Discussion

Lewis (1927) presented evidence that when a substance, other than histamine, was injected into human skin and produced a triple response, it did so by virtue of the liberation of the histamine already present in the skin but held in an inactive form. The demonstration that a triple response is inhibited by treatment with one of the antihistamine drugs supports Lewis's conclusions. MacIntosh and Paton's (1949) observations in man that the second of two injections of a histamine-liberating compound into the same site within 24 hours fails to produce a full triple response, coupled with the work of Feldberg and Talesnik (1952) on the slow rate of recovery of skin histamine following its liberation in animals, also supports Lewis's hypothesis. It follows that if any substance (1) produces a triple response on intradermal injection in man, (2) produces a reduced triple response when the subject is treated with an antihistamine drug, and (3) fails to produce the same response when a second injection is made into the same site within 24 hours, it is likely that the substance is a histamine liberator.

The present observations show that pethidine fulfils these criteria, and so support the conclusions of Nasmyth and Stewart (1950). They also provide an explanation for the observations of Zuck (1951) and Pearce (1951), and show that the walls of the veins in the forearm are permeable to pethidine, histamine, and mepyramine. No other reference has been found to the passage of substances through the walls of veins of comparable size apart from the observation of Magendie, who in the early part of the nineteenth century applied a "spirituous extract of nux vomica" to the outside of the jugular vein of a dog and observed signs of poisoning within four minutes (Franklin, 1937).

In man, the present observations show that the appearance of a triple response in the skin after the intravenous injection of pethidine is to be expected if the injection is made into a superficial vein. Although a fall in blood pressure is common after giving pethidine intravenously (Branwood, 1943), sustained hypotension and collapse are rare. The experiments on the cat indicate that, at least in this animal, the hypotensive effect of pethidine is not due solely to the liberation of histamine. It seems likely that in man some factor other than histamine liberation is involved in the production by pethidine of severe hypotension and collapse.

Conclusions

The triple response in the skin after an intravenous injection of pethidine is due to the permeation of pethidine through the vein wall and the liberation of histamine from the skin overlying it. The walls of the superficial veins in the forearm are also permeable to histamine and mepyramine.

It is unlikely that the occasional case of vascular collapse and marked hypotension following an injection of pethidine occurs as the result of the liberation of histamine.

We wish to thank Professor J. L. D'Silva for continued advice and encouragement. Miss P. Archer kindly drew the illustration for Fig. 1.

REFERENCES

- Branwood, A. W. (1943). *Edinb. med. J.*, **50**, 177.
 Butler, E. B. (1951). *British Medical Journal*, **2**, 715.
 Duguid, A. M. E., and Heathcote, R. St. A. (1940). *Quart. J. Pharm.*, **13**, 318.
 Feldberg, W., and Talesnik, J. (1952). *J. Physiol.*, **117**, 3P.
 Franklin, K. J. (1937). *A Monograph on Veins*, p. 116. Baillière, Tindall and Cox, London.
 Giff, J. H. P. (1947). *British Medical Journal*, **1**, 901.
 Halpern, B. N. (1952). *Ibid.*, **1**, 380.
 Lewis, T. (1927). *The Blood Vessels of the Human Skin and their Responses*. Shaw, London.
 MacIntosh, F. C., and Paton, W. D. M. (1949). *J. Physiol.*, **109**, 190.
 Nasmyth, P. A., and Stewart, H. C. (1950). *Ibid.*, **111**, 19P.
 Paton, W. D. M., and Schachter, M. (1951). *Brit. J. Pharmacol.*, **6**, 509.
 Pearce, C. (1951). *Brit. J. Anaesth.*, **23**, 205.
 Zuck, D. (1951). *British Medical Journal*, **1**, 125.

THE "REFLUX" FACTOR IN ERYTHROCYTE SEDIMENTATION TESTS

BY

W. D. ALEXANDER, M.B., Ch.B.

(From the Gardiner Institute of Medicine,
University of Glasgow)

The most popular methods of determining the erythrocyte sedimentation rate are those of Wintrobe and Westergren. The internal measurements of Wintrobe and Westergren sedimentation tubes are similar except that the length of the former is half that of the latter (2.5 mm. diameter; 100 and 200 mm. long). It was therefore surprising to find that the sedimentation rate of a specimen of oxalated blood determined in a Wintrobe tube differed from the determination in a half-filled Westergren tube. It was noticed that the period of aggregation before fall of the cells started was much longer in the Wintrobe than in the Westergren tube. The technique used to fill the tubes would seem to be the cause, since the columns of blood are identical qualitatively and quantitatively.

