Vol. 15, No. 4 MAY, 1961

JOURNAL

OF THE NEW ZEALAND ASSOCIATION OF BACTERIOLOGISTS

CONTENTS

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JOURNAL OF THE NEW ZEALAND ASSOCIATION OF BACTERIOLOGISTS

Vol. 15, No. 4 MAY, 1961

Communications regarding this JOURNAL should be sent to the Editor, Department of Pathology, Christchurch Public Hospital, Christchurch.

Communications primarily affecting the Association should be addressed to the Secretary, Mr H. E. Hutchings, Pathology Department, Palmerston North Hospital.

All moneys should be paid to the Treasurer of the New Zealand Association of Bacteriologists (Inc.), Mr D. J. Philip, Pathology Department, Middlemore Hospital, Auckland.

Subscription to this JOURNAL is five shillings per year or two shillings per copy, post free.

ADDRESSES

If the address as printed on this envelope is incorrect, **please** notify the Editor as soon as possible of your **correct** address.

A METHOD OF ASSAYING HEPARIN DURING EXTRA-CORPOREAL SURGERY

R. KENNEDY

(*Haematology Department, Central Laboratory, Auckland Hospital*)

In 1958 the cardio-surgical unit at Green Lane Hospital performed the first open-heart operation in New Zealand using the Melrose heart-lung machine.

Prior to surgery the patient is dosed with heparin which is also the anti-coagulant used for collecting the donor blood used initially in the machine.

Some months prior to the first attempt on a human patient, this department was asked to provide a method of assaying heparin that could be used during extracorporeal surgery as it is necessary for the surgeon to have regular checks on the heparin levels, both in the machine, in the patient during surgery and in the patient following neutralisation of the heparin with protamine sulphate.

It was decided that the test should fulfil the following requirements:—

(1) Simplicity of performance and interpretation.
(2) Reproducability.
(3) Speed in obtaining results.

HEPARIN AND ITS ACTION

Heparin is a sulphur-containing muco-polysaccharide naturally present in a number of animal tissues and found in high concentration in liver and lung tissues. Traces of heparin are found in blood and this is thought to arise from the mast cells. Heparin is relatively rapidly eliminated from blood (Eibier, et al 1958).

For its anticoagulant action heparin requires a co-factor which is found in the albumin fraction of plasma (Biggs, 1953). The presence of this co-factor allows heparin to prolong the thrombin-fibrinogen reaction and prevents formation of thrombin. It also interferes with the formation of thromboplastin probably through its action on Christmas factor (O'Brien, 1960).

The combined affect of these actions is to prolong the whole blood clotting time.

A BRIEF REVIEW OF TECHNIQUES AVAILABLE

(1) Chemical extraction methods.
(2) Physico-chemical methods based on titration with protamine sulphate.

(3) Colorimetric reactions with some basic dyes (e.g., toluidine blue).

(4) Anticoagulant action as measured by the whole blood clotting time.

The chemical extraction methods of Bassiouri (1953, 1954) were too time-consuming and involved for our use. In addition they departed somewhat from the testing of blood under physiological conditions.

The protamine sulphate titration method is based on the principle that the free basic group of the protamine sulphate reacts with the acidic groups of the heparin to neutralise the anticoagulant action of the heparin.

Allen, et al (1949) showed that blood heparin at levels which markedly affected the whole clotting time was not always capable of being measured by the protamine sulphate titration. In addition, protamine itself may have an anticoagulant action if present in excess amounts.

The estimation of heparin with some basic dyes, e.g., toluidine blue is based on the principle that if an increasing concentration of heparin is added to a solution of the basic dye, it produces a colour change from blue to purple to reddish-violet shades. However, the reaction is not specific for heparin and indeed may be affected by protein and other substances capable of binding the dye. Walton (1954) showed some of the failures of the reaction.

Warren and Wysocki (1958) compared the whole blood clotting time, protamine sulphate titration and Bassiouri's method for heparin estimations and concluded that a carefully done clotting time was more reliable than the other two methods. The clotting time of whole blood, however, has some limitations.

The main disadvantages are:—

(a) The lack of sensitivity where low levels of heparin are still present after neutralisation.

(b) Difficulty in reading the end point where heparin levels are high.

(c) Possible effect of a lengthened clotting time due to over neutralisation with protamine sulphate.

