of a comparable continuous flow system.

The system purchased consists of one sampler, three peristaltic pumps, seven analytical cartridges, two flow through colorimeters (5-channel), four two-pen recorders, and an 8-channel data processor and printer. For use in the laboratory the system has been augmented with a Technicon Sampler II, and one Pump II. In addition an existing channel of Auto-Analyzer I has been interfaced to the data processor by means of a retransmitting potentiometer. The following modules are in use. A view of the complete system is shown in Figure 1.

#### Sampler

Description

The sampler incorporates a unique probe washing action which is claimed to improve the wash pattern considerably.

The probe washing device consists of a washing chamber through which the prove moves vertically. A sliding 'O' ring on the probe alternately opens and closes on the outlet for the water reservoir.

Suction is applied by means of a manifold pump tube to remove excess water from the probe as it passes through the chamber. A special probe in which three tubes are placed enables the sample to be aspirated by separate channels.

The sampler is constructed of moulded plastic and the controls are mounted on a recessed panel on the front of the sampler. These control knobs enable the user to select sample and wash times ranging from 0-120 seconds.

An Autostop switch allows the sampler to be automatically switched off after the last sample has been aspirated.

A PVC turntable with 40 places is supplied and can be substituted for Technicon II sample trays.

#### **Pumps**

Two models are in use, one has capacity for 15 pump tubes, the other 30 tubes. The pumps have constant speed motors which are fan cooled. The pressure plate assembly is mounted on the top of the pump. A drip tray is fitted internally with an outlet tube which discharges through the base should leaks occur.

#### **Colorimeters**

The colorimeters employ glass fibre optic light guides using quartz iodine lamps. Interference filters are used in the range of 400 nm to 880 nm. Up to five independent colorimeter channels can be fitted to each colorimeter module. Each colorimeter channel has two detectors one for the sample cell and the other for the reference beam. The photoresistors used for the detectors are linked through a bridging circuit and produce a linear output. The standard module incorporates a single 15 mm flow cell with a fixed reference cell. A flow cell can be fitted in front of the reference cell if required for automatic blank correction. The colorimeter is suitable for connection to either potentiometric or amperometric recorders.

The baseline is adjusted by means of a shutter control lever. A fine control knob is used for small adjustment.

A gain control is used to set the peak height of the standard at the desired level.

Analytical Cartridges and Methods Used

Chemlab cartridges constructed of Auto-Analyzer II components are used in the following test methods:

	Technicon method
Albumin	SE4-0030FDK
Protein and Blank	AAII-14
Bilirubin and Blank	AAII-18
Alkaline Phosphatase	SMA-12/60
Aspartate Transaminase	AAII-10
Calcium	CMII-021-01
Phosphate	AAII-04

One channel of Auto-Analyzer I was interfaced through a retransmitting potentiometer from the Bristol Chart Recorder. This channel is alternately used for uric acid, iron and TIBC. *Recorders* 

Four two-pen 'Linseis' potentiometric recorders with 120 mm wide chart paper are used to continuously monitor each channel and as a fail-safe facility.

The output from the colorimeter is directly connected to the recorders in parallel with the data processor. The recorders have an electronic zero control, and selection switch to enable various volt and millivolt ranges to be selected. The variable potentiometer position is used to enable the chart recorders to be calibrated in parallel with the data processor. Variable paper speeds can be selected ranging from 1 to 100 mm per minute or hour.

#### 8-Channel Data Processor

The data processor is a free-standing unit that may be connected to a maximum of eight separate analytical channels. Each channel operates independently and may be used with Auto-Analyzer I or Auto-Analyzer II type systems. The data processor is controlled by an Intel 8080A microprocessor with an application programme on PROM (programmable read only memory) to determine peak heights, calculate data, and determine printing formats.

A small amount of memory is used to store results prior to printing. The 20 column printer can print calibrated values of each sample as it is encountered, batch of samples from each channel and grouping of channel result for each patient.

The data processor is programed by entering numeric codes via a keyboard and depressing function buttons on the control panel.

The following parameters can be selected and changed for each channel:

Value of the top calibrating standard.

Sampling rate.

Number of standards.

Interval of drift specimens.

Interval of wash.

The percent value of each of the standards. (Eight maximum).

First sample sequence number.

Number of specimens before the first drift.

Method of peak picking and printing of results. Window width.

Whidow width.

Minimum height of first peak.

Peak drop value.

Number of data points to be averaged.

With the power left on, the parameters entered are retained. A four digit decimal display shows the value of each parameter as it is entered. In addition this display can be selected to show the digital volt meter reading of each channel and is used when setting baselines and calibration. When a channel is switched on line the status of the channel is displayed. The sampling rate found and the type of peak encountered is shown in numeric code.

Two types of peak picking algorithm are used depending on whether AAI or AAII peaks are used. A sliding seven point second order polynominal curve fit is used to determine the peak height from the data points collected from the point of inflection of the peak and the points in the specified peak window. The programme also includes noise rejection.

A channel is switched on line by depressing the channel 'on line' button. The peak picking programme is activated and scanning of the baseline started. The baseline value is printed when a sample wash cycle has completed along with the parameters selected by the operator for that channel.

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Figure 1.—Shows general arrangement of modules with programmer centre right.

LIVER FUNCTION TESTS					
The Unit of the	Altumin	Protein	Biltrubin	Alk. Phos.	A.S.T.
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Figure 2.—Patients' data (accession list) aligned with printout of results for reporting.

When the synchronising peak is found the actual centre of the peak is calculated. For AAI peaks this is the peak centre, for AAII peaks it is the point of inflection of the plateau. From this the number of data points falling outside the peak window are determined and the interval for subsequent peak detection calculated. For satisfactory operation the sampling rate detected by the first two peaks must be within 10% of that specified by the operator.

The peak height of all the standards and the baseline are used to determine the polynominal curve fit. Linear interpolation is used if only one standard is used.

The drift specimens placed immediately after the standards are used to establish the reference drift value. The drift specimens placed at defined intervals are used to determine the drift of each sub-batch. Linear correction is applied to the subbatch with a further correction to the reference value if required.

The system detects a number of error conditions and prints numeric codes when these are detected. Two types of error condition can be detected. Those associated with discrepancies between operator selected data and the capability of the data processor, and errors detected in peak analysis.

#### Operation

The present configuration consists of three analytical systems:

A five-channel liver function group consisting of albumin, bilirubin and blank, protein and blank, alkaline phosphatase, and aspartate transaminase. In this configuration an Auto-Analyzer II sampler with a timing modification and a Gilford 300N Colorimeter at 340 nm are used. A sampling rate of 72/hour with a sample to wash ratio of 2.3/1 is used.

Two channels calcium and phosphate. A Technicon Pump II is used. A sampling rate of 80/hour is used with a sample to wash ratio of 5.5/1.

A single channel of Auto-Analyzer I interfaced to the data processor as previously described.

Worklists are manually prepared from patient request forms by filling an appropriate formulated worklist. The worklists define the cup positions of patient and drift specimens and the format allows the printouts to be pasted onto the worklist. The patient result data is outlined to facilitate transcription of the results onto the report forms. An example of the liver function worklist is shown in Figure 2.

