CVD

NOTES OF MEETING WITH IMMUNO AT LONDON AIRPORT - 24 JANUARY

Topic: Hepatitis-Reduced Factor VIII and Factor IX Concentrates for Haemophilia Therapy

Dr Eibl (Immuno, Vienna) opened the meeting by informing us that two methods of chemical inactivation of viruses have been investigated by Immuno. The first method involves exposing viruses to chemicals "of the food industry" and incubating at 4°C. The second method involves exposing viruses to a newly synthesised chemical and incubating at 37°C. These chemicals are supposed to degrade spontaneously to harmless metabolites. The viruses inactivated include non-A and non-B, polio and canine hepatitis. Apparently this last is an adenovirus, which is easily killed.

$C_2H_5O-C=O$
C_2H_5O
DIETHYL - PYROCARBONATE (SIGMA)
H_2O
2 C_2H_5OH
+ CO_2

When batches of factor VIII or factor IX are treated with these methods, the additives and their metabolites are then removed (but presumably not completely).

One batch of Kryobulin has been put through method 1 and 10 mls (250 units of factor VIII) with 100 chimpanzee infectious particles of non-A, non-B (assay method unspecified) have been injected intravenously to 4 chimpanzees. Liver function tests and liver biopsies over the subsequent six months showed no evidence of hepatitis. A second 10 mls of the original material untreated was then given intravenously and this was followed by the standard rise in transaminases and positive liver biopsies characteristic of non-A, non-B hepatitis.

Method 2 is thought to be more powerfully viricidal but has not yet been tried on a factor VIII or factor IX concentrate. It is thought to be promising for the prothrombin complexes but not factor VIII (presumably because of the temperature - 37°C). Afterthought - presumably work is going ahead on prothrombin complexes according to the time schedule mentioned later.

There was some discussion about the nature of the treated Kryobulin after Method 1 and the following comments were made:

Yield

The method destroys about 25 per cent of factor VIII - ie from the raw plasma through to the sterilised product the yield is 15 - 17 per cent. There is no actual removal of fibrinogen.

By Immuno electrophoresis and general chromotography, no "neoantigenic" new plasma proteins have been found.

There is apparently no significant changes in the level of factor VIII RAg, CAg and ristocetin cofactors. The level of C1q activity remained unchanged. There is no evidence of complex IgG formation. The final content of factor VIII is about 1 unit per mg of protein; however recovery and half-life studies in humans have not yet been determined.

Work on the prothrombin complex must have gone ahead because the ratio of the activities of II, VII, IX and X were reported as being unchanged. Presumably they are not yet ready to report on chimpanzee studies of proplex modified by method 2.

There was some considerable discussion about the chimpanzee testing. As far as I can gather, only one lot of Kryobulin has actually been put through chimpanzee studies as outlined above. Two other lots have been tested, but from what I gather this may well only be from the factor VIII and protein analysis point of view. There is obviously considerable difficulty in getting access to a large number of chimpanzees. It is apparent that the hepatitis-B virus has not been put through this particular process.

Ari Zuckermann commented that in his view it was important to keep the hepatitis-B and non-A, non-B material separate during the investigation of any treatment efficacy because of an apparent mutual neutralising effect. I personally pressed the idea of working with "real-life" ie large pools in which all sorts of viruses may be mixed, but apparently the academic answer is that if you can have a method that guarantees sterility of all the viruses singly, this method should be applicable to large pool materials with mixtures of viruses. (AZ spoke with authority!) — as always.

He also went on to comment that were other viruses to be used, this may obviate the need for testing in chimpanzees and testing for hepatitis. For example, polio viruses and canine hepatitis viruses could be tested on tissue cultures. In this context the chimpanzee tests would only provide a "gross" safety net.

Clinical Trials Design

There was some considerable discussion over the nature of the design. Dr Craske in preliminary comments pointed out that in work with Dr Rizza, rather to their surprise, over eighteen months 30 patients with no previous history of exposure or only slight exposure had been recruited to a study of the incidence of non-A, non-B after factor VIII concentrate. These patients included patients with von Willebrand's disease and mild haemophilia, and incidentally indicated that completely unexposed patients had 100% incidence of hepatitis with BPL factor VIII and that patients who had previously received 4-5 batches of material had a 50 per cent chance of developing non-A, non-B hepatitis.