It was finally decided that to overcome the limitations of the techniques mentioned and to fulfil the needs of speed, simplicity and reproducability of measurements of heparin that a thrombin clotting time method would be most satisfactory.

Several standard thrombin clotting time techniques were tried but all seemed to lack reproducability. Eventually a modification was evolved which after thorough trials proved entirely satisfactory.

METHOD

The principle of the method is that a sample of the patient's whole blood, serially diluted with donor whole blood, ox plasma (source of co-factor) is added to each dilution and the dilutions are then clotted with a standard thrombin solution which has been checked against a series of control bloods containing known heparin levels.

MATERIALS

Pularin heparin, Parke Davis thrombin, ox plasma (ox blood collected in 3.8% sodium citrate, plasma removed and freeze dried in 0.5 ml. lots).

CONTROL

Normal citrated whole blood containing a known heparin level is diluted in normal citrated whole blood to contain final heparin concentrations as below:—

Heparin Concentrations	*Dilutions*
0.5 to 1 units	1 in 2
1.0 to 1.5 units	1 in 3
1.5 to 2.0 units	1 in 4
2.0 to 3.0 units	1 in 6
3.0 to 4.0 units	1 in 8

0.5 ml. of each concentration is placed in a series of Kahn tubes at 37°C. 0.1 ml. of ox plasma is added and 0.5 ml. of thrombin. The clotting times are noted.

To obtain maximum sensitivity it was found necessary to adjust the thrombin concentration to clot blood containing 2 units of herapin. To achieve this it is usually necessary to dilute the thrombin to a concentration of between 5 and 10 units. Once the thrombin has been standardised it is stable for one day only and should be refrigerated between tests.

TEST

The test is carried out as above except the patient's citrated whole blood is diluted in control blood, ox plasma and thrombin added and the clotting times noted.

Expressing Results	
1 in 8 does not clot	result over 4 units
1 in 8 clots	result 3 - 4 units
1 in 6 clots	result 2 - 3 units
1 in 4 clots	result 1.5 - 2 units
1 in 3 clots	result 1 - 1.5 units
1 in 2 clots	result 0.5 - 1.0 units

SUMMARY

A rapid, simple and reproduceable method of assaying heparin during extra-corporeal circulation is described. Some of the disadvantages of other techniques are described.

This method has been in use for two years and has proved entirely satisfactory.

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Walton, 1954. B.J. Pharmacal. 9, 1.

Warren and Wysocki, 1958. Surgery. 44, 435.

THE EFFECT OF GLUCOSE ON THE ESTIMATION OF TOTAL 17 OH. CORTICOSTEROIDS IN URINE

T. E. MILLER

Chemical Pathology Department, Central Laboratory, Auckland Hospital.

The estimation of corticosteroids and 17 ketosteroids in urine is now frequently requested by physicians, and a number of laboratories in this country are capable of estimating these compounds. In this laboratory the total 17-hydroxy-corticosteroids (total 17 OH. C.S.) are estimated by the method of Norymberski (Appleby et al., 1955) and the 17 ketosteroids according to Drekter et al. (1952). Norymberski et al. (1953) drew attention to the fact that glucose present in the urine may hinder the estimation of total 17 OH. C.S. From discussion with other laboratory personnel it appeared that this hazard is not generally appreciated. This investigation was designed to confirm the findings of Norymberski et al., and to provide a satisfactory technique for the elimination of glucose from the urine prior to the estimation of total 17 OH. C.S.

Experimental.

MATERIALS AND METHODS:

A 24 hour urine with a high total 17 OH. C.S. value and free from glucose according to Benedicts qualitative test was selected for this study. Aliquots were prepared containing 0.0, 0.5, 0.1, 2.0, 3.0, 4.0 and 5.0 per cent glucose. The following technique for eliminating glucose from an aliquot of the test urines was adopted.

(1) The pH. of the urine was adjusted to approximately 7.0 with normal NaOH and a glass electrode.

(2) A suspension of yeast was prepared (1 vol. and 2 vols. of water) after washing the yeast three times with water.

(3) 2 mls. of this suspension was taken, centrifuged, drained thoroughly, and 20 mls. of urine added to the packed yeast. The yeast was resuspended in the urine.

