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V.—Principles of Effective and Safe Transfusion.

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1. Effective Blood Transfusion

If the organisers of this Symposium had invited our colleagues in the pharmaceutical industry to outline their concepts of efficiency, it is probable that two quite separate areas of endeavour would have emerged. In the first place, emphasis would have been made of efficiency in terms of technology, use of manpower and the production of profitable and useful by-products. In the second place, as the result of clinical trials, the persuasive Clinical Advisors would define the type of patient likely to benefit from their preparations, the correct dosage and the possibility of untoward effects. It is these broad aspects of effectiveness and safety which I wish to discuss in relation to blood transfusion, set against the fundamental question: what we are doing with our raw material at the present time and what awaits us in the future?

The product given to us in trust by our donors we call whole blood. It can best be described as a complex soup known as plasma, somewhat thinned by a volume of anticoagulant, in which are suspended red cells, essential for the transport of oxygen; white cells, essential to combat foreign invasion; and platelets which play a fundamental role in haemostatic mechanism.

Approximately 80 per cent. of all the donations used in Scotland during 1971 were given in the form of whole blood. Did all the patients require *whole blood*? The answer is no, for many recipients could have received red cell concentrates (Chaplin 1969; Rush and Stewart 1969; Williams 1969; Gollub *et al.* 1971). The implication of this statement must be that several thousand litres of plasma and billions of platelets were given along with red cells to patients who either had no need of them or could have been managed with simpler and safer alternatives. These thousands of litres were thus lost without trace. Why? Is this important? Is Soctland unique in this practice? The reasons for this interesting state of affairs are basically twofold: firstly the Blood Transfusion Service has seen no clear necessity to conserve this plasma and has, therefore, not made available red cell concentrates on the same basis as whole blood. Moreover, partly because most of our Regional Centres are so isolated from the bedside, it has been difficult to encourage effectively the use of cell concentrates. Secondly, and of much less significance, our clinical colleagues have, on occasion, been somewhat reluctant to use this product. The magnitude of this phenomenon, and its root causes, are almost universal and certainly not unique to Scotland, or the rest of the United Kingdom.

To assess the real significance of this loss of plasma it is necessary to consider three subsidiary questions:

1. Are there some patients who require a constituent in whole blood in an amount which far exceeds that found in whole blood?

2. Are there other patients whose requirements are so small, that one donation, suitably processed, is capable of meeting the needs of several patients?
3. Is it possible to process plasma in some way that in so doing a product is made available which has equal clinical efficacy but is now safe from the transmission of disease?

As the answer to all these questions is in the affirmative, and two of the three items (1 and 3) represent significant reductions in the number of donation units equivalent to patient treatment units, it is essential to examine in some detail the existing and projected requirement of these sort of products. Such an examination may have profound effects on the future way we use our raw material, the development of manpower and the introduction of technological innovations both in Regional Centres and Plasma Fractionation Units. As examples, I have selected coagulation factor VIII, platelet concentrates and purified protein solution (PPS). In doing so we shall seek to ascertain whether there is, or is not, an urgent need to consider new sources of plasma which could, at least in part, arise from the present whole blood input.

Coagulation Factor VIII

While there are no doubts as to the potential efficacy and clinical desirability of concentrated preparations of coagulation factor VIII in the management of patients with haemophilia, the ability of the Blood Transfusion Services to meet the quantities required must soon be seriously questioned. The reasons for this are historical, complex, and may differ from region to region, but one of the important problems is the extraordinary lack of data available on the existing requirements for individual regions coupled with a reluctance to compare clinical practices in different regions. Thus, the only published data from the United Kingdom comes from Oxford in which figures are given for the estimated donations required for a 12-month period during 1966/67 (Rizza and Biggs 1969). Some may regard the figures from this world-famous centre to be of little relevance to the ordinary Haemophilia Centre. However, the data compiled from our own issues to haemophiliacs in the South-East of Scotland are in broad agreement. In 1971, over 5000 donations were required to care for the haemophiliac population in the South-East of Scotland. Moreover, the average consumption of just under 100 donations per patient per year in 1971 compared with 25 donations per year in 1961. It is our impression that at least 20 per cent. of the total whole blood input may be required to ensure really satisfactory cover, based on existing practices. However, the widespread introduction of prophylactic regimes (Hirschman *et al.* 1970; Kasper *et al.* 1970) and a more aggressive approach to controlling haemorrhage in patients with acquired factor VIII inhibitors (Robboy *et al.* 1970) may well push the demand to nearer 50 per cent. If we assume that the national average, at this present time, is perhaps just under 10 per cent. we clearly have some way to go.

