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Blood-Volume: A Commentary

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ABSTRACT

Under physiological conditions and on average, the total volume of blood in the circulation is constant. The total blood volume (BV) and the separate volumes of plasma and erythrocytes vary according to climatic conditions, in pregnancy and in the presence of disease. Such changes can have clinical significance although they are rarely revealed in common clinical practice because of a lack of availability of accurate, cheap and simple techniques for the measurement of BV.

Diminution of BV may occur under intensive-care regimes and is life-threatening; acute exposure to a hot climate leads to an expansion of plasma volume and a corresponding fall in the circulating haemoglobin level. The same changes occur in normal pregnancy and are, perhaps, exaggerated in the tropics. Failure of expansion of PV in pregnancy has adverse effects on the foetus.

New investigations in Singapore of BV in health, disease and pregnancy seem to be desirable now that people, including pregnant women, move frequently between a hot climate and a cold environment provided by air-conditioning.

Keywords: blood volume, control, climatic effect, pregnancy, disease

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INTRODUCTION

The number of erythrocytes, and hence the concentration of haemoglobin, in samples of blood is positively correlated with the volume of red cells (RCV) in the entire circulation. The total volume of blood (BV) which consists of RCV together with that of circulating plasma (PV) normally remains within limits because of "volume homeostasis"[1] which depends on a complex of physiological mechanisms[2-5] including production of atrial naturetic factor[6], activity of the renin-angiotensin system[7,8] and secretion of systemic hormones[9,10]. Therefore, diagnosis of anaemia and polycythaemia should readily be made when the packed cell volume (PCV) of a sample of blood is below or above normal. However, in clinical practice when individual values, rather than group mean values, are determined and when pathological processes may disturb volume homeostasis PCV does not always reflect RCV with sufficient sensitivity[5]. This is usually because of an excess or deficit in PV in pathological conditions and in physiological control of the temperature of the body.

Polycythaemia may be wrongly diagnosed when there is a reduction in PV[11] and such a reduction, due, perhaps, to severe diarrhoea, use of diuretics, or even prolonged standing upright[12], conceals the presence, or degree, of anaemia. A false diagnosis of anaemia may be made when there is an expansion of PV, as in acute glomerulonephritis, cirrhosis of the liver, congestive cardiac failure, beri-beri and some other forms of malnutrition, although not always in nephrosis despite the presence of oedema[13].

Haemodilution in normal pregnancy, which may lead to a false diagnosis of anaemia, has been revealed by many investigators[14]. PV begins to expand soon after conception and finally reaches 45% to 55% above that of the volume before pregnancy[15]. RCV also increases, although to a lesser extent, and there is a consequent increase in BV which allows filling of an enlarged circulatory capacity associated with venous and arteriolar dilatation[16], an absolute increase in metabolic rate[17], an increased cardiac output, a marked decrease in peripheral circulatory resistance[18], and probably, therefore, an increase in blood-flow, which would cause secretion from endothelium of the powerful vasodilator, nitric oxide[19]. Failure to achieve an expansion of PV has adverse effects[20] first observed by W. Zangmeister in 1903 and subsequently by others[21]. Relative haemoconcentration causes, and may precede, the onset of hypertension and toxaemia[16] and retardation of foetal growth[22], and there is evidence that effects of hypovolaemia in pregnancy may result in permanent damage to renal haemodynamics[23,24]. Detection of haemoglobin levels in pregnant women above those compatible with haemodilution should direct attention towards the

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possible use of haemodilution therapy[25] to forestall or limit development of abnormalities.

Apart from pregnancy, failure of physiological control of BV to accommodate to the available intravascular space, even to a moderate extent, may eventually lead to a marked rise in systemic blood pressure[3] and a contraction of PV and BV is associated with hypertension in men[26,27].

There is a low degree of correlation between PCV and anaemia in patients with splenomegaly due to any cause, such as malaria, lymphoproliferative disorders, polycythaemia vera. The circulation in the spleen, in common with those in bone-marrow and placenta, forms an arterio-venous shunt in which erythrocytes are held back whilst plasma is "skimmed off" to proceed into, and therefore dilute, the general circulation[28]. The larger the spleen the greater the volume of skimmed plasma and hence the degree of haemodilution. An accurate diagnosis of polycythaemia or anaemia requires measurement of the size of the spleen, perhaps by the use of gamma camera scintography[11].

