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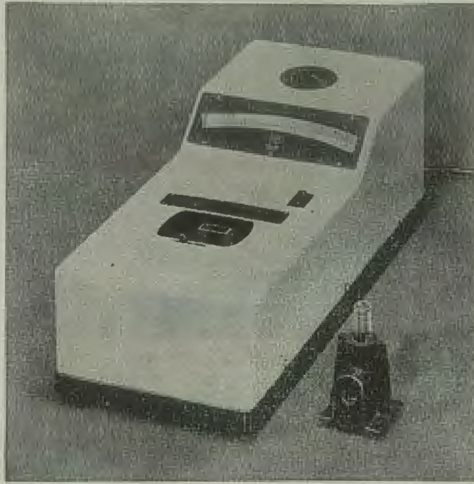

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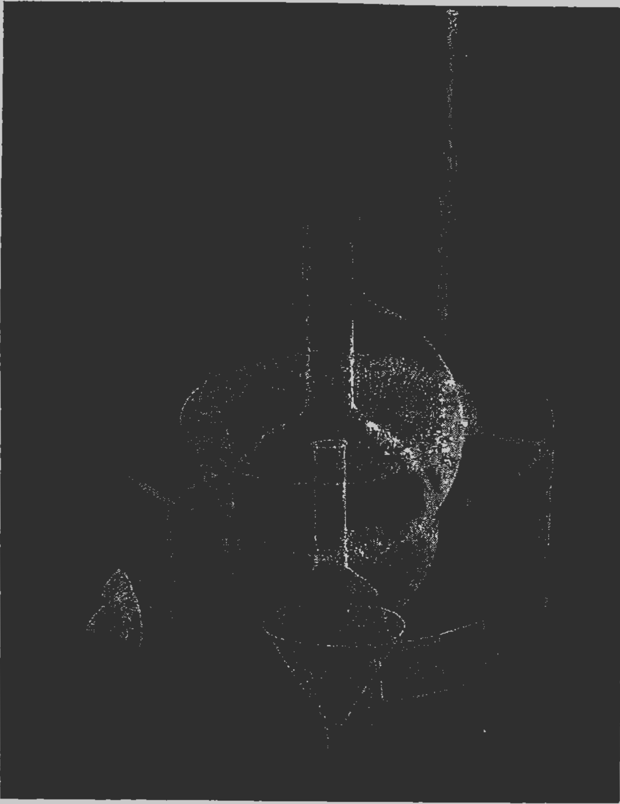

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Vol. 10, No. 2 **July, 1955**

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POLIOMYELITIS IMMUNISATION

Immunisation against poliomyelitis is not a new concept. Attempts were made twenty years ago but they met with little success largely because of the difficulties involved in obtaining virus in a sufficiently high concentration and free of antigenically potent nervous tissue components.

The recent dramatic change in the prospects for successful immunisation has been brought about foremostly by advances in laboratory techniques, that is, the discovery that the three poliomyelitis viruses would grow in tissue culture of human and monkey tissues. Coincident with this advance is the revival of immunological concepts, although established for other virus diseases were hitherto not accepted for poliomyelitis. Finally is the improved understanding of the disease process itself. Tissue culture allows the virus to be obtained in high concentration and in a medium suitable for inoculation.

In order to understand the immunological concepts involved it is necessary to have some understanding of the disease process itself. The measurement of the proliferation of the virus in man is a difficult and therefore somewhat neglected subject. Bodian, however has worked for some years on this problem, using monkeys and chimpanzees as experimental hosts and has pieced together the sequence of events.

It is clear that virus multiplies first in the tissues of the upper and lower parts of the alimentary tract and secondly in the nerve cells of the brain and spinal cord. In the majority of infections only the first multiplication takes place and invasion of the central nervous system must be regarded as unusual, occurring perhaps only once in five hundred infections.

When C N S infection does occur however it seems certain that it is initiated by virus which escapes from the site of primary multiplication and probably travels via the blood stream. Virus can be recovered from the blood only during the pre-paralytic period as neutralising antibodies are present at the onset of paralysis and are presumably formed as the result of the primary multiplication. Virus is excreted in the faeces for many weeks following infection and therefore does not appear to be influenced by antibody. Virus in throat secretions however is of short duration

and disappears with the appearance of antibody. High levels of antibody in the serum does not affect the progress of infection and nerve cell destruction in the central nervous system after the virus has been established there. In contrast however there is little doubt that relatively small amounts of circulating antibody can block the entry of virus into the C N S from the sites of primary multiplication.

At the present time it does not seem possible to eliminate the virus from the alimentary tract or to control in any effective way its spread from person to person. Therefore the only form of control lies in immunisation procedures which will produce serum antibodies at a level which will block the spread of virus from the alimentary tract to the C N S.

In populations where overcrowding and poor sanitation exist infections occurs in infancy and those who escape paralysis are permanently immune. In Cairo most children develop antibodies to all three virus types before they are four years old and the virtual absence of paralysis in those above the age of infancy is evidence that inapparent alimentary tract infection immunises against subsequent paralytic attacks. This equilibrium between virus and human beings breaks down where civilization leads to less overcrowding and improved sanitation. Initial infection is delayed until later in life when for some reason not understood paralytic disease is more likely to result. In this country the high standard of living has resulted in a low population immunity. Only approximately 50% of the children up to fifteen years of age have been found to have antibodies to type I and II viruses and only 11% have type three antibodies. This probably accounts for the high paralytic rate during epidemics.

Two types of immunising procedures are possible. One consists of a killed vaccine injected intramuscularly and the other of attenuated strains incorporated in a vaccine given either orally or by injection. To each there are serious objections and much work has yet to be carried out before the final answer is obtained. The cultivation of viruses in unnatural hosts for several generations may produce a variant which although not producing a severe disease will nevertheless produce an immunising infection. Vaccines used for smallpox, yellow fever, and rabies are examples of viral strains of reduced virulence. Already one strain of polio virus (Type II) has been adapted to the chick embryo and strains avirulent for monkeys have been produced by continuous passage in tissue cultures. Even if these latter strains should prove satisfactory for human vaccination several theoretical problems immediately arise. It is possible for avirulent strains given orally to become virulent again as the result of multiplication in the natural host and thus cause a paralytic infection and transmit virulent virus to other susceptible individuals. This is perhaps a remote

but theoretical possibility. Another danger which could occur with living virus vaccines in tissue culture is the presence of another viral contaminant. Where monkey tissue is used the highly pathogenic Herpes B could be present, or, in the case of human tissue, the virus of Infectious Hepatitis. It would seem therefore that before a truly satisfactory living poliomyelitis vaccine can be produced, the three viruses will have to be adapted to the developing chick embryo. This medium has been used in the past for the production of typhus, and yellow fever vaccines and no virus contaminants pathogenic for human beings have ever been traced to the embryo.

There is no doubt that virus inactivated by formaldehyde and other chemical and physical agents can provoke an antibody response. This has been shown conclusively by Salk and his colleagues. Although apparently non infectious, such virus retains some antigenic power. Likewise there is little doubt that killed virus vaccines are inferior to live vaccines as immunising agents. Howe, working with chimpanzees has shown that excellent antibody response is produced by living polio virus vaccines and that antibody remains at a high level for a considerable time. Rexposure results in a rapid "booster" response.