One of the disquieting trends in the last few years has been the energetic activities of the protein chemists. On the basis of the clinical desirability for a small-volume high factor VIII content product, techniques have been developed which go a long way towards this end (Brinkhous *et al.* 1967; Johnson *et al.* 1971). The serious drawback in this work is the high production losses. The shortage of raw material for the

treatment of all haemophiliacs at the present time is such that until comparable yields are obtained the production of this type of product should be actively discouraged, or at least strictly controlled and its use limited to a small group of patients, such as those with acquired inhibitors.

Platelet Therapy

In the last two years we have been responsible for the preparation of almost 3000 platelet concentrates which were used to cover 175 thrombocytopenic patients. As a result of this work, there is no doubt in our minds that platelet concentrates can be a powerful haemostatic agent and as the expected *in vitro* yield is approximately 70 per cent. (Zucker *et al.* 1969) they represent a good example of efficient blood transfusion.

There has been a precipitous increase in the use of platelet concentrates over the last two years in the South-East of Scotland which was triggered off by the recognition of their importance as an adjunct in the management of patients with acute leukaemia receiving new and powerful cytotoxic drugs. In making a special effort to cover these patients it was discovered that by reorganising certain sections of the department a considerable quantity of platelets could be made available each day, with the minimum of extra effort. One of the pleasing features of this exercise has been the recognition once again of the essential importance of very close co-operation between the medical staff of the Transfusion Centre and the clinicians directly responsible for the patient's care. Thus, although we have now instituted a programme which seeks to provide a constant minimum daily supply, independent of specific requests, the ebb and flow above this base-line is governed by frequent discussions about specific patients.

One of the disturbing features of our results is the high wastage rate. It is difficult to be certain at the present time, but until improved storage techniques have been developed, which are an improvement on the present self-imposed 24 hr., an overall wastage rate of 50 per cent. may have to be accepted. However, it is probable that as more experience is gained this figure can be significantly reduced, even within the present storage restrictions.

One of the gratifying features of this work has been the observations that as the news of the increased availability of platelet concentrates has passed round the region an increasing number of non-leukaemic patients have come to light who require this form of support. In general terms, many of these patients have a much better prognosis than those with leukaemia.

Purified Protein Solution (PPS)

Over the last five years approximately 15 000 bottles of dried plasma have been used per annum in Scotland. Although each unit contains only 500 ml. when reconstituted, the pooling required to avoid transfusion reactions, from natural antibodies, means that every unit contains material from between 10 and 15 donations. Thus, the chance of transmitting hepatitis from this product is significantly higher than the administration of an equivalent volume of whole blood. Moreover, the high free potassium content and low pH of this product may be of some danger in certain clinical situations (Cash 1969). These problems have led to a decision in which it is planned to replace dried plasma by a product which has none of these disadvantages and is notably free from the dangers of transmitting hepatitis (Paine and Janeway 1952). This product is known as Purified Protein Solution (PPS) and is obtained from

the fractionation of plasma. Current fractionation techniques would suggest that approximately one litre of plasma, which would be obtained from 4 to 5 donations of whole blood, will yield $1{\cdot}25 \times 400$ ml units of PPS.

Perhaps largely as a result of the decision to make PPS available in the United Kingdom, efforts have been made to increase the plasma fractionation capacity in this country over the last five years. Thus, extensive building programmes are currently under way in England and Scotland. However, it is possible that less attention has been paid to the quantities of raw material (plasma) required to service these new areas and the ways and means by which this can be achieved.

