been mistaken for a renal condition. We would like to point out that had we seen the case with the bad chest or the case with irregularity of the heart or those with the very severe backache at any other time than during an obvious epidemic we would have sent them into hospital under varying diagnoses, and thus almost certainly have spread the epidemic.

We consider that the period of incubation is between 24 and 48 hours, but it may be as high as 96 hours.

Treatment

All patients were placed on a fluid diet on the first day of the disease and all received diaphoretic mixtures. The diet was increased each day, and the diaphoretics were continued until the patients were fit to leave bed. Cough, where troublesome, was treated with a codeine linctus.

Sulphapyridine treatment was resorted to for all patients whose temperature had risen above 103° F. The dosage was 2 g. as an initial dose and 1 g. four-hourly. No patient had more than 8 g., and the temperature was normal in each case before that dosage had been taken. It did appear as though the sulphapyridine had definitely assisted the drop in temperature. The literature gives conflicting reports on the efficiency of sulphapyridine in influenza A, but it seems logical that in a disease in which the temperature is high owing to virus infection and in which most throat swabs demonstrate the presence of pneumococci or streptococci the use of sulphapyridine or some other drug of the group in severe cases is indicated.

The weakness that so many writers describe after influenza was not apparent in this series of cases, and all patients were fully fit seven days after leaving bed. These seven days were spent doing light duties only. There were no serious complications, except in the case proved to have tuberculosis. It may well be that had this patient not developed influenza his tuberculosis would not have been brought to light at the early stage at which it became apparent.

As stated by many writers, the epidemic occurred in two waves; and we noticed also that the curve of incidence of the cases was not dissimilar in shape to an average temperature curve of the severer cases (Fig. 2). It was observed, too, that

FIG. 2.—Average temperature curve.

the epidemic began in warm dry weather with a mild breeze which persisted from April 14 to April 21. The weather then deteriorated, and there was some rain; but on the 24th it improved, and became quite warm by the 26th. It may be that, somehow, the weather had an effect on the incidence of the disease, as rain fell again on April 28 or 29.

The ultimate object, of course, of the study of any outbreak is to indicate some way of preventing another occurring and, in the case of influenza, especially of preventing such a catastrophe as the 1918-19 epidemic. We can only state that, in our opinion, isolation is of the very greatest importance. Every case of even mild cold that occurs during a known epidemic should be rigidly isolated. By far the greatest proportion of our cases occurred on a satellite aerodrome, and all cases from that aerodrome were taken to the main aerodrome to be nursed in houses separated from all the other buildings on the camp. The only patients reported from the main camp after April 18 were medical personnel and ambulance drivers. Approximately half the medical personnel looking after the cases developed influenza. These medical personnel did not leave sick quarters or associate in any way with other members of the camp. It is probably for this reason that no cases occurred among the thousands of airmen on the main camp.

So far as other preventive methods go, it would certainly be worth while trying experimentally to vaccinate large bodies of men against influenza A a few months before the reckoned time (if it is possible to reckon!) when another epidemic should occur. The work of Horsfall (1939, 1941) in this connexion is especially interesting.

Sterilization of the air by ultra-violet light and propylene-glycol spray has been attempted elsewhere, but in view of our findings that wet weather coincided with the decrease in the waves of the epidemic it may be that simple moistening of the air would have some beneficial effect.

Summary

An account is given of a small epidemic of influenza A.

Indications as to the treatment of such an epidemic when occurring on a military camp are given.

The importance of bearing in mind the possibility of influenza when certain symptoms not normally "textbook" arise, as well as the necessity for excluding tuberculosis, is stressed.

Our thanks are due to Dr. C. H. Andrewes and Dr. A. Morton Gill for the very kind advice and help given, as well as to Wing Commander Hudson, Wing Commander Church, Squadron Leader Wynroe, Squadron Leader Campbell, and our own sick-quarters staff for their great assistance.

BIBLIOGRAPHY

- Adamson, J. D., and Fleet, R. O. (1942). *Canad. med. Ass. J.*, **46**, 121.
 Andrewes, C. H. (1942). *Proc. roy. Soc. Med.*, **36**, 1.
 — et al. (1941). *Lancet*, **2**, 387.
 DeCoursey, C., and Thuss, O. (1941). *Ohio State med. J.*, **37**, 964.
 Emdin, W. (1942). *S. Afr. med. J.*, **16**, 101.
 Gill, A. Morton (1938). *British Medical Journal*, **1**, 504.
 Hirst, G. K. (1942). *J. exp. Med.*, **75**, 49.
 Horsfall, F. L., jun. (1939). *Ibid.*, **70**, 209.
 — et al. (1941). *Ibid.*, **73**, 335.
 Lush, D., Stuart-Harris, C. H., and Andrewes, C. H. (1941). *Brit. J. exp. Path.*, **22**, 302.
 Martin, W. P., and Eaton, M. D. (1941). *Proc. Soc. exp. Biol.*, N.Y., **47**, 405.
 Price, F. W. (1941). *Textbook of Medicine*, London.
 Smith, W., Andrewes, C. H., and Laidlaw, P. P. (1933). *Lancet*, **2**, 66.
 Stokes, J., and Henle, W. (1942). *J. Amer. med. Ass.*, **120**, 16.
 Stuart-Harris, C. H. (1941). *J. R.A.M.C.*, **77**, 123.