One way in which the method of filling the tubes differs is that in the Westergren method blood is drawn past the zero mark and then allowed to fall to it, so that a thin film of blood is left on the inside of the tube above the mark; whereas the Wintrobe tube is filled to the zero mark and the pipette withdrawn, leaving the inside of the tube above the mark free of film. In the first few minutes after a Westergren tube is set up there is therefore a reflux of cells and plasma on to the blood level at the zero mark as the blood adherent to the inside of the tube above the mark runs down to the meniscus. This reflux of blood is of course absent in the Wintrobe tube.

Since the presence or absence of this reflux of blood seemed to exert some effect on the sedimentation of oxalated blood, the effect of this factor on the classical Wintrobe and Westergren methods was investigated. The objects of this paper are: (1) to show that reflux of blood on to the meniscus is a factor frequently affecting the half-hour reading in the Wintrobe method of determining the E.S.R.; and (2) to demonstrate, in four cases in which the readings of the two classical methods were in disagreement, that if reflux of blood on to the meniscus was made to occur in the Wintrobe tube the discrepancy disappeared.

Methods

Comparisons between the readings of the standard Wintrobe and Westergren methods were made on blood from 40 unselected patients. By the Westergren method 7 showed a sedimentation rate between 0 and 10 mm., 14 between 10 and 30 mm., and 19 over 30 mm.

In four cases there was disagreement between the one-hour readings of the two methods, a raised Westergren reading being associated with a normal Wintrobe reading. Each of these patients had active organic disease, and a raised E.S.R. was expected. Blood from these four cases, and from a further 24 in which the readings of the methods were in agreement, was examined to determine the effect of reflux of blood on to the meniscus in each of the two techniques.

In the group of 24 patients in whom the standard methods gave similar readings two pairs of tubes were set up: (1) a pair of Wintrobe tubes (containing oxalated blood), and (2) a pair of Westergren tubes (containing citrated blood). In one of each pair reflux of blood on to the meniscus was allowed, in the other prevented.

Reflux of blood normally occurs when a Westergren tube is filled—in this series the blood was drawn 4 to 5 cm. past the mark and allowed to fall to it. In the tubes in which it was desired to prevent reflux the rubber tube from a W.B.C. pipette was attached to the top of the Westergren tube. Blood was sucked slowly up, and when it reached the mark was stopped by pressing the rubber tube on to the top of the Westergren tube.

Reflux does not normally occur when a Wintrobe tube is filled, the pipette being removed when the blood reaches the mark. When it was desired to cause reflux the tube was filled to the top (1 cm. above the mark), sucked back to the mark, and a small drop of blood left at the top of the tube as the pipette was withdrawn. This small drop runs back on to the meniscus.

In the four cases in which a discrepancy was known to exist between the one-hour readings of the classical methods three pairs of tubes were set up: (1) Westergren tubes (containing citrated blood); (2) Wintrobe tubes (containing oxalated blood); and (3) Westergren tubes filled to the 100 mark with oxalated blood. In one of each pair reflux was allowed; in the other it was prevented.

Results

Of the 24 cases in which the classical methods gave comparable one-hour readings, 5 showed an E.S.R. between 0 and 10 mm., 9 between 10 and 30 mm., and 10 over 30 mm.

One-hour Readings.—In each case the one-hour readings in all four tubes were comparable.

Half-hour Readings.—(a) *Oxalated blood*:—When the half-hour readings in the pairs of Wintrobe tubes were considered the cases seemed to fall into two groups—those with a P.C.V. above 37%, and those with a P.C.V. of 34% and below. The former group numbered 19, and in all these the half-hour reading when reflux was present was twice or more the half-hour reading when reflux was absent. There were five cases with a P.C.V. below 35%. In all these the half-hour readings in each pair were very similar. They were 31, 21 mm. (P.C.V. 34%); 1, $\frac{1}{2}$ mm.; 15, 15 mm.; 13, 12 mm.; 36, 36 mm. (b) *Citrated blood*:—The half-hour readings in the Westergren tubes, with or without reflux, were usually very similar; the average difference was 2.5 mm. Nevertheless in five cases the readings varied in the ratio of 2:1 or more. In all these cases the P.C.V. was 46% or more; and it is interesting to note that the P.C.V. of the blood-citrate mixture would have been 37% or more. The half-hour readings in the five cases mentioned were 3, 1 mm.; 3, 1 mm.; 2, 1 mm.; 6, 3 mm.; and 17, 7 mm.

For the four cases in which the classical methods gave dissimilar results the details of the one-hour readings are set out in the Table.

