

THERAPEUTIC IMPLICATIONS OF HEPARIN METABOLISM IN NEWBORNS. M.M. McDonald, W.E. Hathaway and E.B. Reeve. Department of Pediatrics, University of Colorado School of Medicine, Denver, Colorado, United States.

While thrombotic complications are relatively frequent in the sick newborn, therapeutic heparinization is often difficult to achieve. The clearance of one bolus dose of heparin has been found to be more rapid in the newborn ($T_{1/2}$ of 35.5 min. in babies 33-36 weeks gestation vs. 63.3 min. in the normal adult). Decreased levels of antithrombin III (AT-III) in the newborn (20-50% of normal adult) may explain the relatively short $T_{1/2}$; however, an adult with congenital AT-III deficiency (50% level) was found to have a prolonged plasma heparin $T_{1/2}$ of 106 min. The function of fetal AT-III was studied by isolating the protein from cord blood of nine term and preterm babies by heparin affinity chromatography. Evaluation by SDS-PAGE, immunoelectrophoresis, heparin cofactor specific activity, progressive neutralization of thrombin and Xa at 37°C, pH related antithrombin kinetics and thrombin neutralization relative to heparin concentration revealed a fetal protein with structure and function indistinguishable from that isolated from normal adults.

Clinical use of heparin included the evaluation of the Laidlaw (LL) micro whole blood clotting time. The mean LL time of 79 newborns was 96.2 sec. (SD=23). The LL time was not related to gestational age (24-42 weeks) nor to size (600-3980 g). Nine babies were heparinized for three days to six weeks for major vessel thromboses. Heparin infusion rates were compared with heparin levels achieved, LL clotting time prolongations and AT-III levels. The newborns required heparin infusion rates of 25 to 40 U/kg/hour to achieve plasma heparin levels of 0.3-0.5 U/ml. The prolongation of the LL correlated with the heparin level achieved. In these nine babies, AT-III levels did not decrease during the course of heparin therapy.

These studies suggest that the newborn has an increased heparin requirement for anticoagulation not completely explained by a low level of a normal antithrombin III.

HEPARIN AND ANTITHROMBIN III LEVELS DURING CARDIOPULMONARY BYPASS SURGERY--A PILOT STUDY. P F O'Brien, G F Savidge, B Williams. Depts of Haematology and Cardiothoracic Surgery St Thomas' Hospital Medical School, London, UK.

Heparin and antithrombin III (AT III) levels were followed in ten patients during cardiopulmonary bypass surgery in a pilot study designed to assess the adequacy of the local conventional protocol for heparinization and reversal. Heparin was administered according to the local standard procedure on the basis of body surface area and was neutralized by doses of protamine sulphate calculated on the basis of the initial loading dose of heparin. A fluorometric method was employed to assay plasma heparin and AT III concentrations using the synthetic fluorescent tripeptide, H-D-phe-pro-arg-5-amidophthalic acid, dimethyl ester, as the thrombin substrate. Heparin levels were in the range of 2.0 to 6.5 IU/ml during bypass with significant differences between and within patients in response to periodic additional injections of heparin, due to neutralization of heparin by PF4 released during bypass. All patients showed a sudden drop in the level of AT III of 15 - 35 IU/dl after the initial dose of heparin given before the start of surgery. The AT III levels varied greatly during bypass both between and within patients, falling to as low as 11 IU/dl in one patient whose pre-heparin level was below the normal range, and to zero in another patient. AT III levels rose after administration of whole blood or fresh frozen plasma, and most patients recovered to within 30 IU/dl of original AT III concentration. In all cases the amount of protamine sulphate administered to neutralize heparin was excessive. The diversity of observed patient responses to heparin, heparin neutralization by PF4 release, half-life of heparin, and AT III levels demonstrates the requirement for monitoring heparin and AT III levels before, during and after cardiopulmonary bypass surgery to maintain heparin concentration within a specified range, to facilitate the exact neutralization of heparin at the conclusion of bypass and to assure adequate levels of endogenous AT III prior to surgery and to maintain sufficient concentration of AT III during bypass to ensure the desired anticoagulant effect.

A COMPARISON OF CHROMOGENIC, FLUOROGENIC, AND FIBRINOGEN SUBSTRATE ASSAYS FOR THE DETERMINATION OF PLASMA HEPARIN. R.L. Bick and N.R. Arbegast. San Joaquin Hematology Oncology Medical Group, California Coagulation Laboratories, Bakersfield, California, and UCLA Center for the Health Sciences, Los Angeles, California.

Assays for plasma heparin are now widely utilized in the clinical laboratory and have been found to be particularly useful for monitoring heparin levels in patients receiving subcutaneous or intrapulmonary heparin and for assessing heparin reversal following cardiopulmonary bypass surgery. Three heparin assays were simultaneously used for 40 clinical samples obtained from patients receiving subcutaneous or intravenous heparin and from patients, at mid-bypass, undergoing cardiopulmonary bypass surgery. Correlation coefficients (r values) were then obtained. The heparin samples ranged from 0.04 to 5.1 units/ml. plasma heparin. The assays used were (1) chromogenic (Kabi-s2238), (2) Fluorogenic (Dade), and (3) Fibrinogen (Cutter). It was found that all assays agreed well. The r value for Cutter vs. Dade was 0.93, for Dade vs. Kabi was 0.92, and for Cutter vs. Kabi was 0.92. These studies and data would suggest that all three assays are quite acceptable for the determination of plasma heparin levels, and the relative advantages and disadvantages of each relate only to technique, time, and instrumentation requirements. Based upon these studies, ease of operation and automation, the Fluorogenic (protopath) system was chosen for monitoring heparin levels and reversal in cardiopulmonary bypass patients.

THE ACTIVATED PARTIAL THROMBOPLASTIN TIME AFTER HEPARIN REMOVAL (aPTT/HR): THE BASIS OF A NEW SINGLE TEST STRATAGEM FOR MONITORING ALL PHASES OF ANTICOAGULATION WITH HEPARIN AND/OR COUMARINS. H.E. Branson, C. Engelberg, A. Fagin, S. Puri, J. Broadbrooks. Department of Pathology, University of California Irvine, Irvine, California, U.S.A.

Safe and accurate monitoring of the transition from the antithrombotic protection afforded by the heparin-antithrombin III complex to that of coumarin modified zymogens has traditionally been provided by special dilutional one-stage prothrombin times. The demonstrated ability of ECTEOLA anion exchange columns to remove heparin from plasma containing acarboxy forms of the vitamin K dependent factors led to the development of an alternative test, the protime after heparin removal (PT/HR) (Am J Clin Pathol 71 (6): 665, 1979). Application of this same technology to the activated partial thromboplastin time (aPTT/HR) results in: 1) a superior alternative to dilutional and ion exchange protimes; and 2) a new single test/pathway scheme for following the heparin, heparin and coumarin, and coumarin phases of anti-coagulation therapy. The fitness of the aPTT for monitoring therapy with heparin alone and coumarins alone has been previously established. Preliminary studies indicate that the aPTT/HR is very similar in its ability to determine the adequacy of anticoagulation to a combination of the PT and aPTT. Comparison of the aPTT/HR with the PT/HR for combined heparin and coumarin monitoring indicated that it: 1) more closely approximates the functional activity of factor X; 2) is equally sensitive to heparin and coumarins; 3) is responsive to more common clinically significant depressions of factors; and 4) is the test of choice for pre-surgical screening.