Most of the dried plasma made available in this country is used in those clinical conditions in which acute circulatory volume expansion is required. One of the features in this therapeutic area has been the concern of some clinicians to avoid the dangers of hepatitis and metabolic disturbances and so use agents other than plasma, in particular, dextrans and Ringer-lactate solution. These alternative volume expanders are not without their own limitations and it is almost certain that given sufficient PPS there would be a substantial shift in clinical practice (Lunsgaard-Hansen 1969; Gutteridge and Shaw 1971). Thus, a more realistic figure for future PPS requirements, and hence plasma requirements, is likely to be obtained by consideration of the *total* (dried plasma, dextrans, Ringer-lactate) volume expansion practice as it exists today.

Over the last 12 months, my colleagues and I have attempted to obtain sufficient data from the whole of Scotland which would enable us to form the basis for such a calculation. Every hospital pharmacy has supplied us with details of dextrans (70 and 110) and Ringer-lactate issues over the last 5–6 years. The results show that since 1965 there has been a 100-fold increase in the use of dextrans and a 30-fold increase in Ringer-lactate. In 1970, approximately 15 000 units of dried plasma, 10 000 units of dextran and 150,000 units of Ringer-lactate were used in Scotland. Limited studies within our own hospital would suggest that only 20 per cent. of the dextrans and Ringer-lactate were used for circulatory volume expansion. Thus, the actual *total* annual requirement has been calculated as 45 000 units of PPS. This estimated figure represents 10 units per 1000 population per annum and compares favourably with the current Swiss usage of 11 units per 1000 population (Kistler 1972). The number of whole blood donations in Scotland, from which all plasma must be removed, to meet this expected need is 180,000 per annum. As our total input is only 200 000 donations per annum, it has been calculated that approximately 100 000 new donations are required if we continue to use whole blood in the way that it is currently being used. Thus, considerable efforts will be required to harvest at least part of this supply of plasma from the existing whole blood input. However, unless the plasma fractionators can obtain substantially better yields it is unlikely that we can avoid a significant increase in the total number of donations required, as has been the experience in Switzerland where the donations per 1000 population is 77 compared to the Scottish figure of 40 per 1000 population (Kistler 1972).

The foregoing analysis has clearly revealed that within the next five years, when a marked improvement in the national capacity to fractionate plasma will be a reality, considerable efforts will be needed to find new sources of plasma. Thus, in the interests of efficiency, in terms of the optimal utilisation of raw material to facilitate effective patient-care, Chaplain's (1969) declaration that routine whole blood transfusions is

a 'thoughtless habit' may have to be considered much more seriously by clinicians and Blood Transfusion Services alike.

2. Safe Blood Transfusion

Although the medical profession has long recognised the concept that there are no therapeutic roses without thorns, there is no doubt that the dangers of blood transfusion, in all its forms, have yet to be fully defined. However, in the ardour of therapeutic endeavour, we are frequently guilty of forgetting those hazards which have already been well documented. Moreover, compared to 20 years ago new types of patients, such as those on chronic renal dialysis and marrow ablation for leukaemia are being exposed repeatedly to the hazards of blood transfusion. The recent introduction of laws which legalise abortion provide yet another group. Thus, Stallworthy *et al.* (1971) recorded an estimated blood loss of 500 ml or more in 16·7 per cent. and his was severe enough to warrant blood transfusion in 9·5 per cent.

Recent data published by the Registrar General (1971) would suggest that the number of deaths attributable to blood transfusion are comparable to those complicating general anaesthesia. Almost 50 per cent. of the post-transfusion deaths were due to hepatitis. While not intending to underemphasise the importance of incompatible red cell, white cell and platelet transfusions, allergic reactions to plasma proteins, systemic effects of bacterial pyrogens and heavily contaminated blood and blood products, air embolism, citrate intoxication and haemosiderosis, the magnitude of the hetatitis problem and the recent explosion of highly productive research in this area is so great that it seems appropriate on this occasion to consider this particular feature of safety in some detail.