#### 3. Results and Discussion

#### Sampler

The CS 80 Sampler is currently used on the twochannel calcium/phosphate with a single probe and stream splitter. The probe on this type of sampler requires careful adjustment. The wash water is used to lubricate the probe and sliding 'O' ring. With the triple probe, lubrication was a problem, the probe often sticking after several hours' continuous use resulting in short sampling. For this reason the CS 80 Sampler was replaced on the 5-channel liver function analyser by a Technicon II Sampler and stream splitters which prove just as effective.

Figure 3 shows the wash characteristics of the two samplers in use. Carryover is determined as recommended by the Laboratory Equipment and Methods Advisory Group 1969.

 $K = \frac{b1 - b3}{a3 - b3}$ 

#### Pumps

The CPP 15 and CPP 30 pumps are similar in design to AAI pumps having the same number of rollers in contact with the platen assembly. The pressure adjustment is simply performed by moving two locking nuts on the pump head. The tension on the chain drive can be altered by moving the motor on adjustable mounts. One pump had to be replaced after one year when it developed irregular pumping consistent with the roller chain having been stretched. Adjustments to correct the fault were not possible.

#### Manifolds

With the manifolds in use the bubble patterns are satisfactory.

Colorimeters

The 15 mm flow cell used in Chemlab colorimeters is similar in design to AAII flow cells and produce comparable results. The use of a flow cell for blank analysis in the reference photocell of each colorimeter module is a useful refinement. The careful phasing of total and blank channels is required however to avoid noisy traces, and obtain correct results.

The use of fibre optics for the light guide has a serious disadvantage in that light at 340 nm cannot be transmitted. The manufacturers are aware of this defect and undertook to supply a modified light source using quartz fibre optics. This has not been done although work is progressing on the design of this modification.

In order to perform AST estimations a Gilford 300 N Colorimeter with a flow cell modification has been used. This however, has not proved satisfactory because a blank channel is necessary to avoid false low results found on patients taking

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Figure 3.—Comparative scans for calculating carry-over.

compounds which absorb at 340 nm, e.g., metronidazole. This is to be the subject of a separate report.

The method evaluation of analytical cartridges were determined using the following criteria: analysis of standard profiles<sup>2</sup> to determine ideal, sample rate, interaction experiments, linearity, accuracy and precision. Examples of peak profiles obtained with some of the chemistries are shown on Figure 4. All the methods using AAII cartridges and the Chemlab colorimeters proved linear in the clinical ranges required. One point calibration is used on all AAII methods using one standard and the baseline with linear interpolation to the highest value with that number of significant digits, i.e., Figure 4.-Peak Profiles



Figure 4.-Illustrates the peak profiles obtained for the liver function tests.

if 86.0 was used 99.9 would be the maximum calculated value. This has proved a problem because it is not always possible to obtain calibration serum with values suitable for one point calibration on the Chemlab. For example values of less than 99 are suitable for albumin and protein but for alkaline phosphatase, aspartate transaminase, and bilirubin values in excess of 100 are required.

The procedure of using different sera for some assays is therefore required with the inherent problems.

On the AAI channels uric acid and iron, multiple standards are used and third order interpolation used has been satisfactory.

Precision statistics of all methods before and after the implementation of the 8-channel system are shown on Table I.

#### Chart Recorder

The chart recorders are an integral part of the

system and when calibrated in parallel with the data processor enable the operator to quickly set the baselines and monitor each channel. In the event of data processor failure the tracings although small enable the results to be read from the chart paper.

#### Data Processor

The data processor in the present configuration has one serious deficiency in that results cannot be printed in groups for each patient. This is because the memory of the data processor is exceeded when all channels are on line, due to the fact that the time delay between the fastest and slowest channel on the liver function profile is nine minutes. The manufacturers claim extra memory cannot be added to the existing chassis because of design implications.

The printout currently obtained which shows each peak as it is encountered, plus batches for each channel, eliminates the major clerical pro-

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## Table IAccuracy and Precision

a. Between batch precision (6-month cycles before and after implementation) derived from internal quality control.

	Before		After	
	Avera	-		
	ge		Averag	e
	Level	S.D.	Level	S.D.
Albumin	31	2.02	32	0.68
Protein	70	1.3	69	0.78
Alkaline	102	9.0	149	6.6
Phosphatase				
Aspartate	110	7.0	131	4.6
Transaminase				
Bilirubin	175	3.6	149	5.6
Calcium	2.40	0.034	2.60	0.035
Phosphate	1.6	0.056	1.43	0.033
Iron	24	1.0	22	1.0
Uric Acid	0.40	0.014	0.41	0.014

b. Wellcome group quality control programme (all to date).

	Before		After	
		Pre-		Pre-
	Bias	cision	Bias	cision
Albumin	4.2	3.2	-0.039	0.88
Protein	2.8	1.6	-0.103	1.30
Alkaline	40	8.6	-24.5	13.0
Phosphatase				
Aspartate	20	6.2	25	4.3
Transaminase				
Bilirubin	-1.9	2.5	5.6	3.08
Calcium	-0.038	30.069	0.107	0.070
Phosphate	0.018	30.053	0.007	0.040
Iron	3.2	3.1	-1.14	0.51
Uric Acid	0.005	0.015	0.007	0.009

cedure in continuous flow analysis of chart reading. The specially designed worklists that the batched printouts are attached to have proved useful in the laboratory as a record of results.

The careful selection of first peak criteria is essential for successful operation. The accurate assessment of sampling rate, window width, peak drop value and the number of data points to be used must be critically assessed.

This has proved the most difficult task when methods are first set up. The key to successful operation is analytical methods with good peak profiles and minimal interaction. Occasionally however a swamped peak may result in the data processor going out of synchronisation and data are lost. This occurs most commonly in methods where large variation in peak height occurs, e.g., bilirubin and aspartate transaminase.

Once criteria have been selected problems rarely occur and often synchronisation errors are the result of manifold deterioration. The system therefore ensures that manifold maintenance is of high standard.

One deficiency in the peak picking algorithm is the inability to detect some short samples. We have not been able to fine tune the peak picking programmes to be able to detect short samples. The use of the chart recorders for monitoring purposes is therefore considered essential.

The data processor has rarely been out of action. It was unavailable for one week, the result of a component failure when electronic tests were being carried out on an adjacent Bristol Recorder.

#### **Summary**

In general the system has been well accepted by laboratory staff using it. The modules are simple to maintain and use. The use of the data processor has resulted in a stricter control of procedures which has led to an improvement in the quality control. Two criticisms can be levelled at the system:

- 1. Lack of colorimeter in the uv range.
- 2. Deficiencies in the capability of the data processor.

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#### **Balancing the Quality Control Budget**

J. E. Parker, ANZIMLT, B.Sc.

Chemical Pathology, Diagnostic Laboratories, Dunedin Hospital A paper delivered to the NZIMLT Conference, Auckland 1979

#### Introduction

In any clinical biochemistry laboratory with an adequate quality control system a sizeable portion of the annual budget goes to the purchase of control sera. Automation, by use of an intermittent drift control greatly increases the quantities used and therefore the cost. Faced with the current economic circumstances our laboratory began to examine the possibility of manufacturing some of the material ourselves. We had for some years been obtaining fresh beef blood from the local abattoir for use as a pool and had already established that the sera stored well frozen. Thus we already had a free source of biological material which could be further and better utilised.