Four Cases in Which the Classical Methods Gave Dissimilar One-hour Readings

Diagnosis	Westergren		Wintrobe		Westergren Half Filled with Oxalated Blood		P.C.V.
	R.A.	R.P.	R.A.	R.P.	R.A.	R.P.	
Lung neoplasm ..	45	39	31	7	27	2	42
Bronchiectasis ..	22	19	22	12	33	2	48
Coronary thrombosis ..	60	58	38	10	37	16	46
Pyelonephritis ..	58	55	44	12	46	18	38

R.A.—Reflux of blood on to meniscus allowed.
R.P.—Reflux of blood on to meniscus prevented.

Discussion

It is clear that if the effect of a physical factor on the sedimentation rate of a blood is to be tested, the test should be made in a pair of tubes identical in every other way.

In each of the 28 cases examined a pair of Wintrobe and a pair of Westergren tubes were set up. In any pair the

blood filling the tubes was identical, and they were set up in the same stand. It seems reasonable to attribute any difference between the readings of a pair to the solitary difference between them—that the one was filled so that reflux of blood on to the meniscus occurred, the other so that it was prevented.

Fig. 1 shows the usual significance of this factor on the Wintrobe method of determining sedimentation rate. The half-hour reading is considerably affected, being greater when reflux is present, whereas the one-hour reading is affected slightly or not at all. The one-hour reading in each tube agrees well with the one-hour Westergren reading.

The position was entirely different in the four cases in which the one-hour readings of the standard methods were at variance. In them (with oxalated blood), when reflux was absent, the period of aggregation before sedimentation proper starts was greatly prolonged. In each case the difference between the Wintrobe tube filled so as to produce reflux of blood and that filled without reflux, marked at half-hour, were also present at one hour. The period of "disappearance of the discrepancy" occurred between 1 and $1\frac{1}{2}$ hours, instead of between the $\frac{1}{2}$ and 1 hour as is usual (Fig. 2).

Variation in sedimentation rate in Wintrobe tubes (each pair filled with identical oxalated blood) according as reflux is present or absent. The continuous line shows rate without reflux, the broken line rate with reflux.

FIG. 1.—Usual pattern, with half-hour discrepancy disappearing at one hour. FIG. 2.—The pattern in four exceptional cases—the half-hour discrepancy persisting at one hour and becoming insignificant only at one and a half hours.

To corroborate that this behaviour was dependent on the method of filling the tube, and not on some inherent property of the Wintrobe tube, a pair of Westergren tubes were at the same time half filled with oxalated blood, one so that reflux of blood on to the meniscus occurred, the other so that it was prevented.

The half-filled Westergren tube and the Wintrobe tube in which reflux had been prevented gave readings of a similar order. Those with reflux present gave similar readings. The latter pair were in good agreement with the classical Westergren one-hour reading, whereas the former pair gave much smaller readings.

In all 28 cases examined reflux of blood on to the meniscus did not materially affect the one-hour Westergren reading. As mentioned above, it did affect the half-hour reading in five cases. It seems from the practical point of view that here the factor can be ignored because the Westergren tube is usually filled so that reflux of blood is inevitable, and because half-hour readings (the only ones affected) are not usually made.

The point of practical importance is that if this series of patients had been examined by someone who habitually fills the Wintrobe tube to the top and then sucks out blood to the zero mark no case would have been found in which the one-hour reading differed materially from that of the Westergren method. Whereas had the cases been examined by someone who (like myself) was accustomed to fill Wintrobe tubes to the zero mark, and then remove the pipette so that reflux of blood does not occur, four cases would have been noted in which the one-hour Wintrobe reading was at variance both with the clinical condition of the patient and with the reading of the Westergren tube.

Why the difference, described above, in the filling of the tubes should be the cause of differences in sedimentation rate in a particular blood is a question of theoretical interest. It may be due to the momentum of the cells, gained by flowing down the inside of the tube to reach the zero mark, carrying them on downwards. Their momentum might act in certain circumstances as an accelerating factor on the sedimentation of rouleaux forming near the blood surface. As other suggestions can be offered, further work is required on this problem. For the present, however, it is desired to emphasize only the practical aspects.

It seems quite clear from recent papers that there is more than one cause for discrepancy between the readings of the Wintrobe and Westergren methods. Goldberg and Conway (1952) found in two cases that the discrepancy was due to the retarding effect of the oxalate. Goldberg, Glynn, and Bywaters (1952), in a preliminary note, state that the cause of discrepancy in their cases seems to have been the effect of the anticoagulant on the blood pH. In both these series the discrepancy has always taken the form of a high Westergren reading with a normal or near normal Wintrobe reading. Davis (1946) has reported cases in which a high Wintrobe but normal Westergren reading has been found.

It seems that, if the Wintrobe tube is filled so that reflux of blood on to the meniscus occurs, at least one factor causing unexpectedly low Wintrobe readings will be eliminated.