(4) The urine containing yeast was incubated in a water bath at 37°C. and tested every thirty minutes for glucose using 1 ml. of Benedicts qualitative solution and 0.1 mls. of the urine specimen. When the Benedicts test was negative the preparation was centrifuged and the determinations carried out on the supernatant fluid.

All specimens were treated in this manner. Estimations of the 17 ketosteroids and total 17 OH. C.S. content were performed on the specimens containing glucose and the same specimens after the removal of glucose by the technique described.

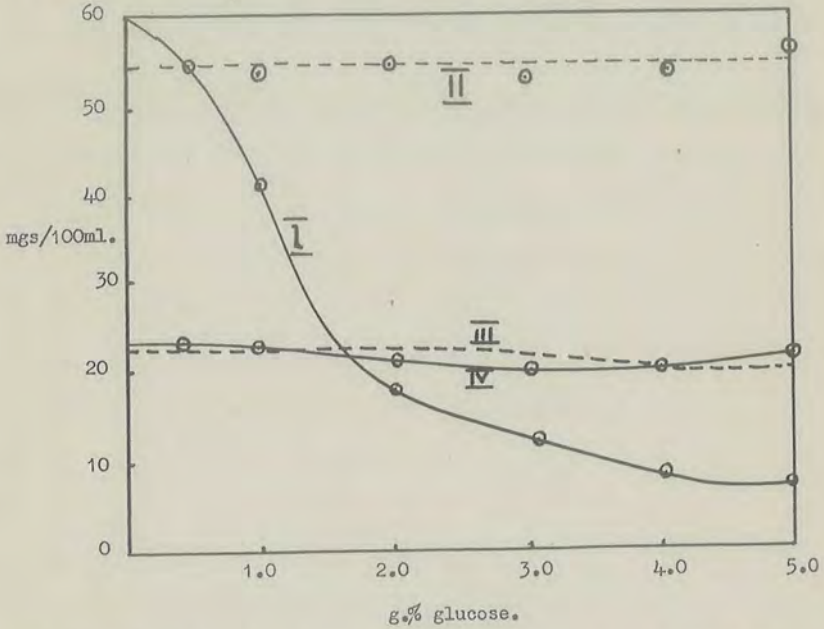

Fig. 1. The effect of glucose on the estimations of total 17 Hydroxy-corticosteroids in urine.

I. Measured concentration of 17 OH. C.S. before yeasting.
II. Measured concentration of 17 OH. C.S. after yeasting.
III. Measured concentration of 17 ketosteroids after yeasting.
IV. Measured concentration of 17 ketosteroids before yeasting.

DISCUSSION:

Norymberski (1952), showed that sodium bismuthate oxidizes several groups of 17 OH. C.S. to ketosteroids, hence any glucose present in the urine will be oxidized at the expense of the majority of the 17 OH. C.S. The estimation of 17 ketosteroids contains no comparable oxidation step and as was expected no significant error was introduced by the presence of glucose in the urine. Useful diagnostic information regarding the activity of the adrenal cortex may be obtained by studying the effect of administered A.C.T.H. on the urinary excretion of total 17 OH. C.S. It is known that the administration of A.C.T.H. can cause an elevation of the fasting

blood sugar, a diabetic glucose tolerance curve, and glycosuria. We have observed that a normal rise in total urinary 17 OH. C.S. following A.C.T.H. stimulations can be masked by the glucose present in the urine and an abnormal stimulation curve obtained instead.

Our observations are similar to those of Norymberski et al. The latter did not however recommend a technique for the elimination of glucose. Recovery results for total 17 OH. C.S. following yeasting indicate that this is a convenient and satisfactory technique for the removal of glucose from the urine.

SUMMARY:

The effect of glucose present in the urine on the estimation of total 17 OH. C.S. and 17 ketosteroids is recorded. A technique for freeing the urine from glucose by yeasting is described and the importance of testing the urine for glucose prior to the estimations of total urinary 170H.C.S. discussed.

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ACKNOWLEDGEMENTS:

I am indebted to Drs. F. H. Sims and R. O. Farrelly of this Department for help and advice in the preparation of this paper.

A METHOD OF PRESERVING AND MOUNTING PARASITIC OVA FROM THE FAECES

I. C. KING

Princess Mary Laboratory, Auckland Public Hospital.