#### Method

#### 1. Urea and Electrolytes

These are performed on a Technicon 6-60 which was being calibrated using a commercial control serum, Calbiochem Reference Serum in every tenth position. This had been used satisfactorily and cheaply for a number of years but was now going out of production. Viable alternatives were going to cost in the vicinity of \$150 a week. The levels of sodium potassium and bicarbonate in the beef sera approximated those of the calibration sera, and urea and creatinine were weighed in to bring them up to accepted levels. This pool was then aliquoted in suitable quantities and refrozen. A batch of four litres supplied sera for approximately three months' use which was carefully calibrated against a reference sera. A daily check was run on values by processing one sample of the commercial reference sera. To date there have been no problems of values 'going off'. We did examine the possibility of freeze drying the product but in terms of the time taken and the necessity for very accurate aliquots the idea was discarded.

#### 2. Liver Function Tests

These are performed on a modular discrete continuous flow analyser (ChemLab) with computerised digital output. The values of the sera did not require to be actually determined as the pool is used for drift control only. Nevertheless the levels of the enzymes in particular were required to be in a fairly narrow optimal range. Protein and albumin levels were acceptable but bilirubin, aspartate transaminase and alkaline phosphatase levels required boosting. Bilirubin prepared from bovine gallstones was purchased from Sigma and weighed into the pool at the desired concentration after dissolving in dimethyl sulphoxide. Aspartate transaminase and alkaline phosphatase were prepared by extraction from fresh human liver obtained from the mortuary. Both enzymes are available commercially at relatively low cost from Sigma but could not be obtained in the time we had available.

Extraction of Liver Morton, (19501).

Blood from the liver was first tested for HBs Antigen and found to be negative. The cadaveric liver which had been stored at -70 °C was thawed, washed, and weighed. It was then cut into small pieces and homogenised with an equal volume of ice-cold distilled water in a Waring blender. This was carried out for a total blending time of two minutes in 30 second units followed by a 30 second rest period. Cold butanol at a concentration of 1.5 ml/gm was added gradually with vigorous overhead stirring of the homogenate. After 30 minutes emulsification the homogenate was filtered through muslin and heated to 35 °C for five minutes. The filtrate was centrifuged at 4°C for 65 minutes 600 rpm in a Damon IEC PR-600 centrifuge and the clear aqueous lower layer collected. This enzyme extract was filtered through Watman No. 1 and dilutions made in 0.1 M Tris buffer at pH 7.4. A 1:200 dilution of the extract in the beef pool gave suitable enzyme levels.

As with the urea and electrolytes this pool was frozen in suitable aliquots for daily use. In future we may calibrate this pool for use in calibration but alkaline phosphatase levels will require a further boosting. A commercial alkaline phosphatase has been purchased from Sigma and a small amount only is required to bring the alkaline phosphatase to the required level. Unless the calibration sera has levels over 100 µmol/1 for bilirubin and enzymes the computer will not printout patient results over this value and they require diluting. Values over 150  $\mu$ mol/1 are inclined to swamp the next peak and cause the computer to get out of synchronisation. It has proved difficult to purchase easily a commercial control sera which fills all requirements.

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#### 3. Calcium and Phosphate

These are also performed on the ChemLab and require a drift control in every tenth position. The beef pool proved unsuitable for use as the phosphate levels were unacceptably high. We arranged for therapeutic bleeds to be bled into a dry bag and the sera obtained from these were aliquoted into suitable amounts and frozen for this use. HBs Antigen testing is carried out on the total pool.

#### Conclusion

We estimate that these procedures still under review, will save us at least five thousand dollars a year on control sera while providing an acceptable standard of quality control.

#### Acknowledgments

I wish to express my thanks to Dr P. Hurst for his assistance in the preparation of the liver extract.

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#### National Immunohaematology Proficiency Survey (NIPS) A Summary of Results J. Austin,

Charge Technologist, Blood Bank Taranaki Base Hospital, New Plymouth A. E. Knight,

Charge Technologist, Immunohaematology Laboratory, Dunedin Public Hospital On behalf of the Technical Sub-Committee of the Transfusion Advisory Committee

Received for Publication. October 1979

#### Introduction

The first National Immunohaematology Proficiency Survey (NIPS) was carried out in May 1978, a total of 49 laboratories participating. This total has now reached 60 with laboratories in Fiji and Australia taking part.

The original aim was that all laboratories both public and private where immunohaematological procedures were being performed, should participate. It was felt that this could be instituted in New Zealand and all laboratories would be encouraged to take part. A minimal charge to cover postage would be made to each hospital board.

Results would be published anonymously and only Regional Transfusion Officers would have a key to the codes of the individual laboratories in their respective regions.

A total of four surveys would be carried out annually.

The object of the surveys is to make available to all laboratories an external quality control programme which may bring to light deficiencies in their own internal quality control programme.

It was the recommendation of the Technical Sub-Committee of the Transfusion Advisory Committee, that a brief summary of NIPS be submitted for publication in the Journal annually, in order to reach as wide a readership as possible.

#### Summaries

It was not the intention in these summaries to pass judgment but rather to present the results and for individual laboratories and technologists to be aware of their short-comings.

With this in mind, summaries are presented of the last four surveys; NIPS 3 (November 1978), NIPS 4 (February 1979), NIPS 5 (May 1979), NIPS 6 (August 1979).

#### NIPS 3

(a) Grouping

Sample 018— $A_x(D)$  Positive,  $R_1r$ , Kell Negative.

Sample 019-O(D) Positive,  $R_1R_2$ , Kell Negative.

Sample 020— $A_2(D)$  Positive,  $R_1R_1$ , Kell Negative.

Sample 021-A, (D) Negative, rr, Kell Negative.

Comment: A number of laboratories failed to correctly identify cell 018 as a consequence of misinterpretation of results, wrong results or failure to use anti-A + B (group O serum). Cells 019 and 020 were correctly typed by all participants. Cell 021 was mistyped by one laboratory and a further laboratory made a transcription error.

(b) Antibody Identification

serum 018-contains Anti-A1.

The majority of laboratories (35/37) who attempted the antibody identification were correct. (c) *Crossmatch* 

All laboratories (42) who performed the crossmatch reported the incompatibility between serum 018 and cell 021.

#### NIPS 4

#### (a) Grouping

Sample 022-O(D) Positive (CDe/cde), Kell Negative, P<sub>1</sub> Negative.

Sample 023—O(D) Positive (CDe/cde), Kell Negative, P, Positive.

Sample 024—O(D) Negative (cde/cde), Kell Negative, P. Positive.

Sample 025— $A_2(D)$  Positive (CDe/cde), Kell Negative, P<sub>1</sub> Positive.

Comment: Cell 022 was correctly grouped by all participants. Cell 023 was misgenotyped by one laboratory, three laboratories failed to detect the weak  $P_1$  antigen and one laboratory switched this tube with cell 024. Cell 024 was correctly grouped by all participants, however the cell was not tested for D<sup>u</sup> by eight of the laboratories. Cell 025 was misgenotyped by two laboratories.