ESTIMATION OF HAEMOGLOBIN BY PHOTO-ELECTRIC ABSORPTIOMETERS

BY

J. M. PETERSON, D.Sc.

Lecturer in Experimental Physiology, the Physiology Institute, Cardiff
AND

D. H. STRANGEWAYS, M.A., Ph.D.

Assistant Lecturer in Histology and Embryology, University College, Cardiff

Changes in intensity of light and in the electric properties of the photo-cell have been advanced as sources of error in the estimation of haemoglobin by means of photo-electric absorptiometers (M.R.C. Report, 1943). This objection to a method which at the moment appears to offer the best means of accurate estimation of haemoglobin may be justified when it refers to absorptiometers which employ a single photo-cell and which indicate the light-absorbing power of the solution by the extent of deflection of a galvanometer. When, however, a null technique is employed with an apparatus incorporating two opposed photo-cells, error due to these changes has been found, in practice, to be eliminated. The procedure by which such error is avoidable can be simply stated without detailed description of method.

Outline of Procedure

The concentration of haemoglobin is measured in terms of the width to which a slit has to be adjusted to pass through the haemoglobin solution that quantity of light which gives a definite output of current from the photo-cell on which the transmitted light is focused. With the light to one photo-cell passing both through the filter whose light absorption in terms of haemoglobin is known and through the adjustable slit set at the haemoglobin value of the filter, the light to the other cell is varied to give a null reading on the galvanometer. The standard filter is then replaced by the blood solution to be measured and the light adjusted by means of the slit till the null reading is again obtained. On replacing the standard filter and resetting the slit to the filter value the instrument should give the null reading. If there has

been a significant change in either or both of the photo-cells or in the light source the null reading will not be obtained, and the light to the balancing photo-cell will have to be readjusted to give a null reading before repeating the estimation. The estimation is reliable only when the null reading is obtained with the standard filter both before and after making the estimation with the blood solution.

As the standard filter and the blood solution do not necessarily transmit the same wave-lengths, it is essential, for this procedure to give accurate results, either that any change in current output from the photo-cell must be the same for all effective wave-lengths or the range of wave-length employed must be so narrow that such a wave-length difference, if it occurs, is without effect.

Our justification for claiming that changes in light intensity and in the condition of the photo-cells need not cause error is this: With an absorptiometer of the opposed-cell type using barrier-layer selenium photo-cells and NaOH-blood solutions, the reading on the instrument for a given solution is unaltered with wide differences of light intensity; further, the haemoglobin value of a solution determined immediately the photo-cells are illuminated is unchanged when redetermined after the apparatus has been in use for some hours, when the condition of the cells has markedly altered (as shown by the galvanometer deflection produced by standard illumination).

Some Observations

In estimating haemoglobin by photo-electric means we have made a few other observations that may be of interest. Certain advantages of the alkaline haematin method have already been advanced by Clegg and King (1942). In some ways the procedure employed in the present work differs from that of other workers. It has been found that accurate measurements can be made with NaOH-blood solutions without the necessity for either heating the alkaline solutions or forming alkaline haematin indirectly by conversion from acid haematin. 0.02 or 0.04 ml. of human blood added to 5 ml. of decinormal NaOH, estimated within five minutes, gives a value greater than 95% of the maximum. For very accurate work it is an advantage to let the solution stand overnight and to use only the supernatant liquid. There is always a slight deposit, but the supernatant liquid gives a constant maximum absorption for about five days. This rapid rate of denaturation is not necessarily true of all bloods. That of the ox, for example, requires some hours for a satisfactory equilibrium to be established.

A further advantage in the use of a strong alkali lies in the fact that while slight clotting in the pipette, which may be undetected by macroscopic examination, causes a considerable error in the absorptiometric estimation of an aqueous oxyhaemoglobin solution, the clot is readily broken down in decinormal NaOH and the supernatant fluid gives a true reading.

