INHERITED DISORDERS OF PROTHROMBIN — NOT UNCOMMON IN NORTHERN PAKISTAN

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SUMMARY

Coagulation screening tests and factor assays were done on fifty suspected cases of hereditary coagulation defects. Six patients (12%) of hereditary prothrombin deficiency were detected out of 15(30%) cases of rare autosomal recessive clotting defects while 35(70%) cases were due to factor VIII and factor IX defects. The age of presentation of all the patients was below on year. Clinically all of 6 patients presented with severe bleeding manifestations. They all showed marked prolongation of Prothrombin time (P.T) and Activated partial thromboplastin time (A.P.T.T.) and factor level was less than 10% in five out of six patients.

INTRODUCTION

Prothrombin is a Vit K dependent clotting factor (mol wt 72,000), its hydrolysis by factor Xa and thrombin results in the formation of alpha thrombin.[1] Inherited defects of prothrombin are rare disorders.[2] Only bout 50 cases have been reported and described in world literature, but they appear highly diverse.[3] The hereditary transmission is autosomal recessive.[4] Clinical features are not typical but usually severe. The disorder is associated with a bleeding tendency that is inversely proportional to the residual factor. The lowest level noted is 2% and no patient with undetectable prothrombin have been found.[5] These patients show mild to moderate prolongation of P.T, A.P.T.T. and factor II assay is reduced while other Vit K dependent factors are normal.[6] A haemostatic level of factor II being 50u/dl, half life of 2-3 days requires infrequent infusion by prothrombin complex or fresh plasma for management.[7]

MATERIAL AND METHODS

All the patients were interviewed with special attention to family history and consanguinity. Complete physical examination of each patient was carried out for any bruises/petechia/echymotic spots on the body. All investigations were performed according to the standard methods described by Dacie and Lewis.[8] Bleeding time and Hess's test were performed on patients. Haematological material obtained from patients:- Three types of blood samples were collected from patients. First in trisodium citrate (9 to 1 ratio), second in plain glass tube and third in EDTA. For control blood samples were collected from normal subjects in trisodium citrate.

Citrated samples were centrifuged at 2000 g for 10 min to separate platelet poor plasma. Prothrombin time, A.P.T.T. and Thrombin time were performed on citrated plasma samples of both test and control.[9] Clotting time of each patient was noted by the sample contained in plain test tube. Hb, T.L.C., TRB.C platelet count and red cell indices were assessed by haematology analyzer. Factor assays were done on citrated blood samples according to abnormalities detected in screening tests.

Factor II assay. Prothrombin assay was done on those blood samples which showed prolongation of both P.T. and A.P.T.T. commercially available freeze-dried factor deficient plasma (reconstituted according to manufacture's instruction) was used. This preparation contained not more than 1% of factor II but adequate concentrations of other factors necessary for prothrombin time test. Three dilutions 1 in 10, 1 in 50 and 1 in 100 of both test and control PPP were made in barbitone buffered saline. 0.1 ml of patient's or control plasma dilution, 0.1 ml of factor II deficient plasma and 0.1 ml of thromboplastin (rabbit brain) were placed in a water bath at 37°C for 60 sec. 0.1 ml of 0.25mol/l $cacl_2$ solution was added to this mixture and clotting time noted. This procedure was repeated with all the dilutions of both test and control plasma. Clotting times were plotted against dilutions on double log paper. The degree of correction of long prothrombin time of factor II deficient plasma after the addition of different dilutions of test and control plasma was noted and is the basis of factor II assay.

RESULTS

Fifty patients suspected of hereditary coagulation defects were screened to see the occurrence of hereditary prothrombin defects. The clinical and haematological data of all these patients was analysed. Six cases (12%) of prothrombin deficiency out of fifteen (30%) cases of rare autosomal recessive hereditary clotting defects were detected, while 35(70%) of cases were due to factor VIII and IX defects.

The patients with hereditary coagulation defects showed a wide age range of presentation but all the six patients of prothrombin deficiency and the one with fibrinogen defect presented in infancy. As haemophilia and Christmas disease are sex linked recessive disorders, these were exclusively seen in males. The autosomal recessive clotting defects including hereditary prothrombin deficiency also showed an increased M:F ratio. Consanguinity was reported in 67% of patients while a positive family history was given in only 17% cases of hereditary prothormbin deficiency. Table-I summarizes mean for Hb, TRBC, PCV, MCV, MCH and MCHC in all the patients having hereditary clotting defect. None of the patient showed thrombocytopenia. Platelet count ranged between 150-600 $X10^9$. With the exception of VWD and fibrinogen deficiency bleeding time was normal in all the patients including six prothrombin deficient cases. Clotting time was more than 15 minutes in all the cases of prothrombin deficiency, few cases of haemophilia factor V and fibrinogen deficiency. PT was markedly prolonged (> 3 min) in all the patients of prothrombin deficiency and fibrinogen defect while factor VII and factor V deficiency showed a moderate prolongation of PT.