Antibody titres produced by killed vaccines are 10-20 times lower than those resulting from living vaccines and appear to fall below detectable levels in less than 12 months. If natural infection takes place while antibody is still detectable a "booster" effect will occur resulting in a "solid' immunity. The crucial question however is whether a rapid "booster" effect will result following infection when antibody has fallen below detectable levels. Salk claims this is so, and the antibody-forming mechanism has been sufficiently sensitised to respond with this "booster" effect. Whether the response under such conditions is rapid enough to prevent virus spreading to the central nervous system is still not determined. Should this not be so repeated inoculations at perhaps 12 monthly intervals will be necessary to maintain a safe level of immunity.

Inactivation of the virus with formalin slowly destroys the antigenicity and so its action must be stopped at a time considered to be sufficient for destruction of infectivity but not long enough to significantly lower potency. If this inactivation is not carried out under optimum conditions, living virus may remain. Unless some alternative method can be found, this will be a major criticism of the vaccine.

At the present time the killed vaccine is the only one available for immunisation but it is unlikely that this type will prove to be the most satisfactory when considering the many factors involved. If it is finally decided that a living vaccine is required, then it will

probably be necessary to wait until chick embryo adapted strains are available and it is not possible to predict when these will be produced.

THE CREATININE CLEARANCE

An easily performed renal function test.

G. F. DAVIS

(Department of Pathology, Auckland Hospital.)

Introduction

The purpose of this article is to bring to notice this test which though not a recently developed one, does not appear to be as widely used as it could be. It does not necessarily supplant other well known tests, but is at least a valuable additional determination giving an estimate of renal efficiency comparable with that obtained by the more common methods such as the urea clearance.

The test may be performed with a short term urine collection period, such as in the manner of the urea clearance test, or, due to the relative constancy of blood creatinine levels, urine collection may be carried out over a period of 24 hours. This eliminates the necessity for strictly accurate timing of collection periods, and complete emptying of the bladder at the end of these periods. Non-observance of the latter, especially at low rates of urine flow, can give rise to large errors in the values obtained when clearance tests are carried out over short periods.

Differential clearances can also be determined, obtaining urine specimens by means of ureteric catheters. A knowledge of the efficiency of each kidney can be extremely important especially where surgery is contemplated.

For the laboratory determinations, preparation and maintenance of accurately prepared standard solutions is not necessary, as absolute creatinine levels need not be determined. The same method of estimation is used for both blood and urine, and the ratio of creatinine concentration in these, together with a knowledge of the respective dilutions employed, is all that is required for calculation of the clearance.

Reagents used in the test are those commonly used in other tests also, and their solutions are simply prepared.

PHYSIOLOGICAL BASIS OF THE TEST

If the concentration of a substance in urine is denoted by U, and the rate of urine flow in mls./minute by V, then the quantity of the substance excreted by the kidneys in one minute is $U \times V$. Then if the blood concentration is B, the same units of concentration being used, this quantity, $U \times V$, is contained in $\frac{U \times V}{B}$ mls. of blood. That is, $\frac{U \times V}{B}$ mls. of blood are completely cleared

of the substance in one minute. This is, of course, a "virtual" volume cleared, as in most cases, including that of creatinine, only a certain proportion of the substance is removed on a single passage of blood through the kidneys. The clearance value represents the volume which would be cleared if removal of the substance was complete, on one passage through the kidneys.

Since filtration in the kidney involves removal of substances from the plasma, and not from the red cells, the clearance is more correctly calculated in terms of the plasma concentration P, and this is the mode of expression for the creatinine clearance. With regard to the urea clearance, the original range of values for normal subjects was established using whole blood urea concentrations. This method is still followed, the results also being expressed as a percentage of the average normal value.

In considering the clearance of creatinine by the kidneys, a number of difficulties are encountered, and the exact mode of excretion is by no means clearly understood.

Some of the difficulties encountered are firstly, that the excretion of endogenous and exogenous creatinine may involve different mechanisms in man, and secondly, the clearance as determined, is a total clearance of the chromogen estimated by the Jaffe reaction, and not a true creatinine clearance.

The exogenous creatinine clearance in which the plasma concentration is elevated to the order of 5mgms./100mls. by the intravenous administration of creatinine, is now used mainly as an investigational technique on dogs, where this clearance gives a true glomerular filtration rate. In man, this is not so, and the exogenous clearance offers few advantages, and numerous disadvantages over other available methods.

Examination of the endogenous clearance in man has produced many conflicting reports, and the consensus of opinion is that it is not a valid investigation method in its present form.

As a clinical method however, the endogenous clearance gives results which, in practice, give a good approximation to the glomerular filtration rate, with two main exceptions.

The non-creatinine chromogen estimated by the Jaffe reaction has a low clearance, and reduces the total chromogen clearance proportionally, so that an increase in this fraction as in certain cardiac diseases, will give falsely low clearances. On the other hand, in chronic renal disease, clearance values tend to be higher than the glomerular filtration rate as determined say, by the inulin clearance, due to excretion of creatinine by the tubules.

It has been estimated that a decrease of renal function to one third of its normal value, results in an overestimate of 10% in the glomerular filtration rate as determined by the endogenous creatinine clearance. Further renal impairment results in proportionately higher over-estimates. Such an error is of little signifi-

cance clinically. For example, if the glomerular filtration rate is determined as 10 mls. per minute, and a value of 13 mls. per minute is found for the creatinine clearance, an over-estimate of 30% has resulted, but clinically this has little importance.

The endogenous clearance gives substantially constant results over a large range of rates of urine flow. Extreme oliguria however may give rise to deviations.

THE DETERMINATION OF CREATININE

This is based on the Jaffe reaction, in which an orange-red colour is produced by creatinine in alkaline picrate solution. The colour is due to a red tautomer of creatinine picrate, the structure of which has not been determined.

This reaction is non-specific and it has been shown that of the chromogenic material in the red cells of normal persons, only about 30-50% is creatinine. However in plasma, 80-100% of the chromogenic material in normal persons, and 70-80% where advanced renal insufficiency is present, can be shown to be creatinine. Thus in the performance of the test, plasma or serum is used. Because of the relatively high concentration of creatinine in the urine with subsequent dilution in the determination, practically all of the colour developed is due to creatinine.

The wavelength of measurement of the density of colour developed in the test (viz. 515 mμ) is such that the density of the blank is as low as possible, that of the test solutions being high enough to permit accurate measurements. This is illustrated in Figure 1.

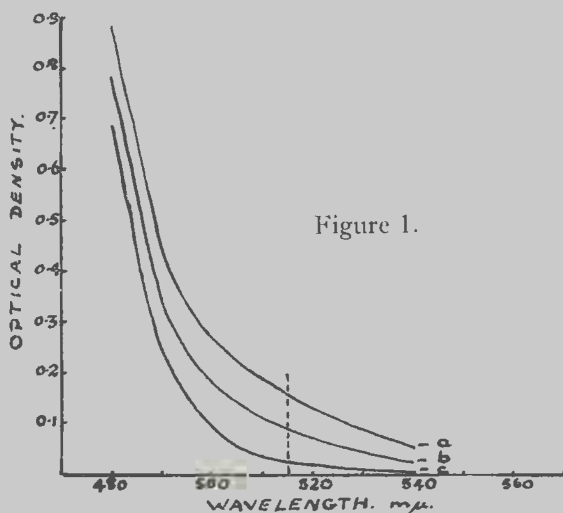

Figure 1. Absorption spectra of coloured solutions obtained in creatinine determination, for a blank (c), blank plus 10μg of creatinine (b) and blank plus 20μg of creatinine (a).

Colour development obeys Beer's law in the range of concentrations likely to be encountered.