These examples indicate that variation in PV and RCV can have effects of clinical importance and the measurement of them should sometimes be made.

TECHNICAL CONSIDERATIONS

The standard procedure for the determination of RCV and PV involves the intravenous injection of a small amount of a dye or of radioactive-labelled erythrocytes, albumin or other substances[29]. The subsequent extent of dilution of an accurately known amount of the marker provides a measure of the volume of red cells or plasma with which it has mixed. When PV alone, or RCV alone, is measured, calculation of one or the other requires determination of the PCV. One opinion is that this procedure can only provide a measure of BV[30,31] whether erythrocyte- or plasma-markers are used and that RCV and PV cannot be measured as separate entities and must be assessed by use of the PCV. Because, therefore, PCV is a mathematical function of derived values of RCV and PV it must be closely correlated with them. Different approaches to the measurement of BV, RCV and PV is at present a matter of debate, which suggests the need for better ways of measuring these values and for their standardisation.

Causes of inaccuracy of results of determinations of BV include, leakage, even of small amounts, of the marker from the vein which is injected, variability between individuals, and in the same individual from time to time, in the proportion of BV which flows to different regions of the body[32], and the extent of turbulent flow[33,34] which could influence the time taken for, and the completeness of, mixing of marker and blood. A particular biological source of error is variability in the amount of albumin, and hence of the dye or radioactive marker which is attached to it, that moves into the extravascular space; use of albumin really measures, at least partially, an "albumin pool", and not circulating volume of plasma. Errors also arise in the course of calculations which depend on PCV. The real factor required is the ratio between RCV and PV, the "body haematocrit", which differs from PCV because erythrocytes may be sparse or absent from some parts of the circulation[35] and PCV should be adjusted by multiplying by a factor "f" accordingly. This factor has been reported to vary from 0.76 to 1.15 between individuals[35], although generally it is accepted as 0.91[36]. Some believe that "f" should be 1.0 and that variations reported are due to errors in the measurement of RCV and PV[29]. Clearly, there is doubt about the use of the body haematocrit in calculations of BV and application of a fixed value might engender appreciable errors. PCV itself is affected by variable amounts of plasma trapped in the packed column of cells according to molecular variants of haemoglobin, as in thalassaemia[37], degrees of deformity of cells under shear stress[38] and dimensions of erythrocytes. Errors may also arise by faulty calibration or careless use of micro-pipettes and micro-syringes, and slight, undetected haemolysis in samples of blood or haziness of plasma which interfere with the optical density of dyed plasma from which its concentration is ascertained.

Thus, there are many sources of biological and technical error involved in the measurement of BV and its determination in any individual cannot be assumed to be precisely accurate. The greater the accuracy desired in the determination of RCV the greater the number of manouevres that are made, each of which is a potential source of error[31]. Nineteen separate variables that are involved in the procedure recommended by the International Council for Standardisation in Haematology (lCSH)[39] have been identified[31]. Nevertheless, high degrees of accuracy have been claimed when careful attention is given to these procedures[40]. However, inaccuracies of measurements of BV made in clinical practice and by different observers are unknown; a wide dispersion of individual results shown in all publications suggests that technical and biological errors are appreciable. In contrast, measurement of PCV is practically free of technical error, although biological variation of this value in the same individual from day to day indicates that the mean of several tests made at intervals should be used.

The results of many different inquiries show that average values for RCV/KgBW for normal men is

close to 30 ml and 25 ml in normal women, although individual values within some "normal" groups have been reported as about 20 ml to more than 33 ml.

According to some investigators RCV cannot be predicted from the PCV in individual patients[5]. However, examination of a large number of determinations of PV and RCV led to the conclusion that a single measurement of PCV does provide adequate, and possibly better, information than measurements of RCV or PV for the accurate diagnosis of polycythaemia vera, except rarely when a decline in the rate of erythropoiesis occurs accompanied by an expansion of PV[31]. A positive correlation has been found between PCV and RCV/KgLBM (lean body mass) and when PCV exceeded 60% all those investigated had polycythaemia vera as had 42% of those with a PCV between 50% and 60%[41]. Similar observations were made in another investigation[42], although 18% of patients with a PCV between 50% and 60% had polycythaemia secondary to a low PV so that in this range measurements of PV and RCV should be made.

