

TYPE OF AGITATION AND PLATELET VIABILITY AND FUNCTION AFTER 22°C STORAGE: CORRELATION WITH MAINTENANCE OF DISCOID SHAPE. S. Holme, K. Vaidja and S. Murphy. Cardeza Foundation, Thomas Jefferson University, Philadelphia, Pa., U.S.A.

Platelet viability as measured by in vivo ^{51}Cr recovery, platelet morphology, and in vitro aggregation with ADP and thrombin were studied with platelet concentrates (PC) stored for transfusion under carefully controlled conditions. The PC were prepared from whole blood with citrate-dextrose-phosphate as anticoagulant. The platelet count was kept between $0.8 - 1.6 \times 10^6$ platelets per mm^3 in a volume of 50 ml. The PC were stored in containers constructed of polyethylene (PE) or polyvinylchloride (PVC) at 22°C for 72 hours. The bags were placed on a horizontal shaker or a ferris wheel type of apparatus during storage. No significant changes in pH or platelet count were observed during storage. PC stored on the wheel showed a moderate loss of viability and marked loss of aggregation response compared to PC on the shaker. Under optimal conditions with PC in PE on the shaker maximal rate of aggregation was reduced only 26% compared to fresh PC. PC stored in PVC showed a statistically significant greater decrease. A good correlation was observed between the percentage of discoid platelets present in the PC as judged by phase microscopy and the extent of platelet shape change response to ADP. Both parameters correlated positively with in vivo ^{51}Cr recovery. We conclude: 1) The type of agitation used significantly effects maintenance of platelet viability and function during storage. 2) Under the conditions used, in vitro function was better preserved in PE than in PVC bags. 3) In vitro function is not inevitably lost during storage at 22°C, but is critically dependent on storage conditions. 4) In vivo platelet viability (^{51}Cr recovery) correlates with maintenance of disc shape. The latter can be quantitated by the shape change response with ADP.

THE IN VIVO EVALUATION OF THE HEMOSTATIC FUNCTION OF STORED HUMAN PLATELETS. M. Blajchman, A. Seny and J. Hirsh. Blood Products Laboratory, McMaster University Medical Centre and Canadian Red Cross Blood Transfusion Service, Hamilton, Ontario, Canada.

The assessment of the hemostatic function of stored human platelets is difficult to assess in human subjects. The use of thrombocytopenic rabbits treated with ethyl palmitate to produce reticuloendothelial blockade, has made it possible to study the hemostatic function of human platelets in vivo. The assessment of hemostatic function has been made using both a jugular bleeding time technique and an ear bleeding time technique, and in both, a close correlation between bleeding time and platelet count has been established. Using both methods, both fresh and human platelets stored for 72 hours at 22°C correct the bleeding time of thrombocytopenic animals to levels appropriate to the platelet count achieved. Platelets stored at 4°C using standard methods of preparation and storage were ineffective hemostatically after 24 hours storage. Platelets prepared and stored at 4°C at a pH of 6.4 were hemostatically effective in thrombocytopenic rabbits for as long as 10 days of storage. No correlation, however, was noted between the hemostatic effect of stored platelets and in vitro tests of platelet function. Similarly, the intravenous infusion of ADP and collagen produced similar falls in platelet count for both hemostatically effective and non-effective platelets. These studies provide further evidence for the limitations of in vitro tests of platelet function for the assessment of the potential in vivo function of stored human platelets. Furthermore, these findings raise the possibility for the prolonged liquid storage of human platelets at conditions which minimize bacterial contamination, yet maintain hemostatic efficacy.

A 3-D VIEW OF SPLENIC PATHOPHYSIOLOGY IN IDIOPATHIC THROMBOCYTOPENIC PURPURA. Marion I. Barnhart and Jeanne M. Lusher. Depts. of Physiology and Pediatrics, Wayne State University School of Medicine, Detroit, Michigan, U.S.A.

The spleen plays a dual role in the pathogenesis of chronic idiopathic thrombocytopenic purpura (I.T.P.) It is generally the major site of phagocytosis of antibody-coated platelets and is also an important site of production of autoantibody against platelets. As splenectomy is the treatment of choice in chronic I.T.P. in children, as well as adults, we have had the opportunity to study the freshly excised spleens from children with chronic I.T.P. We have previously described splenic ultrastructure in I.T.P. as viewed by SEM in two publications (*Excerpta Medica* #357:462-471, 1974 and *Am.J.Hematol.* 1:243-264, 1976). Our most recent studies, to be presented here, employ an additional tool - that of latex particle probes. We have used these probes to identify platelets and macrophages containing phagocytized platelets, and to locate the sites of platelet autoantibody production. 15 spleens were immediately perfused (via the splenic artery) with heparinized Tyrode's solution under physiologic pressure to partially remove blood cells from major vessels. This was followed by perfusion with 0.025% buffered glutaraldehyde and then 0.2 μm latex spheres coated with either specific antiplatelet antibody or normal rabbit immunoglobulin. In I.T.P. spleens perfused with latex spheres coated with normal rabbit immunoglobulin, none of the spheres adhered to splenic cells. In those spleens perfused with latex spheres coated with antibody against platelets, the spheres adhered to splenic macrophages and to morphologically obvious platelets (thus identifying platelet loci in the spleen). In addition, latex coated with platelet membrane protein adhered to lymphocytes in lymphatic nodules, indicating sites of autoantibody production.