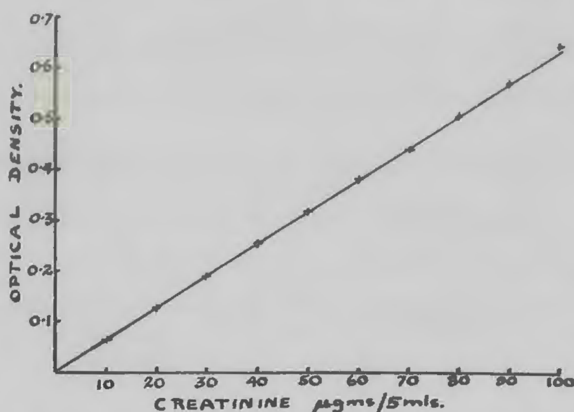

Figure II.

Figure 2. Plot of optical density versus creatinine concentration, for the range equivalent to plasma creatinine concentrations of 0—10mgms./100mls. (0.100μgms. per tube.)

(The data for figures 1 and 2 was obtained with a Unicam SP600 spectrophotometer.)

If required for any reason, the creatinine concentration can be determined by including a standard creatinine solution with the test solutions. Where concentrations are found from a previously constructed calibration curve, such as Figure 2, then the time and conditions of colour development must be rigidly controlled. Temperature in particular has a large effect on the density of the colour development.

The time allowed for colour development must be adhered to always, but strict control of temperature is not necessary if clearance figures only are required, or where a standard solution is included with each set of determinations.

PERFORMANCE OF THE TEST

Brod and Sirota. J. Clin. Invest. 1948, 27, 645.
Shoch and Camara. Methods in Medical Research 1952, 5, 214.)

The patient is given a low protein diet (40mgms. per day) and a complete 24 hour collection of urine is made, the urine being preserved by the addition of 10mls. of toluene to the winchester, or by acidification. At some suitable time during this collection period, 10mls. of blood are withdrawn under fasting conditions.

Alternatively, collection of urine over one or more shorter periods may be made, a specimen of blood being removed during the period as before.

Creatinine determination

(Bosnes and Tauski. J. Biol. Chem. 1945, 158, 581.)

(a) Serum (or plasma).

To 2mls. of serum (or plasma), 5.5mls. of water are added, followed by 1.0ml. of 10% sodium tungstate and 1.5mls. of 0.8N sulphuric acid.

After standing for 10 minutes, the mixture is centrifuged at 3000 r.p.m. for 10 minutes, and the supernatant fluid filtered, using a small paper to ensure maximum recovery of filtrate.

5mls. of this filtrate are placed in a test tube, 2mls. of picric acid are added and mixed. 2mls. of 0.75N sodium hydroxide are then added with mixing.

(b) Urine.

The most satisfactory dilution of urine samples is 1:200 to 1:300. In practice, a dilution of 1.300 will be found to be satisfactory in the majority of cases. 5mls. of this dilution are treated in the same manner as for the serum filtrate.

5mls. of water as a blank are similarly treated with alkaline picrate at the same time.

The optical densities of the three solutions are read at 515mu, exactly 20 minutes after the addition of the sodium hydroxide.
Calculation:—

Let x = Corrected optical density of final colour of serum filtrate (i.e. O.D. serum filtrate colour—O.D. blank colour.)

y = Corrected O.D. of final colour of urine dilution.

d/s = Dilution of serum.

d/u = Dilution of urine.

v = Urinary excretion in mls.

t = Period of urine collection in minutes.

Then the Creatinine Clearance

$$= \frac{U \times V}{P}$$

$$= \frac{\dfrac{y}{d/u} \times \dfrac{v}{t}}{\dfrac{x}{d/s}}$$

$$= \frac{y \times v \times d/s}{x \times t \times d/u}$$

If d/s = 1/5
d/u = 1/300
t = 1440 minutes (24 hours)

Then Creatinine Clearance

$$= \frac{yv}{x} \times 0.0416 \text{mls./minute}$$

Where the body surface area of the patient differs significantly from the standard surface area of 1.73 sq. metres, the clearance figures should be corrected, using the factor $\dfrac{1.73}{\text{Surface area of patient}}$. The surface area may be found from nomograms included in many texts, given a knowledge of the height and weight of the patient.

Due to the difficulty of colour matching, and the relatively high blank density, the method cannot be readily adapted for use with a visual colorimeter.

The normal range of values corrected to standard surface area, established for this method for both males and females is 100-140mls./minute.

Reagents:

1 10% sodium tungstate

2 0.8N sulphuric acid

3 0.04M picric acid. Picric acid saturated at room temperature is approximately 0.08M. This is diluted 1 in 2 to give a 0.04M solution.

4 0.75N sodium hydroxide ($\mp$ 5%) Dilute 15mls. of 20% sodium hydroxide to 100mls. with distilled water.

DISCUSSION

In normal uncontaminated urine, the creatinine concentration remains substantially constant for a period of at least one or two days even at room temperature. However, due to the range of conditions encountered clinically, with some urines being heavily contaminated as for example in pyelonephritis, precautions should be taken to inhibit bacterial activity by the addition of toluene, or by acidification.

Contrary to the results found by some earlier investigations, the alkaline picrate-creatinine colour is found to obey Beer's Law over the range of concentrations illustrated in Figure 2. Deviations which have been illustrated may be put down to instrumental errors and not to deviations from Beer's Law. The behaviour of the particular instrument used must therefore be established.

The table below shows a few values obtained in this laboratory, not as a proof of the validity of the method, but simply as an illustration of some results determined in practice.

Creatinine Clearance	Urea Clearance	Glomerular Filtration Rate Rate (Thiosulphate Clearance)
124 mls./minute	107%	108 mls./minute
83 mls./minute		73 mls./minute
57 mls./minute	33%	42 mls./minute
34 mls./minute		21 mls./minute
1.6mls./minute	1.7%	1 ml./minute

In conclusion, it may be stated that from experience of the test in this laboratory, the results obtained by its use warrant its inclusion in the range of methods of a hospital laboratory. Due to the relative simplicity of reagent preparation and lack of need for accurate standards, the method should appeal in particular to the smaller laboratories.

ACKNOWLEDGMENT

I wish to acknowledge the helpful advice and criticism of Dr. F. H. Sims, Chemical Pathologist, in the preparation of this article.

A METHOD FOR THE RAPID ESTIMATION OF TOTAL CHOLESTEROL

C. E. FELMINGHAM

(*Pathology Department, Whangarei Hospital*)

INTRODUCTION

The most popular methods for estimating Total Cholesterol in this country, seem to be those which rely on the Lieberman-Burchard reaction for final colorimetric analysis. Recently, however, we read of a modification of the Zlatkis method, which utilises ferric chloride in concentrated sulphuric acid as the colour reagent, and which seemed to possess several advantages over the

older methods. We gave this new method a trial, with such complete satisfaction, that we would like to bring it to the notice of those who are not as yet acquainted with it.

Deficiencies of Lieberman-Burchard Techniques

These techniques, of which there are a number of varieties (not differing greatly from each other) and most of which we have tried at one time or another, possess the common faults of being time-consuming, requiring numerous manipulations, and have a rather insensitive end-point. This latter is notoriously unstable in respect to light, moisture, temperature, and time. We have found it to be seriously affected by humidity in the air, and by room temperature, although of course, allowing the colour reaction to take place in the refrigerator, eliminates both temperature and light as variants.

Modified Zlatkis Method

For the benefit of those who may have missed the original article (1), we reproduce its working method below:

Reagents: (1) Iron Stock Solution—Dissolve 2.5gm of $Fe\ Cl_3.\ 6H_20$ in 25.0 ml. of glacial acetic acid. Preserve in the freezing compartment of the refrigerator and thaw when needed.