RCV can be deduced from PCV[31] according to a formula, $RCV/KgBW = 97.2 (PCV)^2 + 20 (PCV)$ when PCV is expressed as a decimal fraction of unity. By use of this formula a diagnosis of polycythaemia can be made with confidence and measurement of RCV is not necessary[31].

STANDARDISATION

Because of considerable diversity in size and shape between individuals and corresponding variation in the capacity of the vascular system there is a need to standardise measurements of BV, RCV and PV. This has commonly been done by expressing volumes in terms of body weight (BW). However, requirements of the body for oxygen and other substances is solely dependent on erythrocytes and plasma so that a primary basis for comparisons of RCV and PV should be related to oxygen consumption[5] or metabolic rate. The measurement of metabolic rate is not usually feasible in clinical practice and can involve appreciable errors. It can be assessed indirectly according to stature and weight, for which various formulas have been proposed[40], although there is appreciable imprecision in their use[43]. When requirements for oxygen are considered, account must be taken of variation in affinity for oxygen between different forms of haemoglobin, as, for example, foetal haemoglobin and those in the haemoglobinopathies[44]. A compromise is to judge adequacy of RCV by relating it to LBM which represents that part of the body mainly responsible for metabolic activity, and RCV is closely correlated with it[45]. LBM can be assessed nearly directly by estimations of natural radioactive body-potassium[46], although facilities for doing so are very rare.

BLOOD VOLUME AND BODY-FAT

Adipose tissue has relatively little metabolic activity and receives a minor portion of the volume of circulating blood so that expression of RCV or PV in terms of fat-free body weight (FFBW) equates with those with LBM. There is a close inverse relationship between RCV and the percentage of the body weight due to fat[41] and measurements of RCV/KgBW can be adjusted accordingly.

The amount of adipose tissue can be calculated from assessments of the volume of body-water, body density, skinfold thickness, or electrobiological impedance[47] as by the Holtain body composition analyser[41]. Skinfold measurements made with care and after practice provide accurate assessments of the amount of subcutaneous fat, which represents, on average, 95% of body-fat.

EFFECTS OF CLIMATE ON BLOOD VOLUME

An expansion of PV, with a consequent fall in PCV, when a person moves from a temperate zone to the tropics has for long been known[48]. The effect is temporary and haematological levels of healthy people who live in the tropics are similar to those who reside elsewhere[49].

Haemodilution due to climatic change and that associated with pregnancy have a common physiological basis and the question arises do they have an additive effect when present together? This does not seem to have been explored, although there is suggestive evidence that expansion of PV in pregnancy in a hot climate may be relatively great[50].

Relationships between haematological indices, BV, and body composition of people who live in the tropics do not seem to have been investigated extensively. Reference to an inquiry in Singapore, although made some time ago[51], may, therefore, be useful in the context of recent observations in Europe and USA.

The investigation in Singapore involved young children, healthy young adults, pregnant women and patients in hospital, some of whom were emaciated and anaemic. Measurements were made of PCV, haemoglobin concentration and PV. The latter was determined by injection of Evan's blue dye and removal of a single sample of blood 10 minutes later, so that the subject was involved for only 20 minutes or so. Those with 10% or less of the body weight as fat had much greater RCV/KgBW than those with more adipose tissue, consistent with observations

made elsewhere[41] and with a suggestion that BV in wasted patients should be judged in relation to "ideal" rather than observed weight[40].

The healthy young men in Singapore had a mean of 23.3% of BW as fat and an RCV/KgBW of 29.3 ml. These values fell on a regression line of a graph based on a number of separate inquiries in temperate zones and made between 1956 and 1962[41]; mean values of the small group of women in Singapore, namely, 33.8% and 20.1/KgBW, fell below this line. Dispersion of individual amounts of BV, RCV and PV in Singapore[51] were the same as those observed in recent times in UK and USA[31,41]. The mean RCV/KgBW of healthy men in Singapore was exactly the same or very close to those of a number of groups investigated in temperate climates[11,52-54]; that of the women in Singapore, 20.1 ml, was relatively low possibly because one or two of them were anaemic. Application of the formula mentioned above[31] to measurements on healthy men in Singapore[51] with a mean PCV of 47.9% showed that, RCV/KgBW = $97.2\ (0.479)^2 + 20\ (0.479) = 31.8$ ml, which is about 8% greater than that actually observed. Possible reasons for this discrepancy might be technical errors, an effect of a hot climate, ethnic differences in body composition, and a relatively low metabolic rate of healthy men which has been revealed in Singapore[55].