(b) Antibody Identification

Serum 022 contains anti-P<sub>1</sub>.

A number of variations occurred in this section of the survey. Some laboratories failed to detect the antibody, others misidentified it and others were unable to carry out the identification due to the condition of their commercial panel upon receipt.

#### (c) Crossmatching

Although all laboratories detected the ABO incompatibility between serum 022 and cell 025 some laboratories had very poor performances with some of the techniques used although other laboratories using identical methodologies found the serum to react strongly with cell 025.

Referee's Comment: "The anti-P<sub>1</sub> is of no clinical significance as it was only reactive below 20 °C and possibly only when using enzyme techniques. In practical terms, incompatibility detected due to this antibody should not have delayed transfusion especially in an urgent situation. Such is the temperature dependence of this antibody that laboratories working in a room temperature above 18 °C may have found that it reacted only with strong P positive cells or they may not have detected it at all."

#### NIPS 5

(a) Grouping

Sample 026— $A_1$  Negative, (rr), Kell Negative. Sample 027—O Positive, ( $R_1r$ ), Kell Negative, *DCT Positive*. Sample 028-O Negative, (rr), Kell Negative.

Sample 029-O Positive, (r"r), Kell Negative.

Comment: Cell 026 three laboratories incorrectly subtyped this cell. One laboratory switched the Rhesus results of this cell with that of 027. Eighteen laboratories failed to carry out a Direct Coombs Test on cell 027 even though the instructions stated that the sample was from a newborn infant.

All laboratories correctly grouped cell 028 and all participants typed all three D negative cells for  $D^{u}$ . Some confusion was caused by the variations used in terminology for the reporting of Rhesus negative typings. All participants correctly grouped cell 029 although one laboratory gave the incorrect shorthand notation for the genotype.

(b) Antibody Identification Serum 026 contains anti-D.

All laboratories who attempted identification detected the anti-D. Some laboratories also detected anti-C and/or anti-E. All were accepted as being correct. The majority of laboratories did not perform titrations of the antibody even though this was from a case of Haemolytic

Disease of the Newborn.

(c) Crossmatching

One laboratory failed to detect the incompatibility between serum 026 and cell 027 by the enzyme technique. Three laboratories found cell 028 to be incompatible by their enzyme technique and two laboratories found cells 028 and 029 to be incompatible by their Coombs technique.

#### NIPS 6

#### (a) Grouping

Sample 030— $A_1R_1^{u}r$ , Kell Negative ss.

Sample 031—A<sub>2</sub>R<sub>2</sub>R<sub>2</sub>, Kell Negative ss.

Sample 032—A<sub>1</sub>R<sub>1</sub>r, Kell Negative ss.

Sample 033-OR<sub>1</sub><sup>w</sup>r, Kell Negative Ss.

Comment: Five laboratories did not perform  $D^{u}$  testing on cell 030 and one laboratory misgenotyped the cell. Four laboratories gave incorrect genotypes for cell 031. One laboratory gave an incorrect genotype for cell 032. Cell 033 was tested for the C<sup>w</sup> antigen by six laboratories and one laboratory made a transcription error in reporting the group of this cell as being group A. (b) Antibody Identification

Serum 030 contains anti-E and anti-S.

Twenty-nine laboratories attempted the antibody identification 18 of which correctly identified the anti-E and anti-S. One referee also found anti- $Wr^a$  to be present.

#### (c) Crossmatching

Cell sample 031 was incompatible with serum 030 due to the presence of anti-E, 12 laboratories

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failed to detect this incompatibility by their enzyme techniques.

Cell sample 032 was compatible by all techniques although four participants found it to be incompatible by enzyme technique, and one by their Coombs technique.

Cell 033 was incompatible by saline and Coombs technique due to the S antigen. Twenty laboratories found it to be compatible by saline room temperature technique and seven made it compatible by saline at  $37 \,^{\circ}$ C. Fourteen participants found this cell to be compatible by their Coombs technique.

Summary

As was stated in the introduction to this article,

it is not our intention to pass judgment but rather to point out to laboratories that errors do occur, errors that may prove fatal.

Laboratories that have a 100% record should not get too complacent—"the best laid schemes, etc."

For laboratories that have made errors, the onus is on them to make corrections and more importantly to be aware of these errors.

It is to the credit of participating laboratories that where the surveys have shown up deficiencies, they have taken steps to improve their performance after consultation either with their Regional Transfusion Officer or another laboratory.

#### A Ten Year Survey of the Efficiency of Anti-Rh Immunoglobulin in the Prevention of Rhesus (D) Sensitisation

Alan E. Knight, FIMLS, ANZIMLT,

Immunohaematology Department, Dunedin Public Hospital. Received for Publication, September 1979

#### Introduction

This paper endeavours to examine the efficiency of this programme with respect to the incidence of anti-Rh (D, CD, DE, CDE) antibodies found at six months following the injection.

The use of anti-Rh (D) immunoglobulin in the prevention of sensitisation of Rhesus (D) negative women post-partum, was introduced in the Otago and Southland regions in early 1969. The control centre for its use and follow-up has been the Blood Bank, Dunedin Public Hospital.

The anti-Rh (D) immunoglobulin is prepared by the Commonwealth Serum Laboratories, Melbourne, from raw material of suitable titre and specificity in part supplied by the New Zealand Blood Transfusion Service. During the period under survey the standard dose has been  $250 \mu g/ml$  of anti-D.

The product is issued to all Rhesus (D) negative women who are either (a) delivered of a Rhesus (D) positive infant, or (b) following an accidental or therapeutic termination of pregnancy, or (c) who are of child-bearing age who have been accidentally transfused with Rhesus (D) positive blood.

#### **Procedure**

The criteria for the product's use has been as laid down by the Transfusion Advisory Committee.

- (a) The mother must be Rhesus (D) negative.
- (b) She should have no detectable Rhesus (D) an tibodies at the time of delivery.
- (c) The infant should be Rhesus (D) positive.
- (d) The product should be issued within 72 hours of delivery.

The following tests are therefore performed at the time of delivery even though the mother may have been fully tested shortly beforehand.

*Mother:* ABO and Rh grouping including D<sup>u</sup>. Screen for atypical antibodies by two-stage enzyme and automated LIS (low ionic strength solution) technique.

*Baby:* ABO and Rh grouping including D<sup>u</sup>, anti-CD and anti-DE (if apparently D negative).

Direct Coombs test.

A Kleihauer test is also performed on the maternal sample. This is an acid elution test which enables the foetal erythrocytes (containing HbF), to be estimated.

Provided all the above criteria are met, the product is issued. If the Kleihauer test reveals the presence of foetal cells in numbers greater than 20 per low power field, a second dose is issued and the Kleihauer repeated 24 hours later to check on the efficiency of the two doses. During the period under review a total of 4,998 doses of anti-Rh (D) were issued. Accompanying each ampoule is a questionnaire which is completed with the time of delivery, time of injection, patient's home address and the name and address of her general practitioner, and then returned to the Blood Bank. A total of 192 questionnaires were not returned.