(2) Colour Reagent—Pipette 1.0 ml. of the Iron Stock Solution into a 100ml. volumetric flask, and dilute to the mark with conc. Sulphuric Acid, with continuous swirling. Discard when any precipitate becomes evident.

(3) Cholesterol Standard Solution — Dissolve 100gm. of pure dry cholesterol in glacial acetic acid and dilute to the mark in a 100ml. volumetric flask.

Method: 1 Pour about 10ml. of a 50:50 alcohol-acetone mixture into a 25ml. volumetric flask, add 1.0ml. of serum with vigorous swirling, and bring to the boil in a hot water bath, mixing constantly while heating, to avoid bumping.

2 Cool, and dilute to the mark with the alcohol-acetone solution.

3 Filter through Whatman 42 paper, keeping the funnel-top covered to prevent evaporation.

4 Pipette a 2.5ml. aliquot into a 6" x 1" test tube and evaporate to dryness in a boiling water bath.

5 Pipette 3.0ml. of glacial acetic acid into the tube, and warm about 30 seconds in the water bath, until the residue comes off the glass.

6 Into each of the three 6" x 1" test tubes, pipette 0.1ml. 0.2ml, and 0.3ml. respectively of the cholesterol standard solution, and dilute each to 3.0ml. with glacial acetic acid. These represent 100mgm, 200mgm, and 300mgm standards.

7 To each of the five tubes, add 2.0ml. of the colour reagent, mix thoroughly, allow to cool, and measure the absorbance at 560m μ, using a 10mm curette, and a water blank. We have found a green filter (such as that supplied with the EEL Colorimeter) to be satisfactory with simple P-E colorimeters. The calculation is:—

$$\frac{\text{Optical Density of Test}}{\text{Optical Density of Standard}} \times \frac{\text{Strength of Standard Used}}{1}$$

When we first started work on this method, we had only a simple EEL P-E colorimeter at our disposal, and found it necessary to add 1.0ml. of distilled water to each tube, before adding the colour reagent, to reduce the final colour sufficiently to enable us to obtain readings. However, we now have a more elaborate EEL absorptiometer, and find that the use of 2.5mm cuvettes instead of 10mm, where necessary, obviates the addition of distilled water.

Comparison: In the performance of several series of estimations and recoveries, comparing our Lieberman-Burchard method (2) with the Zlatkis method described above, several definite advantages of the latter method became obvious, and we find the Zlatkis method superior to the Lieberman-Burchard on the following counts:

(i) The standards and final figures of the Lieberman-Burchard method fluctuated, particularly in wet weather, while the Zlatkis technique was not affected by time, temperature, light, or atmospheric moisture.

(ii) With the Zlatkis techniques we were able to recover cholesterol added to sera of known cholesterol concentrations, with far greater accuracy than with other methods.

(iii) The Zlatkis technique requires five pipettings as against seven, and needs fewer manipulations.

(iv) From start to finish, we can complete a Zlatkis cholesterol determination in 30 minutes as against 70 minutes for the Lieberman-Burchard. These times are for duplicate determinations of both methods, as all our work is performed in duplicate.

On a cost per test basis, we estimate that the Zlatkis method costs 2d. more than the Lieberman-Burchard, excluding the labour involved, but consider that this is more than outweighed by the saving in man-hours, the ease of manipulation, and the greater accuracy obtained.

SUMMARY

A new, rapid, and accurate total cholesterol method, is compared with the customary Lieberman-Burchard methods, with favourable results. We believe that the reproducibility and simplicity of the new method will find favour with the average New Zealand Hospital Laboratory.

REFERENCES: (1) *Zak et al.* Rapid Estimation of Free and Total Cholesterol. Am.J. Clin.Path. 24,1307-15, 1954.

(2) *King.* Total Cholesterol. Micro-Analysis in Medical Bio-chemistry. 2nd Ed. P39.

* WATER AS A SOURCE OF INFECTION

D. WHILLANS

(*Department of Pathology, Auckland Hospital*)

To me, one of the most interesting points about tracing infections carried by water is that it is rarely that the organisms which cause disease are looked for, even when we are determining the suitability of the water for human consumption or for industrial purposes. The reason is, of course, that they will have done their fell work and probably disappeared by the time an epidemic has arisen, leaving it to be carried on by secondary infection.

The other interesting point which was forced upon me in reading for this paper is that the public understanding of hygiene, though still at a low level, is far in advance of that which pertained during earlier times, and I could not but be struck by the public tolerance of every kind of filth even as close to us as the Victorian era.

Cholera

It is instructive to consider some of the great epidemics of the past and the disease of cholera immediately springs to mind. Cholera is now an unimportant disease in England and as far as I know has never been seen in New Zealand. It was endemic in India for centuries and did not spread to the rest of the world till 1817 when between then and 1823 it spread to many parts of Asia ravaging the whole of Europe throughout the years 1832-33. In this year there were 4000 deaths in London alone from this cause. It spread to Canada and New York by the agency of fleeing Irish immigrants and there was a heavy loss of life in Cuba and Mexico.

Further pandemics in 1864-75, 1883-96 with a sixth which started in 1902 have gradually been confined to Asia, Egypt and the Southern Countries of Europe, and though frequently imported into England and America has not succeeded in gaining a foothold since 1873 and with improved sanitary arrangements will probably retract from the West Indies, Southern China, the Philippines, Persia, Arabia and South-East Russia and will be confined once again to a few endemic foci such as the delta of the Ganges.

One of the most famous of the water borne outbreaks of cholera is that of Hamburg in 1892. Hamburg, and its suburbs Altona and Wandsbeck each had a different water supply. Hamburg drew its water from the River Elbe above the city and did

* Part of a symponium on "The Bacteriology of Water" at a meeting of the Auckland Branch of The New Zealand Institute of Chemistry.

not filter it. Altona drew its water from the river below Hamburg after the refuse from 800,000 people had been poured into it but, however, installed a highly efficient sand filter to make the water potable. Wandsbeck not only obtained its water from a relatively non-polluted lake but also filtered it.

Hamburg Epidemic

In August 1892, Hamburg had its first case of cholera and by the end of the month there were 1000 fresh cases a day. The epidemic finally ended with a total of 18,000 cases with 8200 deaths. These were almost all in Hamburg, Altona and Wandsberg having a low incidence of infection and almost all these cases were traced to a start in Hamburg. The epidemic gave an interesting example of test and control, for in many of the streets where Hamburg met Altona, both water mains ran together and served either one house or another in the street. It is significant that without exception, the cases of cholera occurred in the house supplied with Hamburg water and not in those supplied with Altona water. The Elbe was probably infected initially from the excreta of cholera patients in barges on the river opposite the Hamburg water intake. Finally, cholera vibrios were isolated from the river below the mouth of the Hamburg sewer.

The spread of cholera is furthered by the cholera carrier, which though not so serious a problem as the typhoid carrier is still an important source of infection. It is considered that although 90% of cholera carriers are free of the disease in a fortnight, and 99% in a month, there are cases where the vibrio may persist from 3-4 months and even as much as up to a year. Here the vibrios are harboured in the gall bladder and are excreted intermittently in the urine. It is of interest that, unlike a typhoid carrier, the carrier of cholera, though apparently perfectly well, may finally die of the disease thousands of miles from the source of the infection.

In the case of the Hamburg epidemic, no less than 300 places in Germany were infected by carriers from Hamburg.