During the past some diffficulty has been experienced in finding a suitable method for permanently preserving parasitic ova for future microscopic examination. Several methods are available—including one using a water-miscible mounting medium, but none of these have proven particularly successful nor do they seem to preserve the ova morphology. The following is a method evolved at this Laboratory which appears to be quite successful and which does not unduly distort the specimen. It was found that some means of eliminating the fat and moisture content of the faeces was a major necessity and it was with this idea in view that this method finally emerged. The concentration method used is not new but has been found to be particularly reliable and after 20 weeks the original mounted specimens are still in good condition and are optically as perfect as when originally examined.

Concentration Technique:

(a) To 10mls of formal-saline add a portion of faeces. (The volume of faeces to be determined by the number of ova found in the original examination.)

(b) Mix well and add 1ml of ether.

(c) Cork and shake for about 30 seconds.

(d) Centrifuge by slowly bringing up to 2,500 r.p.m. and then allow the centrifuge to come slowly to a stop.

(e) A layer of fats, etc., will be noticed between the ether and formal-saline. Free this by ringing with an applicator stick and decant all the supernatant including this layer.

(f) The remaining deposit contains the ova.

Preparing and Mounting Technique:

Prepare new, clean slides and cover-slips before commencing.

(a) To the deposit remaining in the tube in (f) above add 10mls of S.V.R. Mix well and centrifuge. Repeat this process three times.

(b) After decanting the last wash of S.V.R. add 10mls of Absolute Alcohol. Mix well and centrifuge. Repeat this process three times.

(c) After decanting the last wash of Absolute Alcohol add 10mls of Ether. Mix well and centrifuge. Repeat this process three times. Do not decant all the Ether from the final wash.

(d) Take up the Ether-deposit mixture in a Pasteur pipette and place one or two drops of this on to each of the previously prepared slides. Allow to evaporate. The Ether-ova mixture evaporates quickly and care must be taken not to finish up with the preparation having evaporated in the tube or pipette.

(e) Gently blot the preparation with Whatman's No. 1 filter paper to remove any remaining drops of moisture. I have found ordinary blotting paper tends to leave fibres enmeshed in the preparation and I therefore changed to filter paper.

(f) Mount using DePeX mounting medium.

LIBRARY NOTES

My Life with the Microbes. Selman A. Waksman. Autobiography of the discoverer of Streptomycin.

In 1910, Waksman, aged 22, arrived in America from his native Russia. Through the years, he studied, taught and travelled widely, becoming world famous for his discoveries of antibiotics, until in 1952 he was awarded the Nobel Prize for Physiology and Medicine.

This book is a well written account of his work as well as being an interesting personal story.

Famous Tropical Physicians. G. Rath.

Brief biographies of thirty men who have made contributions to the study of tropical medicine. Includes Hippocrates, Bilharz, Hansen, Manson, Laveran, Reed and Ricketts.

An excellent introduction to a subject with which we have little contact in New Zealand.

The World of Albert Schweitzer. Text by Eugene Exman. Photographs by Erica Anderson.

While new hospitals with every comfort and modern scientific aid spring up in our prosperous country, it is fitting that we should think of others who struggle to heal the sick in conditions of hardship and poverty.

This book of magnificent photographs gives us a glimpse of life in the jungle of French Equatorial Africa where Albert Schweitzer has established his remarkable hospital.

"My Life with the Microbes" and "The World of Albert Schweitzer" may be obtained through any public library.

We are indebted for our review copy of "Famous Tropical Physicians" to the New Zealand distributors of Merck reagents, Messrs Watson Victor Ltd., from whom a limited number of copies may be obtained.

REFRESHER COURSE FOR PRINCIPAL BACTERIOLOGISTS

This course, the first of its kind for our Association, was held at Greenlane Hospital, Auckland, from the 10th-13th October, 1960. Instigated by Dr. S. Williams, Pathologist, and Mr I. Cole, Principal Bacteriologist, both at Greenlane Hospital, the course was designed primarily for people from the South Auckland area.

In opening the course, Dr. Williams stressed the value not only in increased knowledge, but in better liaison between laboratories which could result from the free interchange of ideas. The truth of this became evident as the four-day course progressed and those present became more aware of the problems of their colleagues.