After a period of six months has elapsed following the injection, a letter is sent to the patient with a copy to her general practitioner requesting that a blood sample be collected and forwarded to the Blood Bank. It is the policy of the department not to send these follow-up letters to patients who have had a termination of pregnancy. Occasionally the patient would have changed address following her delivery and therefore no follow-up specimen is obtained. A total of 1,799 injections were not followed up at six months because of the above reasons, i.e., questionnaire not returned, termination of pregnancy, or changed address.

Upon receipt of the six-month sample it is regrouped both ABO and Rhesus and the serum examined for the presence of atypical antibodies by a two-stage enzyme technique and by the low ionic strength autoanalyser. Any such antibodies detected are then identified.

Of the remaining 3,007 injections that were followed up at six months, a total of 76 were shown to have atypical antibodies where none were detected at the time of delivery.

#### **Results and Discussion**

The specificities of the atypical antibodies are shown in Table I.

The incidence of anti-D represents 0.83% of those tested at six months or 0.5% of the total number of anti-Rh (D) immunoglobulin doses issued.

An elaboration of the 25 cases is provided in Table II.

It will be noted that in six cases, 2, 7, 13, 21, 23 and 25 (Table II), there was ABO incompatibility between the mother and her infant. Therefore the mother's plasma would contain naturally occurring anti-A or anti-B to sensitise the foetal cells before they, the foetal cells, would have a chance to sensitise the mother, although apparently this did not occur.

In none of the cases listed was the interval between delivery and injection greater than 72 hours. The minimum interval was four hours and the maximum 64 hours.

In three instances, cases 2, 14 and 18 (see Table II) the appearance of Rhesus antibodies occurred six months after the second recorded delivery. In each case anti-Rh (D) immunoglobulin had been given previously and no atypical antibodies were detected at the six months follow-up.

In none of the 25 cases listed was it recorded that a large foetal/maternal haemorrhage had oc-

curred, warranting the use of a second dose of anti-Rh (D) immunoglobulin.

SpecificityIncideAnti-D (D or CD or DE)2	nce 5 5
Anti-D (D or CD or DE) 2	5 5
	5
Anti-P <sub>1</sub> 2	
Anti-A <sub>1</sub>	9
Anti-H	4
Anti-Le <sup>a</sup>	4
Anti-Le <sup>b</sup>	1
Anti-Kell	2
Anti-E	2
Anti-M	1
Anti-Lu <sup>a</sup>	1
Anti-C <sup>w</sup>	1

#### **Table II**

Interval Detwoor

			Interval Detrices
			<b>Delivery</b> and
	Mother	Baby	Injection in
Case	ABO and R	iesus Group	hours
1.	A Negative	O Positive	24
2.	O Negative	A Positive	28
3.	<b>B</b> Negative	O Positive	49
4.	O Negative	O Positive	21
5.	A Negative	A Positive	57
6.	A Negative	O Positive	23
7.	O Negative	A Positive	48
8.	O Negative	O Positive	5
9.	A Negative	A Positive	31
10.	A Negative	A Positive	22
11.	A Negative	O Positive	44
12.	A Negative	A Positive	11
13.	A Negative	<b>AB</b> Positive	27
14.	A Negative	O Positive	35
15.	<b>AB</b> Negative	?	4
16.	O Negative	O Positive	18
17.	A Negative	O Positive	24
18.	A Negative	A Positive	26
19.	O Negative	O Positive	37
20.	<b>B</b> Negative	O Positive	44
21.	O Negative	A Positive	32
22.	O Negative	O Positive	40
23.	O Negative	A Positive	64
24.	O Negative	O Positive	7
25.	O Negative	A Positive	30

It appears therefore that sensitisation of the mother by the Rhesus (D) antigen has occurred in spite of:

(i) ABO incompatibility between mother and infant

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- (ii) there being no time delay between delivery and injection
- (iii) there being no large foetal/maternal haemorrhage resulting in only partial removal of the foetal cells from the maternal circulation by the use of the anti-Rh (D) immunoglobulin.

Since this paper was first drafted two papers<sup>1,2</sup>, have been produced on the same topic, i.e., prevention of Rhesus sensitisation. In each the authors make mention of the incidence of Rhesus sensitisation even after the prophylactic use of anti-Rh (D) immunoglobulin but suggest that the reduced dose of  $100\mu g$  is sufficient for protection.

The standard dose now used throughout New Zealand is 125  $\mu$ g/ml. This is reduced from the 250  $\mu$ g/ml dose used during the period of this first survey. If another survey is conducted after a further period of 10 years, it will be interesting to see if the incidence of Rhesus(D) sensitisation has increased.

#### REFERENCES

- Proceedings of the McMaster Conference on Prevention of Rh Immunization 28-30 September 1977, (1979), Vox Sang, 36, 50.
- 2. Simonovitis, 1. and Borocz, Irene, (1979), Vox Sang, 36, 21.

#### The Suva Steamer: An Example of Alternative Technology

#### J. A. Samuel, Dunedin

#### (Formerly Superintendent of the Public Hospital Laboratory, Suva, Fiji).

Medawar has said 'It's no use looking to scientific "papers", for they not merely conceal but actively misrepresent the reasoning that goes into the work they describe . . . Only unstudied evidence will do—and that means listening at a keyhole'  $(1967)^1$ .

I had intended to present the 'Suva Steamer' (Figure 2) in its final form—it would have been a brief, simple paper—even elegant.

In defence to Medawar and as a lesson to all who hope to 'write papers' I will describe the evolution of a piece of equipment which, in its final form, turns out to be surprisingly simple and effective.

In 1953, a steamer for controlled temperature below 100°C was described, (Samuel, 1953)<sup>2</sup>.

This was a valiant attempt to adapt an existing steamer of antiquated design. The old steamer needed urgent repairs and the writer's status at that time was not such that he could involve his department in expense for a completely new instrument based only on a good idea.

In Suva in 1955 there was an imperative need to inspissate large batches (about 400 universal containers) of egg medium, so a new design was evolved. Here, I had both the duty and the authority to produce something that worked.

The concept in reference<sup>2</sup> of a plenum chamber feeding a steam/air mixture between trays of media was used in a more logical form:  $4\frac{1}{2}$  kw heated the water in the bottom of the plenum chamber, controlled by a thermostat in the upper part of the chamber, and air from a small compressor was blown into the water. This model was made of galvanised iron in the laboratory with the help of the boiler-house men. It worked.

The whole thing was then built again in its final size, in copper.

Some time later, I thought 'Why heat the water electrically while a steam line is handy?'

Steam from the 15 psi supply to the autoclave was blown into the water at the bottom of the plenum chamber, and air also blown in as before.

I was lucky, as the steam-cock had an aperture of 1/6". Steam at 15 psi issuing from a 1/6" hole is equivalent to 5 kw, so that very little manual adjustment of the steam-cock enabled us to hold the temperature in the plenum chamber at 78 °C without a thermostat.

Eventually, we obtained a small centrifugal blower and came to the final configuration: Blow steam from a 15 psi supply through a 1/8" aperture into the plenum chamber, and co-axially with this blow in air from the centrifugal blower. Blank off the blower by any suitable means to control the temperature. Again fortune was with us.