In 1965, Blumberg *et al.* (1965) reported that sera from two multi-transfused haemophiliac patients formed precipitin lines in the micro-Ouchterlony gel diffusion test when tested against serum from an Australian aborigine. The substance in this serum did not appear to be the usual lipoprotein and was tentatively labelled 'Australia antigen'. Subsequent studies revealed that the presence of Australia antigen was closely associated with viral hepatitis (Blumberg *et al.* 1967; Blumberg *et al.* 1968; Prince 1968), and that virus-like particles could be isolated from antigen-positive sera (Bayer *et al.* 1968). Confirmation of these findings came from all over the world along with the observation by Okochi and Murakami (1968) which clearly indicated that hepatitis frequently followed the transfusion of antigen-positive blood.

These primary observations heralded an explosive research effort in which clinicians, biochemists, geneticists, microbiologists and immunohaematologists have all made important contributions. Thus, hepatitis associated (Australia) antigen appears to be protein with varying amounts of lipid; whether it contains nucleic acids is disputed (Gerin *et al.* 1971; Jozwial *et al.* 1971). Morphologically, three types of particles have so far been described: two spherical in shape of 200 A and 420 A and elongated tubular forms of up to 2300 A in length. The fact that these particles aggregate together in the presence of Australia antibody suggests a close relationship between them and Australia antigen. Whether the particles are viruses is not yet clear: the two major objections are the apparent absence of nucleic acids and the inability to culture *in vitro*. More recent studies would suggest that the latter objection may be overcome (Jensen *et al.* 1970).

From the early beginnings of this work, debate has gone on as to whether Australia antigen is responsible for serum hepatitis alone or both infectious hepatitis and the serum form of this disease. Recent work has shown that the classical long incubation (serum) form, while more commonly acquired by the parenteral route, can also be transmitted orally. This suggests that Australia antigen is responsible for the classical serum hepatitis and sporadic cases of infectious hepatitis and that other agents are casually related to epidemic infectious hepatitis (Simon 1971). However, there seems little doubt that the agents responsible for the epidemic variety can be transmitted parenterally and, therefore, by means of blood transfusion (Koff and Issebacher 1968).

In 1968, Okochi and Murakami first suggested a possible relationship between hepatitis and the administration of Australia antigen-positive blood. This observation was confirmed by Gocke and Kavey (1969) and both groups have confirmed and extended their original findings (Gocke *et al.* 1970; Okochi *et al.* 1970). This work clearly implies that the routine screening of donor blood for Australia antigen will decrease the total incidence of post-transfusion hepatitis. Although the urgency and importance of this approach cannot be overemphasised, the degree of protection offered remains uncertain, as the final analysis will be dependent upon the prevalence of antigen-positive donors in a community, which can very widely (Prince 1970), the quality of the methods used to detect the antigen and the frequency of other potential hepatotoxic agents in the donor population. Further prospective data is urgently required in different communities to clarify these important questions. At the same time, other areas of blood transfusion practice should receive similar attention with a view to diminishing morbidity from hepatitis. It is, therefore, appropriate to consider the ways in which, within the wide boundaries of blood transfusion practice, attempts can be made to reduce the problem of post-transfusion hepatitis.

Clinician's Contribution

It is sometimes forgotten that a much more conservative approach in the use of blood transfusion by our clinical colleagues could make a significant impact on the incidence of post-transfusion hepatitis. In a critical appraisal of tranfusion practice in the surgical units of a large hospital over an 8-month period, Morton (1969) reported that the administration of a significant proportion of blood was either unnecessary or questionable. Rush and Stewart (1969) have also shown that the introduction of a more liberal use of safe plasma volume expanders in the management of operative and traumatic blood loss, with the transfusions given only when the haematocrit was less than 30 per cent. reduced the amount of blood used by 35 per cent. These findings are of great interest for they underline the observations of Gardiner and Dudley (1963) that with good anaesthesia, accurate dissection and the avoidance of bloody mobilisation of diseased structures then the loss of blood in standard operations should rarely approach half a litre. In most elective procedures this volume will not require blood replacement. Thus, the reduction of the use of blood by no transfusion at all or using safe alternatives could have as great an impact on the incidence of post-transfusion hepatitis as current techniques for Australia antigen testing (vide infra).