English Outbreaks

The most famous of the English outbreaks was the Broad Street Pump outbreak of 1854. This occurred in the parish of St. James in an area which housed 222 people to the acre, and in this outbreak about 10% of that population died. Ideas of hygiene were very elementary in those days, and in the area there were many cesspools. In the middle was the Broad Street Pump. This was a shallow well only 28-30ft. deep and not only did the sewer, in poor repair pass within a few feet of it, but there was

a cesspool within 32 inches of the brickwork of the well. The well had a great local reputation, and the water was sent far and wide all over London, some people from distant points even sending their servants to collect it. It was cold and above all sparkling —with the carbon dioxide derived from sewage—so that it can be seen how the stage was set for the tragedy which followed.

It is thought that a child suffered an attack of cholera in the house near the well and that the dejecta and the washings of the linen were poured into the cesspool and from thence infected the pump water. The outbreak was soon on its way and with all the cesspools adding their quota of cholera organisms the epidemic increased explosively. When finally the infection was traced to the pump water, and it must be remembered that this was but an exercise in deduction in those days for it was not until 1884 that Koch announced the discovery of the vibrio, the epidemic was halted and finally died down. There is evidence, however, that it was decreasing of itself by then and it is interesting to speculate now whether this could have been due to a phage on the cholera organism which had come into play. This is not so fanciful as it seems, as clear cases of this action are known.

Finally, the sewage arrangements were bettered, the wells closed and the epidemic was at an end.

Enteric Fever

The next great source of explosive outbreaks is that of Enteric fever. This has been noted from the days of Hippocrates to the present time and the primary cause is the ingestion of human dejecta which, being carried into the drinking water, passes into food, milk, cheese, ice cream or contaminates shell fish, water cress etc.

I can remember the pipi beds at Port Ahuriri, in Napier being closed to the Maoris in 1928, owing to the incidence of enteric fever, and there are numerous accounts of outbreaks due to the storing of oysters in typhoid infected waters.

In contrast to the case of cholera, there have been numerous outbreaks of typhoid in England of fairly recent years, such as the one at Bournemouth in 1936 which was water and milk borne and Epping 1931 which was sewage, water and milk borne.

Typhoid fever is a very serious disease, but is little known in Auckland owing to the excellent watch on cases and contacts provided by the Health Department. Further factors must be, of course, the fairly high level of immunity given by the widespread use of TAB vaccine in travellers overseas and in those who join the armed forces, and in the treatment of sewage, but in view of this latter being a very vexed question I do not propose to say anything further about it.

There was a very instructive outbreak of typhoid fever at Croydon in England in 1937. Here the wells had been in use for 49 years but at last work had to be carried out underground in one of the adits. This continued for a month, and though at first the water was pumped to waste, it was later pumped to the reservoir without filtration or chlorination. Open buckets were used for urination by the workmen and though defaecation underground was denied, one of the workmen was a carrier of typhoid, and in all there were 342 cases of the disease, with 43 deaths. It is perhaps of significance that on phage typing of the typhoid organism isolated from the workman it was found to be of the same type as that causing the outbreak. No typhoid bacteria were, however, isolated from the well.

Apart from the workman, there were several possible sources of infection, such as a latrine dug in the chalk over the adit, and in the fact that the roof and the surroundings of the reservoir were much frequented by the public.

It is obvious that here there was a total lack of recognition of the dangers, that the works were poorly maintained and supervised, that there was a failure to exclude the well from service during the repair work, that the sanitary arrangements for the workmen were poor, and that the workmen should have been medically examined and should have had Widal and Vi agglutinations tests performed before being allowed to commence work.

It is significant to note that in England the case rate for typhoid fever has dropped from 371/M in 1871 to less than 1/M in 1945 and from 150/M to 2.2/M in the same years in America.

Further, typhoid now tends to be a disease of small communities and particularly to be milk borne.

Paratyphoid

Not only does one suffer from typhoid fever, but the paratyphoid fevers are also implicated in water borne infection, thus in 1940 a stream in Dundee where a stream was used to wash dairy utensils was found to be a source of Paratyphoid B infection. The organism was found in the effluent entering the stream from a sewage plant and even in 1947 in Suffolk Paratyphoid B was found in a stream.

However, Paratyphoid fever and dysentery are mostly carried in infected food, but there was an outbreak with interesting features in Leicester in 1950 where the water carried gasteroenteritis and dysentery infection.

On December 20th. 1950, the Health Department in that town was notified that a number of employees of a certain factory were suffering from vomiting and diarrhoeae. As the water was under suspicion, it was checked both chemically and bacteriologically and was found to be so similar to that of the River Soar that

it could not have been derived from the water mains of Leicester. Both town water and river water were used in various processes in the plant and it was found that owing to the river water proving unsuitable for a certain process in the plant, the engineer had interconnected the river and the town supply, and on forgetting to close the connection, had been pumping the river water at a pressure of 240lbs into the town supply of 25-30 lbs. This had contaminated not only the works supply but also the supply to the houses around.

Swabs were suspended in the river just above the intake to the works, and both Salmonella typhi-murium and Shigella sonnei were isolated from them.

Cross connections of sewer and drinking water are of course prohibited but is salutary to know that a similar occurrence, which fortunately did no damage, occurred recently not far from Auckland, so that we should never take up the attitude that "it couldn't happen here."

In the case of the mains in Leicester, it took no less than seven days for the bacteriology of the water in the supply pipes to return to normal after a thorough flushing.

An important point in the infection of water supplies is that the Courts rightly hold the water authorities responsible for sickness and death resulting from the use of polluted water. Thus in Orlean, New York in 1928, there were 248 cases of typhoid fever with 25 deaths and the cost to the authorities, and thus the community, was about $\frac{3}{4}$ million dollars. In this instance, bacteriological tests revealed the coliform organisms in the water two months before the epidemic broke out, but negligence and failure to look for the break in the pipe line, which later was found in a suction pipe within a few hours caused the later tragedy.

Tularemia

Other organisms are of course implicated in outbreaks of waterborne fever. There was an outbreak of Tularemia in Russia caused by the ingestion of B. tularensis. This is generally transmitted by the handling of infected carcases of ground squirrel, hares, jack rabbits, etc., but in the case cited, the drinking water was sucked in from the river, and though chlorinated during the day, was not chlorinated during the night. As the intake was through the bodies of dead rats, it is not hard to see how infection developed.

The "lemming fever" of Norway is thought to be of a similar nature and often follows the consumption of water which has been polluted by the bodies and excreta of the lemming. A further important point is that in this case it is thought that the organism may be able to pass through the intact skin and so cause disease.

Leptospira

Passing now from the bacteria to the leptospira, there are a number of recorded instances of outbreaks of Weil's disease due to a leptospira. Thus in Lisborn in 1931, at least 126 persons were infected with a case mortality of 25%. The water was infected in its passage underground to a fountain through an open conduit by rats from a neighbouring sewer. Here again it is thought that infection can take place directly through the abraded skin or through the mucous membrane. The source of the infection is thought to have been Leptospira from rats urine as the rat is a frequent carrier.

A further instance of an outbreak due to Leptospira occurred in Wyoming in 1942 where, associated with the hot springs at Jackson Hole, Wyoming, there was an epidemic of "Wycon fever," so called. It was shown by investigations 11 years later that infection was almost certainly due to L. canicola. All swimmers in the pool at this time became ill and it is considered that either or both of the drinking and swimming water were contaminated by an infected dog, deer, elk or horse whose excreta contained L. canicola.

Infections are common in damp, badly drained and rat infested situations such as in coal mines, on the Western Front of 1914-18, in the rice fields, the canals of Holland and in sewer workers and fish handlers of England.