To restrict the flow from the blower and at the same time ensure high exit velocity to promote good mixing with the steam, I decided to blank off the blower output with a plate and, for a start, drill three 3/8" holes in the plate.

This *happened* to give us 78 °C in the load, and the equipment has been running for the past 10 years without attention.

So, having paid our respects to Medawar's concept, we may state:

1. Any temperature below 100 °C may be obtained accurately and consistently by mixing

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Figure 1.—Graph of temperature obtained by mixing steam and air in fixed proportion.



Figure 2.—Suva Steamer Mark IV.

steam and air in fixed proportions (Figure 1).

2. At the temperature we are most interested in  $(78 \,^{\circ}\text{C})$  the volumes of steam at atmospheric pressure, and air, are roughly equal and the requisite balance is easily obtained, firstly by holes in the blanking plate on the output of the blower and, if necessary 'fine-tune' with a pivoted plate over the intake side of the blower.

3. The high latent heat of steam means that the steam-air mixture has a high capacity to heat the load quickly.

4. Adjustment of the steam/air ratio leads to virtually instantaneous change on a thermometer in the stream and this adjustment is actually easier than setting a thermostat with its inherent lag.

5. Having got this high velocity mixture it can be applied to a chamber of any desired size or proportions.

6. The stream *heats the load first*. Losses through the walls are of no consequence. This is in contrast to the usual inspissator where the heat must flow by means of indefinite conductivity from the walls to the load.

7. The steam may be obtained from a steam-line

through a pressure-reducing valve, or may be obtained by simply boiling water in the bottom of the plenum chamber. If steam from a line is used, 'meter' the steam flow by a 1/8'' hole at the 15 psi valve, and make the steam inlet to the plenum chamber considerably larger—say four 1/8'' holes. this enables, say  $\frac{1}{2}$ '' nylon hose to be used between the source and the steamer, as the pressure in the hose is then low.

8. If heating water electrically, there is no need for constant-level water feed, as a suitable level of water needs seven litres or more and a half-hour run evaporates only 3-4 litres.

9. If steam is the source of heat, there is little condensation to accumulate—at least that was our experience in the tropics.

#### REFERENCES

- Medawar, P. B. 'The Art of the Soluble.' Methuen and Co., London (1967).
- 2. Samuel, J. A. (1953), J.N.Z. Assoc. Bact. 8, 17.

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#### Correspondence

## How Much Information can you Extract from a Gram Stain?

Sir,—Some years ago I received a swab from a post mortem with the message, 'see what you think of this; it might be a Proteus or Pyocyaneus; it stinks.'

After examining the gram stain, I thought to myself, I've never seen this organism before nor expected to, but I believe it is *Bacillus necrophorus*.

This is a gram negative, non-sporing strict anaerobe which grows only on enriched media. It was cultured on blood agar, cooked meat and deep serum stabs made. The latter culture produces a fir-tree appearance.

If you refer to Bergey, you will find an array of gram negative anaerobes, including one with such morphology that it has been named *Bacillus ridiculous!* This is not what I saw. The slide I examined showed a bacillus with a highly uniform morphology and this was the clue. We all know that since micro-organisms multiply by binary fission, that at any stage of their life cycle there must be some slight variation in size and shape. This is a constant finding. However this particular organism appeared to be identical in morphology, each organism stamped from the same die. One other aspect of note was that they were not fusiform but had parallel sides with unusual ends. The best description is perhaps that the ends appeared to be 'chamfered.' Later editions of Bergey now refer to this organism as *Bacillus biacutus*.

So never despair, amid the tedious collection of organisms you see every day, you too may have the pleasure (if only once in 40 years), of recognising something completely new, and from a gram stain at that! January 1980.

> ALF SAMUELS, (Formerly) Laboratory Superintendent, Public Hospital Laboratory, Suva, Fiji.

#### Abstracts

#### Shirley Gainsford, Maree Johnstone, N. J. Langford and L. M. Milligan

#### **CLINICAL BIOCHEMISTRY**

A Simplified, Colormetric Micromethod for Xylose in Serum or Urine with Phloroglucinol. Eberts, T. J., Sample, R. H. B., Glick, M. R., and Ellis, G. H., (1979), *Clin. Chem.* 25, 1440.

Using Phloroglucinol instead of p-bromoaniline in the colour reaction, this method for Xylose required a four-minute heating period and a total assay time of 10 minutes. Glucose interference is less than from that of Roe and Rice and its reproducibility, linearity and accuracy is comparable to, or exceeds that of the p-bromoaniline procedure. N.J.L.

**Evaluation and Application of Magnetizable Charcoal for Separation in Radioimmunoassay.** Al-Dujaili, E. A. S., Forrest, G. C., and Landon, J., (1979), *Clin. Chem.* **25**, 1402.

Charcoal and magnetizable ferric oxide can be trapped in a polyacrylamide gel to form magnetizable charcoal that enables separation with the use of a magnet in place of centrifugation. Conventional charcoal separation of antibody—bound that free antigen is compared with the magnetizable particles. Use in the R.I.A. method for Aldosterone is covered in detail.

N.J.L.

Screening for Cushing's Syndrome using Early Morning Urine Samples. Walker, M. S., (1979), Ann. Clin. Biochem. 16, 86.

A study reporting on the effectiveness of urinary free 11-hydroxycorticosteroid/creatinine ratios (UFC/Cr) as a screening test for differential diagnosis of Cushing's syndrome and simple obesity. The UFC/Cr ratio is said to give a clear distinction between the pathological and normal group, but does not exclude the possibility of intermittent adrenalcortical over-activity in apparently normal obese patients on its own.

N.J.L.

Solid—Phase Enzyme Immunoassay for Serum Ferritin. Anaokar, S., Garry, P. J., and Standefer, J. C., (1979), *Clin. Chem.* 25, 1426.

Described is an E.I.A. for Serum Ferritin. This

The Progressive Appearances of Multiple Urinary Bence Jones Proteins and Serum Paraproteins in a Child with Immune Deficiency. Bushell, A.C., Whicher, J. T., and Yuille, T., (1979), *Clin. exp. Immunol.* 38, 64.

Described is a child in whom multiple progressively appearing Bence Jones proteins in urine and serum paraproteins were found with no evidence of a malignant disease. N.J.L.

Hemoglobinuria and Hematuria: Accuracy and Precision of Laboratory Diagnosis. Bee, D. E., James, G. P., and Paul, K. L., (1979), *Clin. Chem.* 25, 1696.

Assessment of blood in urine by five routine methods showed poor association between dipstick and cell counting procedures.

N.J.L.

Rate Nephelometric Measurement of Rheumatoid Factor in Serum. Finlay, P. R., Hicks, Jane M., Williams, Jean and Lichti, D. A., (1979), *Clin. Chem.* 25, 1909.

Measurement of Rheumatoid Factor with a Nephelometer is described. This rate reaction method measures the complexing of antigen—antibody in a reaction taking 17-20 seconds. Precision, linearity and accuracy are stated to be excellent with results being compared with the (Hyland) R.A. test latex slide method.