The region of the spectrum best suited for accurate photo-electric estimation of alkaline haematin has been investigated, employing a wide range of colour filters. A spectroscopic examination of two alkaline blood solutions, representing extremes of concentration of haemoglobin, show marked differences in light absorption in three regions of the visible spectrum—viz., (1) in the band about 600 $m\mu$, (2) in the long-wave end about 750 $m\mu$, and (3) in the short-wave end to about 495 $m\mu$ for the weaker solution and to about 525 $m\mu$ for the stronger. Decidedly the best results, as measured by change in photo-cell current with change in haemoglobin concentration, have been obtained in this last region using a Wratten 45A filter, which gives light transmission from 430 $m\mu$ to 540 $m\mu$, with maximum at 470 $m\mu$ and a smaller transmission in the ultra-violet. This absorption region is not so specific for alkaline haematin as is the 600 $m\mu$ band, but the degree of change of absorption with change in concentration of alkaline haematin, as indicated by the photo-cell, is so much greater here than in the 600 $m\mu$ band, as delimited by available colour filters, that its use is justified. In estimating total blood pigment, the lack of specificity of the shorter-wave region could be significant only if one were dealing with abnormal blood containing an appreciable quantity of some pigment not convertible to alkaline haematin by decinormal NaOH. Further, in the wave-length range transmitted by the Wratten 45A colour filter the absorption by blood plasma is insignificant, as shown by the extent to which the absorption by decinormal NaOH is increased by the addition of plasma to it. In a series of experi-

ments the addition of 0.02 ml. of plasma to 5 ml. of decinormal NaOH did not, in any instance, increase the absorption by more than the equivalent of 0.1 g. Hb per 100 ml. of blood, and in some instances there was no increase. It is obvious that the plasma plays no significant part in the estimation.

The accuracy obtained in photo-electric estimation of alkaline solutions of blood is highly satisfactory. Replicates of 0.02 ml. from the same sample of blood with an oxygen capacity of about 20 ml. per 100 ml. of blood can be estimated with a standard deviation of 0.1 and with a standard error of 0.04 for twelve estimations. When 0.04 ml. of blood is used the error is less.

In the absence of an absolute standard of reference we have expressed our measurements in terms of oxygen capacity, and have found that the curve relating absorptiometer reading to relative concentrations of haemoglobin, as determined by exact dilution, coincides satisfactorily with that relating absorptiometer reading to oxygen capacities of the same relative magnitudes and determined by the van Slyke manometric method.

From the observations made it seems justifiable to claim that the reliability of the absorptiometric method of measurement here described is such that, provided the blood does not contain appreciable quantities of haemoglobin derivatives inconvertible to alkaline haematin by decinormal NaOH, the only significant errors involved in estimating the total pigment lie in the sampling (Peterson, Strangeways, and Jordan, 1943) and in the lack of an absolute standard.

Summary

An answer is given to the criticism that changes in light intensity and in the electric properties of the photo-cell result in unavoidable errors in the absorptiometric estimation of haemoglobin. Certain facts relating to the accuracy of the estimation are discussed.

REFERENCES

- Clegg, J. W., and King, E. J. (1942). *British Medical Journal*, 2, 329.
M.R.C. Report (1943). *Ibid.*, 1, 209.
Peterson, J. M., Strangeways, D. H., and Jordan, R. C. (1943). *J. Physiol.*, 102, 5P.

Medical Memoranda

Staphylococcal Pyopneumothorax in an Infant: Recovery

The following case record may be thought interesting enough to merit publication.

CASE HISTORY

A female infant was born of a healthy mother on Dec. 14, 1942, at the West Middlesex County Hospital. The infant weighed 7 lb. 5 oz. at birth and progressed satisfactorily until Dec. 20 on breast-feeds complemented with national dried milk. On that date she developed diarrhoea, with orange stools, and vomited once. She was transferred to the children's isolation ward, where she was found to be slightly dehydrated. She seemed to make a little progress on graduated feeds (glucose-saline, dilute lactic acid milk, and expressed breast milk) until Dec. 31, when she suddenly became much worse, with watery orange stools and severe dehydration. An intravenous drip was necessary to correct the latter, and, owing to the poor oral intake of fluids, was continued until Jan. 6, 1943. By that time she had developed thrombophlebitis of the internal saphenous vein, into which the drip was running. She had a slight temperature for two days, but this subsided when the drip was stopped. She also had two septic fingers and a septic toe. She improved sufficiently to go to the breast, for the mother's milk was coming in better by then. On Jan. 13 she suddenly collapsed at the breast, and was found to be pale, cyanosed, and almost pulseless. Later, when examination was possible, her temperature was 103° F., pulse 140, and respirations 48. The percussion note was impaired at the left base, where bronchial breathing and pleural rub were found on auscultation. A diagnosis of pneumonia was made, and the child was put in an oxygen tent and given a course of sulphadiazine (8 g. in six days).

During the next week the temperature subsided but did not reach normal, and the respiration rate increased. On Jan. 21 slight oedema of the legs was noticed. By the next morning the oedema was so gross that the legs looked like sausages, and the body wall was also dropsical. On examination of the chest the respiratory excursions were found to be minimal, respiration rate 60, heart sounds muffled at the left base and accompanied by harsh crepitations. The apex beat could not be located. A radiograph (portable) revealed gross displacement of the mediastinum, which was bowed out almost into the right axilla. It was thought that this bowing might have kinked the inferior vena cava, thus causing the oedema of the legs.