Viruses

Coming now to viruses, the picture is at the present confused, but there is one paper which shows that Coxsackie or "C" viruses which can be isolated from the faeces of patients and also apparently healthy individuals and from sewage is rather resistant to chlorination and may require from seven to 46 times as much chlorine to kill as a suspension of E. coli, but that 1.4 ppm of free chlorine would in general be sufficient. In a mixture of faeces, however, it might well be more resistant, though flocculation of the material and its filtration from the water would achieve a desirable end.

An interesting account was given some years ago of an outbreak of viral hepatitis spread by drinking water and by contact in a remote part of Pennsylvania where the drinking water from a well was probably polluted by seepage from a cesspool higher than the well and some 50ft. away in the shale. A high level of ground water was a factor at the time and there was clear bacteriological evidence of pollution by reason of the high coliform count which returned to normal on the source of pollution being removed.

Amoebae

It is instructive, also to consider the amoebae. Thus infection with Entamoeba histolytica is caused by the infection of food and drink with cysts of the organism. It is a common source of infec-

tion in the tropics during military operations and is found in all tropical countries where human excreta are used to fertilise vegetable gardens and where wells are dug so that human excreta can gain entrance.

The cysts on being ingested form trophozoites and give rise to the disease which is a fairly serious one. The cysts themselves are fairly resistant and may survive for three months at freezing temperatures or ten days at 20°C. and are only killed by hyperchlorination. In New Zealand it has never been a problem, and all known cases are kept under surveillance by the Health Department. It is, however, not impossible for E. histolytica infections to occur here and there is clear evidence that a few cases of infections have actually occurred.

Worm Infections

Turning finally to the worm infections, whipworm or Ascaris infections are common in tropical countries with a high rainfall, high humidity and dense shade. The egg, on being swallowed, forms a larval state and this grows to a worm in about three months.

Rather more involved is the case of infection with Ascaris lumbricoides. The fertile eggs on being ingested form larvae which burrow through the small intestine and make their way through the right heart to the lungs. After two moults they break through the pulmonary capillaries and are carried up into the epiglottis where they are swallowed, and after two further moults mature in the intestine. It is possible that infection through the intact skin can occur from the larval stage. Although it is normally an infection of the tropics and the sub-tropics, there is a record of a waterborne infection in Hertfordshire in 1923 where a local well was infected and gave rise to a series of cases.

One of the most important of the Nematode parasites of man is the Hookworm. This is distributed over most of the world, and is found particularly in backward communities. The eggs form embryos in moist soil and these, on contact with intact human skin, usually the interdigital spaces of the toes, reach the venules and are carried from there through the heart to the lungs, where they migrate to the air sacs, and thence to the stomach and on passing to the intestine develop into adult worms in about five weeks. This is a common infection of the Americas and in the miners of Europe and is common in the tropics and in the Pacific Islands.

An important and rather horrible worm infection is that of the Guinea Worm. The female of this species is about 120 cm. in length and migrates through the tissues, till coming near water it bursts through a blister in the skin and discharges a host of motile larvae. These are taken up by small crustaceans of the species Cyclops, develop in them and are swallowed by swimmers

or are drunk in infected water, developing into adult worms in 8-12 months. Simple filtration, liming etc. kill the crustacean which is however resistant to heavy doses of chlorine.

Further examples of organisms capable of infecting water are the Cestodes, where according to the species, either the ovum or the hexacanth embryo on being swallowed becomes after several stages a scolex and then a worm. These are mostly dependent on man as the definitive host. In the case of T. echinococcus, the cause of hydatid disease, the dog is the usual definitive host. Hydatid disease is common in parts of New Zealand and in Hawke's Bay, where I began my training it is quite common. Here the dog carries the infection, pollutes drinking water which is swallowed and infection results.

In the case of D. latum the course of events is longer, here the ovum forms the embryo which enters a crustacean which is swallowed by a fish which is eaten by a human in an insufficiently cooked state and after this "House that Jack Built" story the human is infected.

My last example is that of Schistosomiasis in which the ova are hatched to miricidia. These are ingested by water snails in which they form cercariae, which on passing into the water penetrate the skin of the bather or drinker. They then pass to the lung and from there to the portal circulation where they mate, and finally pass ova around the bladder. These penetrate the bladder and are discharged into the water to begin the cycle again.

This is a common waterborne infection of Egypt and may be kept under control by removing the snails which act as the intermediate host and by keeping the water containing the miricidia for two days when they die. Insanitary disposal of human faeces, urine or sputum provides the source of the pollution of the water from which the intermediate hosts becomes infected.

Cercaria dermatitis from non-human schistosomes is known in Canada and America and may cause urticarial whealing in infected water.

MENINGITIS CAUSED BY LEPTOSPIRA POMONA

JEANNETTE GREY

(*Pathology Department, New Plymouth Hospital*)

Only recently has leptospirosis become a common word in New Zealand hospital laboratories. Leptospiral infections of cattle, pigs and humans had been frequent in many countries for many years. In 1937 Clayton and Derrick of Australia described a new species of leptospira which they called L. pomona after the district of Pomona (Queensland) where it was first isolated. Since 1952 when L. Kirschner (Dunedin) described the first cases of L. pomona infection of humans in New Zealand, L. pomona has gradually become more and more noticed in our hospitals, as the cause of febrile illnesses, which were previously undiagnosed, or merely labelled "influenza."

Cases of L. pomona infection from districts around New Plymouth have all been diagnosed by serum agglutination tests done at the Leptospiral Reference Laboratory at Medical School, Dunedin. The histories of the 19 cases diagnosed from New Plymouth Hospital during the last 18 months reveal that nine of them showed sufficiently severe meningeal symptoms to warrant lumbar puncture. The cerebrospinal fluid reports are interesting for the total leucocyte counts and protein levels are all moderately raised to within the same limited range, and of the seven cases where a glucose estimation was done, six gave normal figures. The C.S.F.s were all taken on the eighth or ninth day after the onset of the illness. All urine and blood count reports mentioned in the following case histories refer to tests done at the time of admission to hospital. All were male patients working on or connected with farms.

Brief Case Histories

CASE 1 (L.)—aged 17 admitted with P.U.O. after eight days of general abdominal pain, limb pains and headaches, and three days of neck stiffness, vomiting and érythematous rash over trunk. Suffused conjunctiva.
C.S.F.: 130 leucocytes/cu.mm. 90% polymorphs. Protein: 59 mgs/100ml.
Urine: Small amount albumin; occasional leucocytes and casts.
W.B.C.: 16,400 per cu.mm. with 78% lymphocytes, B.S.R.: 57 mm. in 1 hour. Discharged after one month. (Subsequent tests on sera revealed that at least 5 cows and 2 pigs were carrying L. pomona on the farm where patient worked.)

CASE 2 (A.C.)—aged 29, admitted after a week of headaches, nausea, limb and neck pains. Temp. 101. Vomited on admission.
C.S.F.: 170 leucs./cu.mm. 36% polymorphs. Protein: 41 mgs/100 ml.

W.B.C.: 4100/cu.mm. with normal differential. B.S.R.: 16 mm. in 1 hour. Discharged after nine days.

CASE 3 (W.B.).—aged 38, admitted with P.U.O. after five days intermittent vomiting and diarrhoea, headaches, oliguria and neck stiffness. Temp. 101.

C.S.F.: 445 leucs/cu.mm. 87% polys. No protein done.
Urine: Moderate amount albumin. Blood T.N.P.N.: 57 mgs/100 ml.

W.B.C.: 9300/cu.mm. with 78% polymorphs.
Patient discharged after 10 days.

CASE 4 (C.L.)—aged 51, admitted (as possible intestinal obstruction) after four days pyrexia, vague abdominal pain and some vomiting.