Normal men in Singapore had an average BV/KgBW of 69.4 ml, the highest individual value being 90 ml, whereas anaemic patients often had greater amounts, some more than 100 ml. When individual amounts of BV of all subjects were plotted against total amounts of haemoglobin in the circulation values for anaemic subjects all fell above the regression line for normal men. Therefore, deficits of BV due to reductions in RCV had been amply compensated by increases in PV; in some cases these increases may have been excessive.

The results obtained in Singapore showed that, except in some pregnant women, BVs were not obviously affected by a tropical climate and that a diagnosis of anaemia can be made by examination of a sample of circulating blood, although demonstration of degrees of haemodilution in pregnancy and expansion of BV in anaemia required measurements of PV. When needed, a simple method for determining PV apparently yields results similar to those obtained by more sophisticated, elaborate and expensive methods.

PRACTICAL CONSIDERATIONS

Four important observations should be kept in mind when measurements of RCV and PV are interpreted. First, RCV/KgBW is normally correlated inversely with the amounts of body-fat and observed, or calculated, and amounts should be adjusted accordingly. Second, deficits in BV due to a reduced RCV in anaemia are usually made good by an expansion of PV and an expansion of RCV is accompanied by an appropriate reduction in PV so that BV remains constant. Third, in abnormal states and in pregnancy expansion, or in other circumstances shrinkage[56], of PV can be excessive, thus leading to an enlarged or diminished BV and, also, obscuring the presence of anaemia or polycythaemia. An undue expansion of PV may, at least partially, be due to an excessive retention of water by anaemic patients[57], which could be determined by measurement of the volume of urine excreted after ingestion of a known volume, say, 500 ml, of water. Information about BV is important when the use of transfusion of blood is being considered because of a danger in some cases of overloading the capacity of the cardiovascular system and, although nowadays, transfusion therapy tends to be avoided because of possible transmission of serious infections[58] it is sometimes imperative. Hypovolaemia may be present, although unsuspected, in patients undergoing major surgery or who are in intensive-care regimes[29]. In these patients blood can be diverted from some regions and tissues so deprived, for example, the mucosa of the gut, may develop local pathological changes and lead, secondarily, to enhancement of inflammatory and immunological responses, which endanger life[29]. Fourth, a demonstration of the presence of anaemia, substantiated by measurement of RCV, may not tally with clinical signs and symptoms, because of the operation of adaptive mechanisms.

People with severe anaemia sometimes seem able to carry on their usual activities[50]. Under laboratory conditions reduction of haemoglobin levels to around 5g/100 ml are needed before subjective symptoms appear[59] and at this level oxygen requirements can be met. In the tropics anaemic patients are often wasted because of primary or secondary undernutrition. When weight is lost there is an appreciable depletion in LBM as well as of fat, a decrease in metabolic rate[60] and in energy expenditure. These changes reduce the amount of oxygen needed compatible with a reduction in RCV. A decrease in PCV allows an enhanced flow of blood partly associated with a fall in blood-viscosity[61] and, importantly, by an appreciable decrease in peripheral circulatory resistance[53,62]. In addition, because haemoglobin has a strong affinity for nitric oxide[63,64] lack of haemoglobin in anaemia may release more than normally of this vasodilator. Dissociation of oxyhaemoglobin, by which oxygen becomes available to cells, is enhanced by an increased rate of

blood-flow and also by an increased concentration in erythrocytes from anaemic patients of 2, 3-diphosphoglycerate[65]. Diversion of blood from some areas, for example, the splanchnic region, to others of more immediate vital importance has already been mentioned. Thus, through these various mechanisms the human body can adapt to the anaemic state and, thereby, confuse clinical assessments and interpretations of measures of BV.

THE FUTURE

The history of the study of the blood volume extends back over many years. However, aspects of the subject remain controversial and relevant papers continue to be published. As to the future, there is a need to devise new, simple and accurate techniques for the determinations of BV, RCV and PV and for the amounts needed, in various circumstances, according to metabolic indices. A phenomenon that should also, perhaps, be explored is the extent of the haemodilution of pregnancy under different climatic conditions. If, indeed, PV expansion is relatively great in the tropics suitable allowance for this should be made when decisions are needed about haemodilution therapy, and about a haemoglobin level below which iron or other supplements should be distributed in ante-natal clinics.

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