FORMAL LECTURES COVERED THE FOLLOWING:—

Liaison with Medical Staff and other Laboratories—Dr. Williams.
Recent Advances in Haematology—R. Kennedy.
Training of Laboratory Staff—D. Whillans.
Recent Advances in Serology—A. Fischmann.
Blood Bank Practice—R. Douglas.
Antenatal Lab. Problems—J. Sloan.
Advances in Biochemistry—Dr. Sims.
Cytology and Histology—J. Cole and Dr. Williams.
Equipment and Sterilization—D. Whillans.
Pediatric Lab. Problems—Dr. Beecroft.
Viruses—R. McKenzie.
Medico-Legal Problems—Dr. Cairns.
Recent Advances in Bacteriology—J. Holland.
Lab. Administration and Records—Dr. Williams.
T.B. Methods—G. Chambers.

Additional to the formal lectures was an interesting session on the "Heart-Lung" machine and a visit to Biological Laboratories and to Dr. Burton's Cancer Research Unit.

Those attending the course were:—

Dr. Simcock, Opotiki.
Mr G. George, Rotorua.
Mr R. Barrington, Hawera.
Mr D. Smith, Tauranga.
Sister Killian, Mater Hospital, Auckland.
Mr G. Kuru, Wairoa.
Miss Davies, Waikato.
Mr Clapson, Waikato.
Mr B. Robertson, Thames.

R.W.B.

FINAL EXAMINATION FOR THE CERTIFICATE OF PROFICIENCY IN HOSPITAL LABORATORY PRACTICE

Examiners: Dr. F. Cairns, Dr. W. Kenealey, Mr H. T. G. Olive.

Tuesday, 30 August, 1960.

9.30 a.m.-12.30 p.m.

WRITTEN EXAMINATION

1. Write notes on—
   (a) Bacteriophage
   (b) Anaphylaxis
   (c) Mantoux Test
   (d) Preparation of autogenous vaccines for patients with recurring boils.
2. Discuss a scheme for processing formalin-fixed tissue to a stage where it is suitable for cutting and staining. Give the reasons for each of the steps and approximate time intervals.
3. A patient has had a haemolytic transfusion reaction—
   (a) List the laboratory tests that may be done to confirm the diagnosis.
   (b) Describe the steps you would take to find the cause of the reaction.
4. Discuss methods for estimating serum proteins and the various fractions thereof, as carried out in a hospital biochemistry laboratory.
5. Write brief notes on the following—
   (a) Beer's Law.
   (b) Milli-Equivalents.
   (c) Diffraction grating.
   (d) Sulphaemoglobin.

Tuesday, 30 August, 1960

2.30-5.30 p.m.

PRACTICAL I

BACTERIOLOGY

CULTURE A

Organism from faeces of case of suspected enteric fever: Identify the organism.

(S. typhi).

CULTURE B

Organism from urine: identify the organism.

(Enterococci).

CULTURE C

Organism from swab of boil: identify organism and determine sensitivity to antibiotics supplied.

(Coagulase positive Staphylococcus aureus).

SERUM E

Do Widal and give interpretation of results.

BIOCHEMISTRY

1. Carry out an alkaline phosphatase determination on serum (labelled D) by the King Armstrong method. Explain the various steps and principles involved.
2. Write brief notes on the exhibits set out.
   1. Bottle Potassium Hydrogen Phthalate.
   2. Bottle Trichloracetic Acid.
   3. Stained Electrophoretic Strip.

4. Volumetric Pipette.
5. Thermostat.

Wednesday, 31 August, 1960
9.30 a.m.-12.30 p.m.

PRACTICAL II

BACTERIOLOGY

1. Continue work with cultures from yesterday.
2. Write notes on "spots."
   Enterobius vermicularis.
   P. humanis.
   Malarial parasites.
   Ringworm slide.
   Incubator capsule.
   Element assembly of McIntosh & Fildes Jar.

HAEMATOLOGY

3. Do an osmotic fragility test on the blood provided (incubation *not* required).
4. You are given A1 and A2 cells and anti-A serum. Determine the titre of Anti-A in saline.

Wednesday, 31 August, 1960
2.30-5.30 p.m.

PRACTICAL III

HAEMATOLOGY

1. You are provided with oxalated blood and films. Estimate the packed cell volume, haemoglobin and calculate the mean cell haemoglobin concentration. Report on the film.
2. Stain and report on the films P, Q, R. Report on the films S, T, U.
   P.—Normal infant's blood.
   Q.—Untreated P.A.
   R.—Hypochromic anaemia in an infant with pneumonia.
   S.—Film showing Pelger anomaly of the leucocytes and nucleated red cells.