N.J.L.

Serum  $\gamma$ -glutamyltransferase Isoenzymes in Extrahepatic Biliary Obstruction. Wenham. P. R., Price, G. P., and Sammons, H. G., (1979), *J. Clin. Path.* 32, 902.

Using sera from patients with Extrahepatic Biliary Obstruction five isoenzymes of  $\gamma$ -glutamyltransferase are shown to exist, by the use of electrophoresis, gel filtration, and ultracentrifugation. Three of the isoenzymes found were not able to be demonstrated in normal sera. The results are discussed and compared with studies of Alkaline Phosphatase. N.L.

High Molecular Weight Alkaline Phosphatase: A Clinical Study. Crofton, Patricia M., Elton, R. A., and Smith, A. F., (1979), Clin. Chem. Acta. 98, 263.

High molecular weight ALP is low or undetectable in the serum of healthy individuals or patients with bone disease, but appears in the serum of some patients with liver disease. Using sera from 72 patients with a variety of forms of liver disease, 14 patients with bone disease, and eight healthy individuals, high molecular weight ALP activity was measured and results compared with measurement of other indices of liver function in order to evaluate the enzymes usefulness in diagnosis of liver dysfunction. It was proved to be both sensitive and specific for detecting liver disease and may be useful in early detection of liver metastases. N.J.L.

Evaluation of Four "Kit" Immunoassay Methods for Determination of Alpha-Fetoprotein in Serum during Pregnancy. Wong, P. Y., Doran, T. A., Ho, F. F. K., and Mee, A. V., (1979), Chin. Chem. 25, 1905.

Four commercially available Immunoassay Kits; Damabott from Abbott Lab. Ltd., Amersham from Amersham from Amersham Corp., Pharmacia from Pharmacia Diagnostics, and Behring RIA., from Behring Ltd., are evaluated for measurement of Alpha-fetoprotein in serum from pregnant women. All were found acceptable with respect to sensitivity, stability, precision, linearity, and analytical recovery. N.J.L.

Abnormal Lipoprotein Patterns in Human Serum as Determined by Agarose Gel Electrophoresis. Papadopoulos, N., (1979), Clin. Chem. 25, 1885.

Described as a reference for the detection of lipoprotein abnormalities are electrophoretic lipoprotein patterns that include those of Tangiers, Abeta-lipoproteinaemia, and the patterns that show an extra band as found in Cholestasis and Multiple Myeloma. M.J.L.

#### IMMUNOHEMATOLOGY

Low Ionic Strength Salt Solution (LISS): Its Effective use in Routine Compatibility Testing. McPherson, A. J., (1979), *Path*, 11, 615.

Over the last three and a-half years, 30,000 units were compatibility tested for routine and emergency blood transfusions using LISS suspended cells. No sero-positive red cell-induced transfusion reactions have been encountered during this time. L.M.M.

#### Improved Yields Factor VIII From Heparinised

Plasma. Rock, G. A., Cruickshank, W. H., Tackaberry, E. S., and Palmer, D. S., (1979), *Vox Sang*, **36**, 294.

The Factor VIII activity of Cryoprecipitate processed from Heparinised plasma was significantly higher than that prepared from CPD plasma.

#### MICROBIOLOGY

Rapid Diagnosis of Anaerobic Infections by Gas-Liquid Chromatography, Ladas, S., Arapakis, G., Malamou-Ladas, H., Palikaris, G., and Arseni, A., (1979), J. Clin. Path. 32, 1163.

Eighteen pus specimens from various sources were analysed by Gas-Liquid Chromatography (GLC) and the short chain fatty acids detected were compared with the micro-organisms isolated by conventional methods. It was found that the detection of propionic, isobutyric, butyric or isovaleric acids by direct GLC of pus specimens is strong evidence for anaerobic infection. It can be concluded that direct GLC provides a rapid and reliable presumptive method for the differentiation between anaerobic and aerobic infections.

M.J.

### Group G Streptococcal Meningitis, Gopaul D. D., (1979), Canad. J. med. Tech. 41, 148.

A beta haemolytic streptococcus was isolated from the cerebrospinal fluid of an elderly patient. The organism was identified as belonging to Lancefield Group G. M.J.

Rapid Identification of Medically Important Yeasts, Land, G. A., Fleming, W. H., Beadles, T. A., and Foxworth J. H., (1979), *Laboratory Medicine*, **10**, 533.

An identification scheme for yeasts is presented which will allow the clinical laboratory to speciate most medically important yeasts within 24 hours of pure culture. A mini-scheme is also described which allows small laboratories to screen adequately for *Candida albicans, Candida stellatoidea* and *Cryptococcus neoformans* within 24 hours from a mixed culture. M.J.

#### Campylobacter Enteritis-An In-Patient Study, Pentland, B., (1979), Scot. med. J., 24, 299.

The variety of clinical features encountered in campylobacter enteritis in 26 patients is described. The article emphasise the need to consider campylobacter infection in patients presenting with bloody diarrhoea, acute abdominal pain or pyrexias of unknown origin. M.J.

**Detection of Bacterial Phosphatase Activity by Means of An Original and Simple Test.** Satta, G., Grazi, G., Varaldo, P. G., and Fontana, R., (1979), J. clin. Path. 32, 391.

Details of a phosphotase test that only requires the inoculation of an agar plate containing phenolphthalein diphosphate and methyl green are given. The test was tried with straphylococci, micrococci, streptococci, coynebacteria and enterobacteria and found reliable. S.G.

An Evaluation of 12 Methods for The Demonstration of Penicillinase. Lucas, T. J., (1979), J. clin. Path. 32, 1061.

Twelve methods that demonstrate penicillinase production were compared using strains of Haemophilus influenzae and Staphylococcus aureus. The acidometric agar plate method appeared the most suitable for routine use.

S.G.

**Evaluation of Bacteriological Swabs and Transport Media in The Recovery of Group B Streptococci on Laboratory Media.** Cumming, C. G., and Ross, P. W., (1979), *J. clin. Path.* 32, 1066.

Various swabs and transport media were assessed on their ability to maintain the viability of Group B Streptococci. The best results were obtained with swabs not held in transport media and kept at 4°C rather than room temperature.

S.G.

**Biochemical Identification of Clinically Important Yeasts.** De Louvois, J., Mulhall, Anne and Hurley, Rosalinde, (1979), *J. clin. Path.* 32, 715. The AP1 20C, API 20C (auxanogramme), Mycotube and Auxodisk kits for the identification of yeasts were tested against 50 clinical isolates. The advantages and disadvantages of each system are given. S.G.

Medium for Isolation of Yersinia Enterocolitica. Dudley, M. V., and Shotts, E. B., Jnr, (1979), J. clin. Mic. 10, 180.

The authors describe a new medium (cellobiose argenine lysine) for the isolation of Yersinia enterocolitica. It was found to be as sensitive as MacConkey agar in supporting growth of Y. enterocolitica and superior to MacConkey agar in differentiating it from other enteric organisms. The CAL media is incubated at 25 C for up to 40 hours and colonies of Y. enterocolitica appear bright burgundy. S.G.

