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NO EVIDENCE OF HEPATITIS OR HIV TRANSMISSION IN VIRGIN HAEMOPHILIC BOYS TREATED WITH BRITISH HEAT TREATED FACTOR VIII CONCENTRATE (8Y). K.J. Pasi and F.G.H. Hill. Haemophilic Unit, The Children's Hospital, Birmingham, U.K.

HIV and hepatitis virus transmission is of major concern with factor VIII therapy. Non-A, non-B hepatitis (NANBH) has a near 100% incidence in patients previously treated with unheated large pool factor VIII concentrates. NHS heated high purity factor VIII concentrate (8Y) undergoes severe protracted heat treatment of the freeze dried concentrate theoretically sufficient to inactivate hepatitis viruses as well as HIV. Infusions of 22 different batches of 8Y have been given to 18 children with haemophilia A (10 virgin patients; 8 who had only received single donor cryoprecipitate) have been treated for up to 18 months. Regular testing for viral antibody seroconversion and biochemical liver enzymes have been made. None had had clinical or biochemical evidence of liver disease prior to the commencement of 8Y therapy. All these boys were immunized with HBVax at the time of the first treatment with 8Y and were HIV antibody negative. Liver function tests were to be performed monthly but due to patient non-compliance this was only achieved in 60% of patients.

All patients receiving 8Y have remained anti-HIV seronegative. Only the virgin patients can be considered suitable for evaluation with regard to the transmission of NANBH. These boys by this time have received multiple batches of 8Y (mean 5 batches, range 1 to 14). In only 1 patient has an isolated rise in aspartate transaminase (AST) been noted (AST 27 to 131 IU/l) 6 weeks after treatment with a new batch, but no rise in alanine transaminase (ALT) or clinical evidence of liver disease was found. Viral serology was performed. AST returned to normal within 12 days. This batch was received by 3 of the virgin and 2 of the previously cryoprecipitate treated boys. All these 5 boys who were exposed to the suspect batch had normal liver enzyme levels when measured within 4-6 weeks of exposure.

Of the 10 virgin patients receiving multiple batches of 8Y a transient rise in AST but with no rise in ALT has only been noted in 1 patient. In the absence of firm biochemical evidence of liver disease NANBH is an unlikely cause. Lack of transaminase rises in other virgin patients strengthens this assumption. We conclude that 8Y reduces the incidence of NANBH and HIV transmission.

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NO ANTI-HIV SEROCONVERSION AFTER REPLACEMENT THERAPY WITH PASTEURIZED F VIII CONCENTRATE. A STUDY OF 151 PATIENTS WITH HEMOPHILIA A OR VON WILLEBRAND'S DISEASE. Kl. Schimpf*, H.H. Brackmann, W. Kreuz, B. Kraus, F. Haschke, W. Schramm et al. Rehabilitation Hospital and Hemophilia Center Heidelberg, Rehabilitation Foundation, 6900 Heidelberg, Federal Republic of Germany*.

Transmission of hepatitis viruses and HIV has proven to be a risk of replacement therapy in hemophilia. As regards F VIII products a concentrate (Hemate HS or P) in which viruses are inactivated by heat-treatment over 10 hours at 60° C in aqueous solution is available since 1979. Our clinical studies have shown that this product does not transmit HBV and HNANBV. As the product was manufactured by 80% from US plasma it was necessary to prove that it also does not transmit HIV. As it is, for ethical reasons, not possible to treat a control group with non-virus-inactivated F VIII, non-transmission of HIV can only proven if anti-HIV seroconversion does not occur in larger groups of patients treated exclusively with this virus-inactivated product. We collected data from 151 patients treated with Hemate HS (P) who had never before received blood or blood products. Therapy was started between Feb. 1979 and Jan. 1986 (median July 7, 1983). The median length of observation till the last anti-HIV testing was 24 (3 - 83) months. 112 patients were observed longer than 13 months. The median total dosage was 17,000 (500 - 2,155,375) IU of F VIII, the median patient age was 6 (0,5 - 68) years. In none of these patients anti-HIV seroconversion (ELISA test) was observed. According to the rule of three, the upper 95% confidence limit for a random sample of 60 cases with zero events would be 3/60 or 5%. For greater numbers of n cases, as in our study, the range of confidence narrows increasingly. The period of observation of this study is hitherto the longest.

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CHANGES IN PLATELET SURVIVAL AND SIALIC ACID CONCENTRATION IN PLASMODIUM BERGEI INFECTED RATS. E.M. Essien and A.L. Inyang. Departments of Haematology & Pharmacology & Therapeutics, University of Ibadan, Nigeria.