BIOCHEMISTRY

3. Examine the urine specimen provided for reducing substances, acetone and urobilinogen.
4. Write brief notes on the exhibits set out.
   a. Bottle Acetic Anhydride.
   b. Bottle Sulphanilic Acid.
   c. Retort Splash Trap.
   d. Chromatography Strip.
5. Do van Gieson stain on paraffin section provided.

ORAL EXAMINATIONS

Dr. Cairns:

Life cycle E. granulosis, H. influenzae, Refrigerators, Complement fixation tests, Postal regulations, Casoni tests, Fixation of gross specimens, Filters, Determination of non-lactose fermenters motility.

Dr. Kenealy:

Paul Bunnell Tests, Abnormal mononuclear cells, Antibody titres, Enzymes used in Rh work, Cross-match technique, Platelet counts, Methods of haemoglobin estimation, Significance of abnormal R.B.C. osmotic ragility tests, Howell-Jolly bodies.

Mr Olive:

CSF Protein, CSF sugar in infection, CSF chlorides in T.B., Spider web clots, Effect of blood on CSF colloidal gold reactions, Electrolytes, Urea, TNPN, Blood sugar, Protein fractions, Amylase, L.D.H., Transaminase, pH, Bilirubin, Cholesterol, Urinary Diastase, Porphyrins, Normal solutions, Molar lactate, Oily lye, Topfer's and phenolpthalein indicators, Flame photometry, Nessler's reagent, Protein precipitants, Postal regulations.

The following candidates were successful:—

Miss M. D. LEADLEY, 5 O'Rorke Street, Auckland.
Mrs C. J. TAYLOR, Public Hospital, Auckland.
Miss S. COXHEAD, Public Hospital, Auckland.
Miss J. LEVIEN, Public Hospital, Auckland.
Miss S. FURKERT, Public Hospital, Auckland.
Mr K. WATTS, Public Hospital, Auckland.
Mr G. J. McMAHON, Public Hospital, Wellington.
Mr A. HOWELL, Waikato Hospital, Hamilton.
Mr K. R. JAMES, Waikato Hospital, Hamilton.

INTERMEDIATE EXAMINATION FOR HOSPITAL LABORATORY TRAINEES

Examiners: Dr. L. Sefton, Mr G. George.
Wednesday, 19th October, 1960.
Time allowed: 3 hours.
Answer all questions.

WRITTEN EXAMINATION

1. Describe, with the aid of a diagram, a normal standard labouratory autoclave. Detail the steps taken, and the reasons for them, to sterilize an average load. State the optimum operating conditions for efficient sterilization, and indicate any precaution for personal safety.
2. Excluding basic components, state the important and distinctive ingredients of the following culture media. Indicate how they function in each medium and the particular purpose and method of use for the complete medium.
   (a) MacConkey agar.
   (b) Tetrathionate or Selenite broth.
   (c) Urea agar or broth.
   (d) Loeffler.
   (e) Hoyles.
3. Write *brief* descriptive notes on the following. Use diagrams where necessary.
   (a) Numerical aperture.
   (b) Reduced oxygen tension.
   (c) Coagulase test.
   (d) Penicillinase.
   (e) Bimetallic thermostat.
   (f) Hydrogen Ion Concentration.
4. Describe your method for estimating haemoglobin in blood. What errors can occur in this estimation? Describe the advantages and disadvantages of your method compared with other methods which are available.

5. Give an account of your method of performing the following examinations:
   (a) A platelet count.
   (b) A routine examination of a pleural fluid.
   How should specimens for these examinations be sent to the laboratory?
6. How would you determine the T.N.P.N. content of a specimen of blood? Explain the chemical reactions which take place. From what substances in blood is the non-protein nitrogen derived?

PRACTICAL I

Wednesday, 19 October, 1960
2.30-5.30 p.m.

BACTERIOLOGY

1. Perform routine qualitative examination of urine provided, including deposit, culture and sensitivity, with the discs provided. Make a smear and gram stain, from the deposit. Report your findings.
   (Pus, RBC, Acetone +, Sugar +, Albumin +, enterococcus, Staph aureus, coliform bacilli.)
2. This is a pure culture of a non motile organism isolated from faeces. Complete its identification.
   (a) for an urgent report;
   (b) for reliable confirmation of identity.
   (S. sonnei.)
   List briefly the steps taken, and summarise your findings, supporting the identification.
3. Report on the four slides provided from:
   (1) Urethra (Gonococci).
   (2) Mouth (Vincent's organism).
   (3) Sputum (T.B.)
   (4) Thoat (C. diphtheriae).
   Very briefly detail the staining procedure for each (one method only for No. (4).
   Baceriology to be completed tomorrow.