APTT was markedly prolonged (> 3 min) in all the six patients of prothrombin deficiency and also in fibrinogen deficiency, while all of the factor VIII, Factor IX, factor XI and factor V deficient patients showed moderate to marked prolongation of APTT.

TABLE – I

SUMMARY OF HAEMATOLOGICAL DATA IN PATIENTS HAVING HEREDITARY CLOTTING DEFECT

(n=50)

Haematological parameters	Range	Mean
Hb (g/dl)	8.04–13.56g/dl	10.36
TRBC (x 10^{12}/1)	3.02–4.45	3.90
PCV (1/1)	0.295–0.398	0.112
MCV (f1)	64–72	70
MCH (pg)	20.4–22.6	21.5
MCHC (g/dl)	30.2–33.0	32.3

Thrombin time was normal in all the cases except the one with fibrinogen deficiency.

Factor assay

None of prothrombin deficient patient showed > 5% of factor activity. Five out of six patients showed factor activity between 5-10% while one patient showed 11% factor activity. Rest of the H.C.D patients showed factor activity between 1-5% except some of the haemophiliacs and patient of Christmas disease who showed <1% of factor activity.

DISCUSSION

Prothrombin is a single chain polypeptide consisting of a carboxy terminal half, the thrombin forming part of the molecule and an amino terminal half, PT fragment 1-2 that is liberated during activation by factor Xa.[10] This prothrombin fragment F1+2 can be determined in patient's plasma as a measure of thrombin generation.[11] The thrombin so formed not only converts soluble fibrinogen into insoluble fibrin polymers but it also activate factor XIII, V and VIII, can stimulate platelet aggregation and back activate prothrombin.[12]

Various types of genetic abnormalities including point mutation, and gene deletion are seen in prothombin genotype. One one hand these mutations can cause elevated plasma prothrombin levels associated with increased incidence of venous thrombosis,[13,14] while on the other hand these can cause structural variants of prothrombin molecule (dysprothrombinaemias and low level of prothrombin (hypoprothrombinaemia) associated with bleeding tendency.[15,16]

Hereditary prothrombin deficiency is a rare disorder. Only 40 cases were reported in world literature till 1985 (Table –II).[17] In the last fifteen years there is addition of only 10 cases. In our study the patients included were suspected of having hereditary clotting defects by the history and clinical examination. We detected six cases of prothrombin deficiency out of fifty cases of hereditary clotting defect. This reflects that like haemophilia A and haemophilia B, prothormbin deficiency is also not uncommon in Northern Pakistani population. No comparable data is available as almost no work has been done on the incidence of autosomal recessive hereditary clotting defects in Northern Pakistan.

Unlike factor VIII and factor IX, there is little correlation between clinical severity and factor level in hereditary prothrombinaemia. Our observation is the same

TABLE – II

APPROX: NUMBER OF CASES RARE CLOTTING DISORDERS BASED ON CASES REPORTED IN THE LITERATURE AND OTHER DATA (PERSONAL COMMUNICATIONS FROM VARIOUS CENRES, PERSONAL INFORMATION EDUCATED ESTIMATES, ETC.)

Defect	Approx: number of cases
Fibrinogen defects	300
Factor XI defect	200
Factor VII defect	150
Factor XII defect	150
Factor V defect	150
Factor XIII defect	150
Factor X defect	40
Factor II defect	40
Combined factor V and factor III defect	40
Combined factor VII and factor III defect	20
Combined factor VIII and factor IX defect	10
Antithrombin III defects	250
Protein C defects	50
Plasminogen defect	10
a2-Antiplasmin defects	10
Fibrinolysis activators defects	10

as all the six patients presented with severe bleeding symptoms, five having factor levels between 5-10% and one had 11% factor activity. All showed marked prolongation of clotting time, PT and APTT.

It was believed that in hereditary prothrombin deficiency life is not possible if the patient has <10% of factor activity, but patients having 2% of factor activity have been detected and reported.[18] In our study five patients showed factor levels below 10%.

The conclusion is that like factor VIII and IX defects, hereditary prothrombin deficiency is also not uncommon in this area of the country. As consanguinity is positive in 67% of cases, the reason may be intermarriages. Positive family history is observed in only 17% of cases, therefore mutation of gene is important in hereditary prothrombin deficiency as in haemophilia.

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