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DEVELOPMENT OF FACTOR VIII CONCENTRATES

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Development of Factor VIII Concentrates

Development to Date.

During the latter part of 1972, laboratory scale (2 - 10 l) batches of plasma were fractionated by the methods of Newman and Johnson to intermediate potency (approximately 3 U/ml) and high potency (HP) (>20 U/ml) factor VIII. These early experiments tended to confirm the authors' contention that little activity would be lost during the conversion of intermediate to high potency factor VIII.

When, in February 1973, we progressed to the 10 - 60 litre scale, using equipment of the type required for large-scale production, the factor VIII yield of the high purity preparation dropped below 15%. Since by April the P.F.C. had built up more than six months' stock of Fraction I A.F. at the current rate of issue, August 1973 was set as the deadline for solving the yield problem of HP material, or specifying a less ambitious product to replace Fraction I.A.F.

In the course of a vigorous campaign to improve the yield of HP factor VIII, several major variables were discovered in addition to those described by the original authors, and by August it was clear that at least 6 months' more work would be required to raise the yield consistently to the 20 - 25% required for large scale production. In accordance with the original deadline for replacement of A.F, the emphasis of the work was diverted to improvements of the intermediate preparation.

Product.

A product of intermediate type has now been developed to the 100 litre scale and is obtained in 30 - 40% yield. It has the following composition at the point of dispensing:

Factor/

Factor VIII: 4 - 8 U/ml.

Protein concentration: approximately 3 g%, of which approximately 60% is fibrinogen.

Citrate 0.02 M, tris 0.02 M, pH 6.8.

This preparation has been dispensed in 100 ml volumes, and freeze-dried. If dissolved in half the dispensed volume, the preparation as injected into the patient is approximately isotonic, and each bottle contains 400 - 800 units factor VIII in 2 - 4 g protein. Its potency is therefore 8 - 16 U/ml, compared with 20 U/ml for Hemofil; Hemofil is not a high potency preparation, by the defintion adopted by most fractionators. For comparison, the old A.F. product contained about 400 U factor VIII, and was infused at a potency of 2 U/ml. At this concentration, the preparation was slightly hypertonic and contained about 4 g protein.

Further reduction of the citrate content of the new product may improve the potency of the preparation at the point of use to approximately 20 U/ml, but some clinical experience with the existing product would be expected before this was undertaken.

The presented dose (up to 800 U) may be rather high for some applications. It should be noted that patients may receive only about 250 U from a 6-unit cryoprecipitate dose. If there is a strong demand for a smaller dose, it is likely that 30 ml at 6 - 8 U/ml could be dispensed into a vial for re-solution in 10 - 15 ml. The total dose would therefore be in the region of 200 U, conveniently about half the old A.F. dose.

Production Levels.

Large-scale crushing and thawing equipment was commissioned in early September, 1973, and is functioning adequately on/

on a load of 100 l plasma. It is expected that with minor improvments the batch size may be increased to 180 l.

In the present accommodation, and with other current developments and production commitments, no more than one batch per week can be expected. The maximum production will therefore be about 150 doses/month, rising to about 250 doses/month if scale-up is successful. This assumes that thawed plasma activity will remain above 0.6 U/ml, and that a dose will contain 480-800 U.

A minimum production rate cannot be estimated honestly. The first fraction recovered commonly inherits most of the sins of the parent plasma, and the product is extremely vulnerable to contamination during the use of pilot equipment in an environment inimical to rapid and hygienic operations. The earliest results indicate that the problem of contamination is being successfully contained.

Future High Potency Concentrates.

The incidence of haemophilic emergencies which can be treated successfully only with high potency (>20 U/ml) concentrates has not yet been defined. The availability of a new concentrate at a potency approaching 20 U/ml may assist such a definition. It is not disputed that all haemophiliacs would prefer low volume injections, but a 50% penalty in yield, i.e. a 100% increase in plasma input and fractionation cost, cannot be ignored in any rational accounting system.

Development of the next generation of factor VIII concentrates is expected to be slow in the next 12 months, and it would be prudent to emphasise the production of large stocks of intermediate material before committing large resources to the next phase. Possible candidated for investigation are:

(a) Newman and Johnson's HP: potential potency 100 U/ml.

potential/

potential yield 25%. Work interrupted in August may be resumed at a reduced rate. Experience gained in large-scale production of intermediate material will be helpful, but not crucial, to a solution of our difficulties with this method.

(b) Kabi method using immobilised dextran sulphate. potency: 30 U/ml immediately, >100 U/ml with re-work. Yield probably about 50% at 30 U/ml potency.

(c) Methods using specific polyelectrolytes: potency >100 U/ml, yield unknown.