Laboratory's Contribution: Australia Antigen Testing

The original method for detecting Australia antigen was the micro-Ouchterlony gel diffusion (ID) technique. The method is simple but lacks sensitivity and requires

several days before a result is available. It is, therefore, a particularly unsatisfactory technique for donor screening, although it may still be of use for the determination of specificity by using known standard reagents.

The counter-current immunoelectro-osmophoresis technique (CIEOP) is a method in which antigen migrates in an electric field through a suitable medium against antibody which migrates in the opposite direction as a result of endosmotic flow. Precipitation occurs at the point antigen and antibody meet (Cullingford 1964). This technique is 10 to 15 times more sensitive than ID in detecting Australia antigen and definitive results are available within 30–75 minutes (Prince 1970).

Using a complement-fixation test (CFT) Shulman and Barker (1969) have claimed a 200 to 300-fold better sensitivity than ID for the detection of Australia antigen. However, the findings of Purcell *et al.* (1969) were nearer 24, and Hierholzer and Le Bouvier (1969) found it to be only 16 times more sensitive than ID for the detection of antigen and barely two times more sensitive for antibody. Bruce White *et al.* (1971) concluded that the sensitivity of CFT and CIEOP are comparable. There are, at present, three major disadvantages of this technique which have important implications to routine blood donor screening. In the first place the CFT requires high titre antisera and this currently is in short supply. Moreover, it is now recognised that false-positive reactions can arise from sera derived from multi-transfused patients. Finally, the very high sensitivities recorded requires an overnight incubation period. Despite these limitations there is no doubt that this technique has two important features which may require serious future consideration by the Blood Transfusion Services; the method can be automated using microvolumes (Sturgeon *et al.* 1971), and the data of Shulman and Parker (1969) would suggest that the detection of an upsurge in anticomplementary activity in the serum may precede the detection of Australia antigen and biochemical evidence of hepatitis by more than 4–6 weeks.

Walsh *et al.* (1970) have recently described a highly sensitive radioimmunoassay which they claim is 200 times more sensitive than CFT, for both antigens and antibody. There is no doubt that this approach represents the most sensitive means of detecting Australia antigen and antibody. However, at the present time, the technique is cumbersome and not readily adapted for routine use. It could, however, be of great value in a retrospective analysis of antigen-negative sera which have been linked, in a prospective study, with hepatitis in the recipient.

Haemagglutination inhibition techniques are commonly used in blood transfusion laboratories and a report of the application of this principle to Australia antigen testing by Vyas and Shulman (1970) was therefore welcomed. The technique appears to be simple, at least as sensitive as the CFT and is readily automated. There is an urgent need for this approach to be developed.

In deciding the most appropriate technique for large-scale screening of blood donations, considerations of simplicity, reliability, sensitivity, availability of reagents, time required to produce a result and cost must all be borne in mind. At the present time, there is no doubt that the method of choice for routine screening is the CIEOP technique. This conclusion concurs with a statement issued by the *ad hoc* Committee on Hepatitis-Associated Antigen Tests of the Committee on Plasma and Plasma Substitutes in the United States (1971).

Although the recent introduction of total donor screening throughout the length and breadth of Scotland must be regarded as a major step forward there is still much

to be done. This concerns the quality of the existing facilities and development of more sensitive screening procedures for future use. While it is accepted that the CIEOP technique is basically simple it is full of pitfalls (Vierucci *et al.* 1970; Khon and Morgan 1971) and liable to give false-positive and negative results. Both these events could have serious consequences on the donor and recipient respectively. False-negative results on plasma destined for fractionation could also endanger the staff of the Plasma Fractionation Units. The quality of reagents, particularly the specificity of antibody, may also be important, as different antigenic determinants on the virus particles seems to be a real possibility (Levene and Blumberg 1969; Millman *et al.* 1969; Le Bouvier and McCallum 1970; Raunio *et al.* 1970). The studies of Das (1972) have emphasised the consequences of this heterospecificity, when it was demonstrated that some sera containing Australia antigen will not react to certain antibodies. Such antibodies should therefore be excluded for blood donor screening purposes.