C.S.F.: 255 leucs/cu.mm. 48% polymorphs. No protein done. Blood T.N.P.N.: 106 mgs/100 ml.
Urine: Large amount albumin, occasional leucs, and many granular casts.
W.B.C.: 12,900 per cu.mm. with 88% polys. and toxic granules. This patient's kidneys were "hard hit by the infection."

CASE 5 (W.R.).—aged 38, admitted with meningo-encephalitis after a week of severe headaches, fever, vomiting and body pains. Mild conjunctivitis. Temp. 101.

C.S.F.: 227 leucs/cu.mm. 53% polys. Protein: 64 mgs/100 ml.
Urine: Small amount albumin, occasional leucocytes.
Patient discharged after five days.

CASE 6 (B.G.).—aged 20, admitted as influenzal relapse, after a week of stiff and aching limbs and joints, headaches and intermittent pyrexia. Painful stiff neck and erythematous spots on legs. Temp. 101.

C.S.F.: 435 leucs/cu.mm. 54% polys. Protein: 61 mgs/100 ml. Glucose: 41 mgs/100 ml. (The lowest figure recorded in seven cases; the others were normal.)
Urine: Small amount albumin and occasional leucocytes.
W.B.C.: 9700 per cu.mm.

CASE 7 (C.B.).—aged 29, admitted (as possibly acute nephritis) after three days nausea, abdominal and limb pains, headaches, stiff neck and oliguria. Temp. 101. After admission, intermittent vomiting for four days; erythematous spots on legs.
C.S.F.: 125 leucs/cu.mm. 80% polys. Protein: 50 mgs/100 ml.
Urine: Moderate amount albumin and occas. leucs. and casts.
W.B.C.: 5050/cu.mm. Patient discharged after 10 days.

CASE 8 (S.S.).—aged 28, admitted (as acute nephritis or infective hepatitis) after four days of headaches, vomiting and limb pains. Temp. 103.
C.S.F.: 325 leucs/cu.mm. 73% polys. Protein: 75 mgs/100 ml.
Urine: Small amount albumin and occasional leucs. and casts.
Normal W.B.C. and differential. Patient discharged after 11 days.

CASE 9 (S.B.).—aged 7, admitted after four days of headaches, general abdominal pain, vomiting, slight neck stiffness. Temp. 103.
C.S.F.: 450 leucs/cu.mm. 64% polys. Protein: 62 mgs/100 ml.
Urine: Trace albumin and occasional leucs. and casts.
W.B.C.: 11,400. Discharged after 19 days.

SUMMARY

Nine cases of leptospirosis, with meningitis, caused by L. Pomona are described, with emphasis upon the laboratory findings in each case.

The cerebrospinal fluids show a leucocyte elevation of 100-450 per cu.mm., a protein elevation ranging from 40 to 64 mgs. per 100 ml. and a normal glucose figure with the exception of one case with a figure of 41 mgs/100 ml., which may be lower than normal.

Acknowledgments

I wish to thank Dr. Dennis Allen, Pathologist, for his helpful criticism of this article and Dr. L. C. McNickle, Medical Superintendent, for permission to publish the clinical notes.

Reference

Kirschner et al. N.Z. Medical Journal. Vol. 51, No. 282., pp. 98-108 (April 1952).

DEPARTMENT OF HEALTH
INTERMEDIATE EXAMINATION
HOSPITAL LABORATORY TRAINEES

Central Laboratory, Auckland Hospital, May 26th and 27th, 1955.

THEORETICAL EXAMINATION

Examiners: Dr. S. E. Williams, Mr. D. Whillans.

(Attempt all questions).

1 Describe in detail the method you would adopt to estimate the percentage of urea in urine. Draw a diagram of the apparatus used, and write brief notes on the underlying principles of the estimation.

2 Discuss fully the preparation, storage and use of material for performing routine antibiotic sensitivity tests by the method you use in your results?

3 How would you carry out a platelet count and estimate the bleeding and clotting time of a patient? Give the normal values for the methods described.

4 Give a table showing the interaction of cells and serum of all the ABO groups. Describe in detail your method of performing a blood grouping and cross matching for a non-urgent blood transfusion.

5 Describe your method for carrying out a bacteriological analysis of milk and reporting the results. List briefly the pathogens which may be found in milk.

6 Write brief notes on:—
- (a) Specific gravity.
- (b) Sugar estimation in C.S.F.
- (c) The disposal of sputum infected with M. tuberculosis.
- (d) The sources of error in blood sedimentation rate estimation.
- (e) The Postal Regulations for the transmission of pathological materials.

PRACTICAL EXAMINATION

Examiners: Dr. S. E. Williams, Mr. D. Whillans.

You will be allowed an hour to perform the work in each of the three groups. Label ALL work and ALL answers with your examination number. Do not discard any preparation.

You will be allowed one hour on Friday morning to complete Question No. 2.

1 (a) The packed cell volume of the blood provided, which has been collected in haematology oxalate is 42. Estimate the mean corpuscular volume. Show your working.

(b) Report on the three blood films, A, B and C, provided, and do a differential count on D or E.
(Eosinophila; myelogenous leukaemia; erythroblastosis normal count).

2 (a) Examine and report on a centrifuged deposit of the urine provided, test for albumin and estimate the percentage of sugar present in the specimen. Show your workings.
(Red blood cells, small amount of albumin, sugar 1.7%).

(b) Examine bacteriologically the broth culture provided.
(20 cultures provided, half being haemolytic staphylococcus and half haemolytic streptococcus. Candidate chose one.)

3 (10 minutes for each of the following sections).

(1) a Give the names of the parts of the microscope labelled.
b Read the two verniers on this mechanical stage.
c Write notes on this object. *(Incubator capsule).*

(2) What is this and how is it used?
(Seitz filter, set up for use).

(3) and (4) What ingredients are used in these preparations and for what purpose are the various substances included?

(3) *McConkey's medium, Hayem's solution.* (4) *Carbol fuchsin, Benedicts qualitative solution).*

(5) These two oxalate tubes are used for the collection of blood for Haematology and for Biochemistry. What is the difference in their preparation, and why?

(6) This is a tube of Robertson's Cooked Meat Medium. How is it prepared, and what is its use?

EXAMINATION RESULTS

The following candidates were successful in passing their Intermediate Examination for Hospital Laboratory Trainees held at Central Laboratory, Auckland Hospital, on May 26th and 27th 1955.

Misses S. A. Jury, D. G. Ludbrook, P. A. Furkert and Messrs J. G. Meredith, and A. Fischman, Auckland Hospital; Mr. D. J. Dunlop, Hastings Hospital; Misses N. M. Snowden, M. A. Kennedy and J. H. Johnstone, Waikato Hospital; Miss J. A. Mills, Rotorua Hospital; Miss M. M. Eales, Christchurch Hospital; Miss M. G. Woolley, Mater Misericordiae Hospital; Mrs. J. V. Dobbs and Miss A. N. Sheldon, Whangarei Hospital; Mr. G. A. Kuru, Dannevirke Hospital; Messrs J. C. Beattie and R. H. Jones, Cook Hospital; Miss J. B. Speden, Medical School, Dunedin.

MINUTES OF A COUNCIL MEETING OF THE N.Z.A.B. HELD AT THE WELLINGTON HOSPITAL, AT 10 A.M., 21ST MAY, 1955.

Present: Messrs McKinley, Donnell, Patterson, Rush-Munro, Bloore, Adamson Reynolds.

APOLOGIES: Miss Scott, Mr. Samuel.

MINUTES OF COUNCIL MEETINGS 14th and 16th July, 1954: Confirmed.