#### Application of The Fortner Principle to Isolation of Campylobacter from Stools. Karmali, M. A., and Fleming, P. C., (1979), J. clin. Mic. 10, 245.

A culture of a rapidly growing facultative anaerobe is included in the same bag as cultures for Campylobacter species to reduce the oxygen tension. This method does not need anaerobic equipment and can be used for the isolation of C. jejuni from stools. S.G.

Reliable Urease Test for Identification of Mycobacteria. Steadham, J. E., (1979), J. clin. Mic. 10, 134.

A urease test is described in which the urea broth is modified by lowering the pH, concentrating the buffer and then increasing the length of incubation. 1346 isolates representing 17 differnt species were tested and gave reliable results. S.G.

Modern Trends in The Laboratory Diagnosis of Gonorrhoea. Low, A. C., and Young, H., (1979), *Med. lab. Sci.* **36**, 275.

This is a review of the laboratory diagnosis of gonorrhoea including transport of specimens, media, identification antibiotic sensitivity testing and serological methods. S.G.

#### HAEMATOLOGY

The Osmotic Fragility of Red Blood Cells: A Re-Evaluation of Technical Conditions. Godal, H. C., Nyvold, N., and Rustad, A., (1979). Scand. J. Haematol, 23, 55.

The osmotic fragility of red blood cells is influenced by even modest environmental changes. Consequently, the technical procedure must be strictly standardised. The authors have found that even a shift of 0.2 of a pH unit markedly influences the result. -E.R.C.

**Evaluation of the Activated Whole Blood Clotting Time (ACT) in Vitro.** Stenbjerg, S., Berg, E., and Albrechtsen, O. K., (1979), *Scand. J. Haematol.* 23, 239.

The activated whole blood clotting time (ACT) was analysed in vitro using blood samples from

normal individuals and from patients with heart disease, scheduled for open heart surgery. The authors established a linear relationship between ACT and heparin concentrations and conclude that the ACT is a reliable uncomplicated technique to monitor heparin, including the monitoring of heparin during open heart surgery. —E.R.C.

A Simple Enzyme—Immunoassay (EIA) Test for Factor VII—Related Antigen. Ness, P. M., and Perkins, H. A., (1979), *Thrombos. Haemostas.* (Stuttg.) 42, 848.

An enzyme immunoassay (EIA) has been developed to measure factor VIII-related antigen. The authors state that this assay gives similar results but greater precision than the Laurell electro immunodiffusion technique. -E.R.C.

Evaluation of the In Vitro Detection of the Hypercoaguable State Using the Thrombin Generation Test and Plasma Clot Impedance Test. Peck, S. D., (1979), *Thrombos. Haemostas.* (Stuttg). 42, 764.

This study reports the correlation of the thrombin generation test and the plasma clot impedance test with the clinical evidence of hypercoagulability, and shows that these two tests are practical, rapid and useful tests for detection and monitoring of the hypercoagulable state.

-E.R.C.

Hereditary Acanthocytosis Associated with the McLeod Phenotype of the Kell Blood Group System. Symmans, W. A., Shepherd, C. S., Marsh, W. L., Oyen, R., Shohet, S. B., and Linehan, B. J., (1979), *Br. J. Haemat.* 42, 575.

This paper presents the findings from a family that have the McLeod phenotype in the Kell blood group system, yet do not have Chronic granulomatous disease. It represents the first occasion that a person of a rare blood group has been recognised because of an associated anomaly in red cell morphology. —E.R.C.

Haemophilia "A' in a 46, X, i (Xq) Female. Mori, P. G., Pasino, M., Rosanda Vadala, G., Bisogni, M. C., Tonini, G. P., and Scarabicchi, S., (1979), Br. J. Haemat. 43, 143.

A phenotypically normal female, with negative family history for bleeding disorders, was found to be affected by severe haemophilia A. Chromosome analysis showed the presence of an X isochromosome of the long arm in every cell. -E.R.C. International Committee for Standardisation in Haematology: Recommended Screening Test for Glucose-6-Phosphate Dehydrogenase (G-6-PD) Deficiency. Beutler, E., Blume, K. G., Kaplau, J. C., Lohr, G. W., Ramot, B., and Valentine, W. N., (1979), Br. J. Haemat. 43, 469.

This paper describes the ICSH recommended screening technique for detecting G-6-PD deficiency, including preparation and storage of the reagent, performance of the test, interpretation etc.

-E.R.C.

A Precision Study of a Photo-optical Factor VIII Assay Technique. Brown, G. D., and Palkuti, H. S., (1979), Am. J. Clin. Pathol. 72, 204.

This brief scientific report discusses a photooptical factor VIII assay technique and the authors provide data to show the reliability of the method. This technique may be of interest because of the number of photo-optical clot timers on the market in New Zealand at present. -E.R.C.

Platelet Counts in Capillary Blood. Feusner, J. H., Behrens, J. A., Detter, J. C., and Cullen, T. C., (1979), Am. J. Clin. Pathol. 72, 410.

The authors collected venous and capillary bloods simultaneously from healthy adults to assess the accuracy of capillary platelet counts by electronic particle counter (Coulter Model ZB). For one quarter of the subjects the capillary blood count underestimated the venous blood count by 25 percent. The authors conclude that caution must be exercised in the interpretation of platelet counts performed on capillary blood by such methodology. -E.R.C.

Quality Control of the Latex-fixation Test. Singer, J. M., Edberg, S. C., Selinger, M., and Amram, Manuela, (1979), Am. J. Clin. Pathol. 72, 591.

The authors mention the preparation of a standard reference serum, and suggest a number of guidelines for the quality control of precision and sensitivity of the test. -E.R.C.

Bromochlorophenol Blue. A New Stain for Erythroblasts. Kass, L., and Gardner, F. H., (1979), Arch. Pathol. Lab. Med. 103, 565.

In erythroblasts of varying maturational stages, cytoplasm is stained bright yellow with bromochlorophenol blue. Since this staining reaction appeared to be distinctive among various cytological types of marrow cells, it may constitute a rapid method for visualisation of cells of erythroid origin. —E.R.C.

Discovery of an Inherited Bisatellited Metacentric Microchromsome in Amniotic Cell Culture. Romain, O. R., Columbano-Green, L., Smythe, R. H., and Dukes, P. C., (1979), *Clinical Genetics*, 16, 183.

The identification is reported from Wellington Hospital of an extra bisatellited metacentric microchromosome in amniotic cell culture from the third pregnancy of an identical twin, and its subsequent finding in the maternal parent as an inherited familial marker. The authors believe theirs to be the first bisatellited microchromsome to be clearly identified from an amniotic cell culture using silver staining. -E.R.C.

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已 で	Ortho Diagnostics Travelling Fellowship 1980	D D
민	"An Announcement"	ם ס
	"Notices were sent to Biochemistry Departments of all Laboratories regarding the Ortho Diagnostics Travelling Fellowship. Applications closed on 29th	ם ס
	February 1980, and were to be sent to the Secretary of the A.A.C.B. Judging will take place in Australia. Bev Gain thanks all those who applied and hopes that a New Zealander wips the award	٥ ٥
민	All the best to those who put in an application. The next opportunity to win the award is in 1982."	

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