Reduced circulating platelet count sometimes to thrombocytopenic levels in man and normally severe thrombocytopenia in animals are well known features of acute Plasmodium falciparum or experimental P. bergei infections in these respective organisms. Suggested mechanism(s), disseminated intravascular coagulation or immune mediated mechanism, thought to be involved in these observations are disputed. Shortened platelet survival has been reported in man.

We now present data on platelet survival and total platelet sialic acid concentration in P. bergei-infected Wistar rats. A total of 52 rats were used. For the platelet survival studies each of the 8 suckling test animals was infected by intraperitoneal route with mouse-passaged P. bergei 4-5 days before injection of ^{51}Cr-labelled homologous rat platelets (50 µCi $Na^{51}CrO_4$/rat) the platelets being obtained from adult Wistar rats. Blood samples were then collected 2 hr after the injection (zero hr sample) and subsequently at 17.0, 42.5 and 66 hrs. Platelet recovery and survival curves were determined on these samples. It was found dat fewer platelets (as % recovery) were obtained from each infected rat sample compared with control, the difference was significant in the 42.5 and 66 hr samples: 7.9 ± 8.1 (test) vs 41.4 ± 15.2% (C) for 42.5 hr and 2.8 ± 4.1 (t) vs 26.8 ± 6.2% (C) for the 66 hr samples ($p < 0.005$ for each). For sialic acid determinations, 40 suckling Wistar rats (30 test, 10 control) were treated as for survival studies. At identical periods, blood was collected, washed platelets obtained, lysed and protein and total sialic acid determined by Lowry (1951) and Aminoff (1961) methods respectively. Total sialic acid of 7.02 ± 4.21 nM/mg protein at 42.5 hrs and 4.8 ± 2.14 at 66 hrs were significantly less than control value of 11.43 nm/mg protein and also showed a negative correlation ($r = -0.95$) with % parasitaemia.

It is concluded that P. bergei infection causes a reduction in total platelet sialic acid with resultant significant shortening of the platelet life span.

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PSEUDO-THROMBOCYTOPENIA ASSOCIATED WITH AN ANTICOAGULANT INDEPENDENT IgM PLATELET AGGLUTININ IN A PATIENT WITH PROLONGED BLEEDING TIME. M.H. Horellou (1), C. Capelle (1), T. Lecompte (1), C. Kaplan (2), C. Lecrubier (1), J.M. James (3), R. Le Menn (4), J.Y. Muller (2), M. Samama (1). Laboratoire Hématologie, Hôtel-Dieu, Paris (1), C.N.T.S. Paris (2), Hôpital Saint-Joseph, Paris (3), and Centre Microscopie électronique, Talence, France (4).

An original observation of platelet agglutination was found in a 63 y.o. man during 5 years following, without spontaneous bleeding. Low platelet counts (10 x 10^9/liter) and large agglutinates were observed on blood specimens collected into EDTA (1 to 15 mg/ml whole blood WB), citrate (1/10 volume citrate 0.11 M), sodium heparin (100 units/ml WB), citrate plus acetyl salicylic acid (5 to 100 mg/ml WB), citrate plus prostacyclin (Upjohn 10^{-5} to 10^{-6} M), and buffering anticoagulant (0.8 M citrate, pH 4.65). Low platelet count and large agglutinates were also observed in capillary blood obtained by finger puncture kept at 22° or 37°C. All techniques revealed significant clumping making assessment of overall platelet number impossible. Bleeding time Ivy horizontal incision is longer than 20 minutes.

Patient's serum induced normal platelets agglutination up to a 1/512 dilution at 22° and 37°C temperature. Lack of agglutination after treatment of the serum by IgM and indirect immunofluorescence tests identified the IgM nature of the agglutinating factor. This antibody reacted with Glanzmann thrombasthenia platelets ruling out the IIb/IIIa complex as the target of this agglutinin.

There was no evidence of platelet activation following agglutination of autologous or allogenic platelets : electronic microscopy of native blood platelets clumps did not show any sign of activation, and plasma level of B-thromboglobulin was normal in this patient. Normal platelets agglutination by patient's serum was not accompanied with ATP secretion (luciferin-luciferase).

This IgM agglutinin was associated with elevation of IgM (700 mg/dl) without clinical or biological signs of auto-immune disease.

No similar case with irreversible platelet agglutination in both capillary or venous blood has been reported before. The significance of the observation remains obscure.