For a new trial of hepatitis-free factor VIII material, Dr Craske suggested that ideally 4-5 separate batches should be prepared and two patients allocated to each batch. The follow-up period after initial exposure should be six months.

Dr Hill and Professor Hardisty pointed out the ethical difficulties of using newly diagnosed children as first candidates in the trial. This is because children may be safer on cryoprecipitate because of the possible toxic effects of the added chemicals, and also because of the need for considerable

follow-up venepunctures which in the context of general ethical difficulties with working on children as experiments, must be a severely limiting factor.

After some discussion it was agreed that trials should initially take place in adults, and particularly those who require large doses of factor VIII frequently. Such patients could be used for assessing data on recovery, half-life and toxicity. When such patients reveal satisfactory data, the next stage is going to adults who have only occasional requirements. (However, these adults would have to have a good reason for not receiving the cryoprecipitate or DDAVP - such as major surgery). After an initial period of satisfaction in such patients, one could then go to children.

Immuno commented that both factor VIII and factor IX concentrates (!) could be available from April 1983 as it is anticipated that the patenting problems will be solved by then. The meeting felt that the overall time scale might be two years.

Acquired Immunodeficiency Syndrome (AIDS)

This was discussed in the after lunch period. Dr Kraske summarised the current position. He gave a clinical description of the AIDS syndrome which included demyelinisation and also cases of acquired thrombocytopenia. Many of the infections which such patients sustain are characteristic of the immunocompromised host.

The basic lesion appears to be intractable and to be caused through impaired cell-mediated immunity. The population groups affected include promiscuous homosexuals, heroin addicts, immigrants into the US from Haiti (these are not necessarily homosexuals or addicts, and one such individual may well have acquired his disease outside the US). Incidents of clustering have been reported but an early report involving amile nitrite appears to have been disproved.

Other clinical features and lymphoreticular tumours, Kaposi sarcoma (of the acute type similar to that seen in Africans) repeated attack by various of the herpes viruses and also parasites and fungi.

Up to 10 December 1982, some 800 people had been reported as suffering from the AIDS, and there was a 45% mortality.

Ten haemophiliacs in the US have been affected and five have died. The youngest was aged 7. All cases have had prolonged treatment with factor VIII, but there is no specific implication of one particular product or batch. Other cases involving blood and blood product transmission have included platelets transfused in three cases. In one of these cases, one of the donors was a young New York man in his twenties. A second case was a 20 month old child with Rhesus HDN who had received several units, including platelets known to have come from a homosexual donor who was asymptomatic at the time, but who later died. The child has developed autoimmune haemolytic anaemia and a possible AIDS state.

The incubation period for the syndrome appears to be six months to two years.

In the UK, so far only one or two cases have been reported from the communicable diseases centre.

The infectious precautions include discouraging homosexuals from donating blood or organs. Protocols from the United States are being considered by the Hepatitis Working Party in the UK. Apparently the American fractionation companies are very aware of the problem and are taking some unspecified measures to screen out such donors.

The attention of the meeting was then drawn to the two articles on the editorial in the New England Journal of Medicine of 13 January, which in summary indicates that the T4/T8 ratios among haemophilics receiving factor VIII is greater among those who have been exposed to concentrates than those exposed to cryoprecipitate only. However, cryoprecipitate in the US comes from volunteer unpaid donors and therefore are presumably well motivated people.

Final comments on the possible nature of the transmissible agents indicated that there may not be just one agent, but a mixture, ie - a barage of viruses including hepatitis B, non-A, non-B, CMV, and many others, possibly transmitted from asymptomatic healthy blood donors.

Those present included:

Dr Evans (Manchester)
Dr Barrowcliffe
Dr Rizza
Dr Hamilton
Dr Ludlam
Dr Colvin
Professor Hardisty
Dr Preston
Dr Mayne
Dr J Davidson
Dr Aronstam
Dr Hill
Dr Edgecombe
Dr Prentice,
Dr Savage
Dr Kernoff
Dr J Leslie
Dr Winfield
Dr Wensley
Dr Mibashan
Dr Craske
Professor Zuckermann
Dr Bloom
Dr Shinton

The Immuno team were led by Dr Eibl.

E E Boulton