These potential defects in the existing facilities have serious clinical and legal implications and it would seem to be essential that a national organisation such as the Blood Transfusion Service considers as a matter of urgency the institution of a National Reference Laboratory from where standardised reagents can be made available, all positive reactions checked and technical training and refresher courses organised. It would also play an essential role in the qualitative and quantitative aspects of hyperimmune gamma-globulin production, the clinical use of this material and organisation of the much-needed prospective data on the fate of recipients receiving antigen-negative blood. Such a laboratory should be funded and staffed to ensure that by direct participation and collaboration with other workers throughout the world, the most advanced and appropriate new techniques are introduced to Regional Blood Transfusion Centres as soon as possible. In short, the Blood Transfusion Service must be up in front rather than behind, for we must not assume that the elimination of all antigen-positive units will solve the post-transfusion hepatitis dilemma. Current evidence strongly suggests that the present limitations, which have been calculated to represent a detection rate as low as 25 per cent. cannot be entirely explained on insufficient sensitivity of existing methods, and that other agents are responsible for a significant proportion of the problem (Gocke 1970; Otto-Servais *et al.* 1970).

Miscellaneous Contributions

Although the use of immunoglobulin will be discussed in detail elsewhere in this Symposium there is one aspect of this topic which is of particular relevance in the general context of safe blood transfusion. The Blood Transfusion Service should be concerned and involved, if appropriate, in those measures which seek to reduce the evidence of all forms of transmissible hepatitis in the community at large, as it is from this same community that we obtain our raw material.

Normal immunoglobulin has been shown to be an effective agent in the prophylaxis of infectious (epidemic) hepatitis (Stokes and Neefe 1945; Krugman 1963; Pollock and Reid 1969) and there seems little doubt that those persons in close contact with the patient are at highest risk. Ashley (1954) demonstrated that the secondary attack in families given gamma-globulin was only 0·6 per cent., compared to 15·2 per cent. in untreated families. In 1968 the United States Communicable Disease Centre (1968) recommended the inoculation of home contacts and this recommendation conforms

with the report of the WHO Expert Committee on Hepatitis issued in 1964. The United Kingdom has not adopted this policy and it is difficult to avoid the conclusion that the primary reason for this apparently negative approach is the limitation in the supply of gamma-globulin (Public Health Laboratory Service Report 1968). This problem is largely due to the shortage of plasma fractionation capacity, although in Scotland calculations indicate that sufficient material could be made available at the present time to cover the family contacts of the 4000 cases notified each year.

Other important areas which assist in the progress towards diminishing the incidence of post-transfusion hepatitis are the introduction of purified protein solution and frozen red cells which have been discussed elsewhere.

3. Conclusions

The peculiar role of the Blood Transfusion Service is to provide a link between the healthy section of the community and its sick. In some respects this is a delicate link; for the manner in which a professional body (ourselves) works in harmony with a voluntary community organisation is quite unique. Perhaps as a result of this presumed delicate and sometimes emotional balance between professional and volunteer we have shied away from the harsh realities of efficiency, and have not asked the right questions and in a way which demands a diligent search for constructive answers which require definitive plans for action.

As the future bores in upon us, the increase in complexity and clinical demands on the Blood Transfusion Service becomes disturbingly evident. Thus the answers to some of these fundamental questions are a matter of urgency. Such questions include: What is the role of the Blood Transfusion Service? Should Regional Centres be sited in the urban areas or within major hospital complexes? What is the optimal size of a Blood Transfusion Region? How can we organise ourselves so that systems of research can be developed which seek to isolate, define and quantitate groups of patients requiring any one of the increasing number of products available? Are the current plans for increased plasma fractionation capacity in the United Kingdom really going to meet the needs for the 1980s? In order to insure the high quality of the second and third generation of Regional Directors and their supporting staff, is it not time to take the necessary action, as has been done at an International level, to establish Blood Transfusion as a separate clinical/laboratory subspeciality and in so doing break the increasingly inappropriate all-embracing nature of our attachment to general haematology?

In the answers to these questions lies much that is important for the future effectiveness and safety of blood transfusion practice in this country, for the most important factor of all will be the creation of an environment in which the availability of suitably trained high-quality professional and technical staff will be secured (Greenwalt 1969; Shively *et al.* 1971). The least of our problems will be the availability of our good friend and colleague the blood donor.

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