NEW MEMBERS: The following were elected: Misses L. Kirkup, S. Snook, S. O'Sullivan, P. Harrison, G. Prebble, N. Eccles, J. McKenzie, J. Levien, A. Brooks, Messrs T. Scott, M. Churchouse, J. Thomas, E. Clarke, J. Pryor (AUCKLAND), Miss G. Rutherford (NEW PLYMOUTH) Miss J. Styles (NAPIER) Miss K. C. Handley, Mr. A. C. Howell (TAURANGA), Mr. D. F. Henry, (WAIKATO), Miss B. L. Moore (CHRISTCHURCH), Mr. R. Smith (GREYMOUTH), Miss H. Chesterman (AUCKLAND), Miss P. A. Waters (BLENHEIM), Miss R. A. Donnelly (WAIPUKURAU), Mr. S. W. Entwhistle (TIMARU).

ALL JUNIOR MEMBERS.

Mr. R. Wales (WANGANUI), Mr. G. E. Pearmain (TAURANGA), Miss L. Evans (CHRISTCHURCH), Miss J. Sorensen (WELLINGTON). SENIOR MEMBERS.

RESIGNATIONS: The following were accepted: Mrs. S. Lawton (nee Jenkins), (BLENHEIM); Miss Blodorn (TIMARU).

CORRESPONDENCE: From Mr. K. B. Ronald, inquiring about examinations dates

Moved:

That application be made to the Director-General of Health for notice to be given to the association of examination dates.

(Adamson—Rush-Munro).

ANNUAL CONFERENCE, 1955: The President's action in transferring Annual Conference 1955 from Napier to Auckland was confirmed, and the setting up of a Conference Committee was authorised. (Bloore-Adamson). At this stage Dr. Mercer attended to report on action at the Pathologists' Conference about the proposed syllabus for the final examination. The interim report of his committee and the syllabi of Mr. Whillans and Mr. Reynolds have been referred to the Pathologists Executive under the chairmanship of Dr. Pullar. The pathologists have been circularised and are to report

to their Executive by the 15th June, 1955. Dr. Mercer then left after being thanked by the President. Mr. Olive as representative on the S.A.C. attended by invitation for discussions on correspondence about salaries.

FROM MR. CLAPSON, WAIKATO, re the payment of a Married Minimum Wage. Moved: That Mr. Clapson be informed that he should make application through his Hospital Board to the D.G.H. for consideration of his case as a special case. (Bloore-Adamson).

FROM CHRISTCHURCH MEMBERS re salaries of Staff Hospital Bacteriologists, suggesting that the annual increments be reduced from four to two of £80½ and £63¾. Moved: That the annual increments of the salaries of Staff Hospital Bacteriologists be reduced from four to two of £80½ and £63¾. (Rush-Munro—Bloore).

FROM MR. V. JONES, AUCKLAND, re salaries asking that consideration be given to the subject of salaries, the payment of penal rates, etc., and that this matter be placed on the agenda for the coming Conference. After some discussions it was moved that a recommendation from Council be sent to Annual Conference, 1955 for approval that the Director-General of Health be asked that immediate steps be taken to bring salaries of Hospital Bacteriologists into line with those recently established in the Public Service. (Reynolds—Bloore).

FROM MR. A. FISCHMAN, AUCKLAND (through Mr. Rush-Munro), suggesting that local branches be formed. Moved: That Mr. Fischman be informed that the Council feels that it is not the time for the official formation of such branches but that they could be organised locally and unofficially. (Reynolds—Bloore).

MR. SAMUEL'S RESIGNATION FROM THE COUNCIL was accepted with regret.

BALANCE SHEET FOR CONFERENCE 1954 was accepted.

ACCOUNTS: In response to a suggestion from the Hon. Treasurer regarding a figure in the accounts of unpaid subscriptions prior to his taking office it was moved that subscriptions owing prior to 1st April, 1954 be writen off as bad debts. (Bloore—Adamson).

Authority was given for the following accounts to be paid:

Item		
Auckland Trade Linotypes	£1 2 6	
Leightons	£1 18 6	
Mr. Reynolds (Peoples Palace)	£1 10 0	
Council Meeting Expenses:		
Adamson	£4 0 0	
McKinley	£4 4 0	
Toll Calls	£2 16 1	£7 0 1
Rush-Munro	£12 18 0	
Donnell	£14 1 0	
Patterson	£14 1 0	
Bloore	£3 15 0	

Mr. Bloore left for his plane at this stage.

GENERAL BUSINESS:

JOURNAL EDITOR'S RESIGNATION. After some discussion it was decided to ask Mr. Murphy to edit the October 1955 Journal in order to give the new Editor a reasonable time to take over Editorial affairs.

REPORT ON CONFERENCE 1955. Mr. Patterson reported verbally of satisfactory progress on Conference arrangements, from the Conference Committee in Auckland.

MR. GARNHAM, NAPIER. In reply to correspondence from Mr. J. B. Rankin, Napier, it was moved that Mr. Garnham be told that the Association supports his application to the D.G.H. to sit for the Final Examination. The Council suggests that if this is agreed to he should attempt the Intermediate Examination first as a preparation, and if successful, then the Final Examination as soon as possible afterwards.

(Rush-Munro—Patterson).

In 1896, Mr W. G. Pye, after several years' experience at the Cavendish laboratories of Cambridge University, started his business of manufacturing scientific instruments and laboratory apparatus for educational purposes. Today the Pye Group of companies includes in its manufactures the widest possible range of instruments, equipment and accessories.

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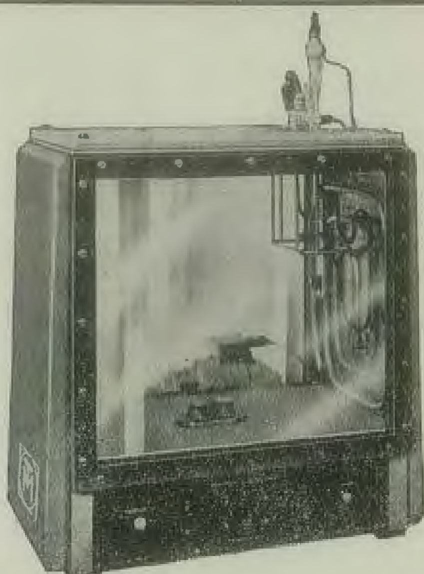

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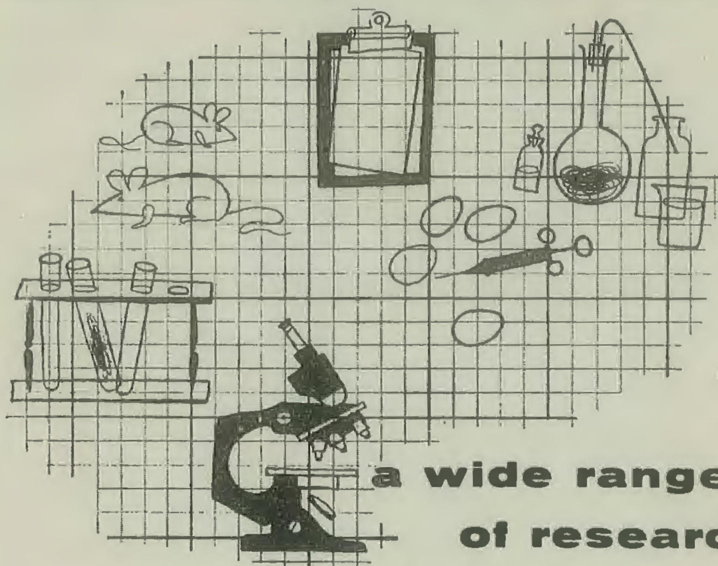

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