Summary

The Wintrobe and Westergren readings were compared in 40 cases and found to be at variance in four, the Westergren reading being more compatible with the clinical state of the patient.

The effect of reflux of blood on to the meniscus was examined in 28 cases, including the four in which discrepancy was noted.

This factor is of no practical importance in the Westergren method.

In the Wintrobe method it affected the one-hour reading in four cases—the cases in which discrepancy was noted. In these when reflux was present the one-hour reading agreed with the Westergren reading; when it was prevented the reading was discrepant.

One of the factors sometimes causing unexpectedly low Wintrobe readings will be eliminated if the Wintrobe tube is filled so that reflux of blood on to the meniscus occurs.

Grateful acknowledgments are due to Professor Sir John McNee for providing facilities in the Gardiner Institute and for guidance in the preparation of this paper; also to Drs. H. Conway, A. Goldberg, and J. Cochran for their helpful advice and criticism.

REFERENCES

- Davis, L. J. (1946). *Practitioner*, 157, 13.
Goldberg, A., and Conway, H. (1952). *British Medical Journal*, 2, 315.
Goldberg, Sylvia, Glynn, L. E., and Bywaters, E. G. L. (1952). *Ibid.*, 1, 202.

A note on dahlia tubers as a source of food is contributed by Mr. S. G. Harrison to the February issue of the *Journal of the Royal Horticultural Society*. The tubers consist partly of inulin, and though not palatable either to man or domestic animals they could be a useful source of laevulose if a cheap process were devised for converting the inulin into that monosaccharide. The present small demand for inulin is being partly met by a firm which is processing three tons of dahlia tubers a year to produce it; the tubers used are normal horticultural surplus. It is thought that breeding experiments might lead to the development of tubers with a higher content of inulin than the present 9% to 13%, for the sugar content of sugar beet has been increased from about 7% sucrose to about 20%.

A SURGICAL METHOD FOR MANAGEMENT OF PATIENTS WITH PORTAL CIRRHOSIS

PRELIMINARY REPORT OF A CASE

BY

CARLO A. MONTAGNANI, M.D.

(From the Surgical Clinic of the University of Florence)

Ligation of the hepatic artery, alone or together with the splenic and left gastric arteries, has been used in recent years as a treatment for portal hypertension and its complications. Success has been claimed in a number of these cases. Ascites and bleeding oesophageal varices appear to be favourably influenced by a reduction in arterial blood supply to the liver, and liver function seems to improve.

The rational basis for this surgical method can be traced back to the studies of the intrahepatic circulation in portal cirrhosis by Kretz (1904), Herrick (1907), McIndoe (1928), and Dock (1947), and to the more recent demonstration by Montagnani and Paci (1952) of a tremendous increase in the arterial circulation in the cirrhotic liver with widespread arterio-portal shunts of varying size.

Berman *et al.* (1950, 1951) and Berman and Hull (1952) have suggested that the method works by reducing arterial blood flow and pressure in the liver parenchyma so that more blood can pass through the portal bed and portal venous pressure is lowered.

Ligation of the terminal part of the hepatic artery carried great hazard, and as a compromise the artery has been tied proximal to the origins of the gastroduodenal and right gastric branches so that a free collateral circulation is possible. Experimental studies in dogs (Montagnani, 1952) have led to the development of a method of gradual occlusion of the hepatic artery or its terminal branches by wrapping around it a "cellophane" band treated with dicetyl phosphate. This led to a fibroblastic reaction of the surrounding tissues, ending in complete obliteration of the hepatic artery. In the meantime a collateral circulation developed towards the liver through the inferior phrenic arteries and minute arterial branches along the portal vein and the common bile duct. No impairment of liver function or damage to the liver parenchyma was observed after such a procedure. No antibiotics were given at any time.

The favourable results of these experiments encouraged clinical application of the procedure in the following case.

Case Report

A man of 58 was transferred from the medical service with Laennec's hepatic cirrhosis and ascites, which had not responded to medical treatment. His father had died of hepatic cirrhosis and he himself had worked for years in a winery, drank 1.5–2 litres of wine each day, ate little, and smoked much. For two months before admission he suffered from dyspepsia, alternating diarrhoea and constipation, asthenia, and loss of weight.

On admission he was found to be wasted and breathless. The sclerae were icteric. The abdomen was greatly distended by fluid and the lower half of the body was oedematous. Distended veins were present on the lower abdomen. There were no abnormalities in the chest. The relevant laboratory findings were: total plasma protein 5.8 g.% (albumin 2.3 g., globulin 3.5 g.); prothrombin 50%. During