BIOCHEMISTRY

1. Determine the glucose content of the specimen of blood (J).
2. Solution (K) contains 6.0 gms. of sodium carbonate per litre. Use it to determine the normality of the solution of hydrochloric acid (L).
3. Estimate the chloride content of the c.s.f. (M).

PRACTICAL II

Thursday, 20 October, 1960.
9.30 a.m.-12.30 p.m.

BACTERIOLOGY

1. Complete Bacteriology from yesterday.
2. Make brief notes on the four spots provided.
   1. Sintered glass filter.
   3. Two micro pipettes.
   3. Pneumococci on blood agar.
   4. Blood specimen bottle with label to indicate EDTA as the anti-coagulant.

HAEMATOLOGY

1. Are the cell suspensions (A) (B) and (C) suitable for transfusions to a patient, a specimen of whose serum (D) is supplied?
2. Carry out a red count of the specimen of blood (E). Calculate the standard deviation and coefficient of variation of your result. Using

the Hb value and P.C.V. supplied, calculate the mean cell haemoglobin, the mean cell volume, and the mean corpuscular haemoglobin concentration.

3. Make a blood film from the specimen (E) used in the previous question. Leave it for inspection by the examiners. (Staining not required.)
4. Write a report on blood films (F) (G) and (H). Credit will be given for an accurate description of the appearance rather than a diagnosis. A differential count is not required.
   F. P.A.
   G. Chronic lymphatic leukaemia.
   H. Nucleated RBC, Howell-Jolly Bodies Polychromasia.

ORAL EXAMINATIONS

Dr. Sefton:

Normality of H_2SO_4, Dilution of H_2SO_4, Normal solutions, Molar solutions, Indicators for H_2SO_4 titration, Red cell development, Cross-Matching, Occult blood, Absolute values.

Mr George:

Hoyle's medium, Growth factors of H. influenzae, Br. abortus, Bile pigments in urine, Volumetric pipettes, Disposal of T.B. sputum pots, Anaerobic jars, Albumin in urine, Staining of Vincents, Blood culture medium inactivators of Sulpha and Penicillin, Balances.

Successful candidates were:

Mr F. B. O'MEARA, Rotorua.
Miss H. M. RANFORD, Waipukurau.
Miss J. POOLE, Tauranga.
Mr T. E. BROWN, Dunedin.
Miss A. K. BARRACLOUGH, Wellington.
Miss P. M. LAWN, Wellington.
Miss M. H. BURNETT, Wellington.
Mr L. R. TAYLOR, Wellington.
Miss J. A. WOLLEY, Wellington.
Miss L. B. WILLS, Wellington.
Mr J. E. DAVIES, Invercargill.
Mr D. A. CATHCART, Invercargill.
Miss P. K. OAKLEY-BROWNE, Auckland.
Mr H. F. STUNZER, Auckland.
Miss M. B. HUDSON, Christchurch.
Miss E. A. SLEE, Christchurch.
Miss B. DRAYTON, Hamilton.

A well aimed blow by Pyrex

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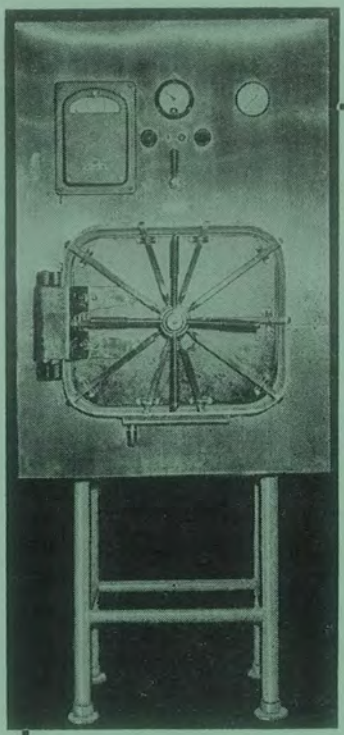

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