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Heterozygous FXII deficiency is not associated with an increased incidence of thrombotic events: Results of a long term study☆

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ABSTRACT

Objective: To investigate the incidence of thrombotic events in patients heterozygous for FXII deficiency during a long observation period.

Patients and methods: 103 heterozygotes for FXII deficiency, 49 female and 54 male were followed for 19.6 years (range 5–32 years). As controls 103 unaffected family members of same sex and similar age (± 5 years) were enrolled. The thrombotic end points were: myocardial infarction, deep vein thrombosis and ischemic stroke.

The mean Factor XII level in the heterozygotes was 48.5%: range (35–60%) that of control was 96.5% (range 70–155%).

The heterozygotes showed one myocardial infarction, two deep vein thromboses and no ischemic stroke. The unaffected family members observed 2 myocardial infarctions, one deep vein thrombosis and one ischemic stroke.

There were seven deliveries (five women) among the heterozygotes and six (five women) among the controls. Furthermore, four and five surgical procedures were carried out in the patient and in the control group, respectively.

Immobilization times for surgical procedures or pregnancies were 50 days and 57 days for the heterozygotes and the unaffected family members, respectively.

Heterozygotes for FXII deficiency did not show an increased incidence of thrombotic events as compared with unaffected family members during a long follow up.

1. Introduction

The relation between FXII deficiency and thrombotic events has been a hotly delated subject. The observation that Mr. Hageman, the index patient with this disorder, died in 1968 of pulmonary embolism has been the basis for the existence of a causative role of this deficiency in the pathogenesis of thrombosis [1]. Several case reports published before and after that date have been maintained to be the proof of the validity of the association [2–7].

Subsequent studies have demonstrated that homozygous patients with FXII levels of less than 10% of normal are not associated with an increased risk of thrombotic events [8–11].

Occasional reports of patients with intermediate levels of FXII and others with proven or probable heterozygous with FXII deficiency have been reported to have thrombosis [12–13].

Furthermore, FXII 46C/T polymorphism which causes slightly decreased levels of FXII has been proposed to show an increased incidence of thrombotic events [14].

These data would indicate that, as far as thrombosis is concerned, a mild deficiency could be more dangerous than a severe one. This interpretation has found support in a study which shows that mortality rate in FXII deficiency has a U shaped curve, namely it is linear between the 100% level to the 15% level, but then it turns upward sharply with FXII levels of less than 10% of normal [15]. In other words it would seem that patients with less than 10% FXII activity level have the same mortality rate as subjects with 90% activity. Since thrombotic events play an important part in mortality rates the possibility that a mild to moderate decrease in FXII activity could be, contrary to a severe deficiency, a cause for thrombosis remained open.

The purpose of the present study is to report on the thrombotic incidence seen in a population of proven heterozygotes for FXII deficiencies during a long observation period.

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Table 1
Main results of the follow up study.

	N° of cases	M.I.	Ischemic stroke	Deep vein thrombosis	Comments
Heterozygous FXII deficiency	103	1 a)	0	2 c)	a) Heavy smoker c) One patient took oral contraceptives; in the other case it occurred after trauma.
Unaffected family members	103	2 a)	1 b)	1 c)	a) smokers; one of them had also hypercholesterolemia: b) Transitory ischemic attack. No sequels. Subject had moderate hypertension. c) Venous thrombosis of left leg at the end of pregnancy

2. Patients and methods

All heterozygous patients with FXII deficiency seen in Padua during the years 1968–2010 were followed for a mean period of 19.6 years (range 5–32 years). All these patients were related to the 39 homozygous patients seen during the same period of time, namely they were all members of families which contained at least one proven homozygote [12].

Forty nine were female and fifty four were male for a total of 103 patients. The normal counterparts had the same female-male composition. No patient or control had complained of any thrombotic event before entering the study.

Patients were first diagnosed as heterozygous at the time homozygous patients were discovered. Diagnosis was obtained by clotting tests and molecular biology techniques. The mutation found were −13C > T (1); −8 G > C (2); −1 G > A (3); Gln501Stop; Phe574Leu.

These patients were matched with sex and age (± 5 years) unaffected family members. These were parents or children of the homozygous patients, or otherwise related to them (siblings, cousins, etc.) In seven instances there was no close member of the family available to act as a normal counterpart. In this case distant relatives were chosen.

The patients and the normal counterparts were seen in Padua every year. Caring physicians were also contacted every year in order to clarify or confirm their health condition.

The thrombotic events taken into consideration were Myocardial Infarction (M.I.), venous thrombosis (V.T.) and ischemic stroke (I.S.). The number of prothrombotic conditions (pregnancy, oral contraception, trauma, immobilization) which occurred during the observation period were recorded for both patients and controls.

The thrombotic events had to be demonstrated by objective methods.

Coagulation test were carried out according to accepted procedures as previously reported [11]. Congenital FXII deficient plasma was used as substrate for the FXII activity assays. FXII antigen say was carried out in 36 patients and in 16 controls by the Laurell is method using an anti FXII antiserum obtained or purchased from Behringwerke, Marbug, Germany [16].

The common polymorphisms FV Leiden and GtoA20210 were investigated in almost all patients and controls by means of accepted procedures as previously reported [17,18].

Both patients and controls during and after the thrombotic event took oral anticoagulants and heparin or antiplatelets drugs for a variable period of time and dropped out of the study. The corresponding control or patient was concomitantly excluded.

During the study six patients and five controls took for short periods of time, aspirin, antipyretics, cough medications several antibiotic and non-steroid anti-inflammatory drugs.

There were four surgical procedures among the patients (inguinal hernia repair, appendectomy, benign thyroid node removal and hemorrhoidectomy). In the control group, the surgical procedures were five (caesarean section, appendectomy, skin cyst removal, laparoscopic gall-bladder removal, duodenal ulcer surgery).

There were seven pregnancies in five women among the patients and six in five women among the controls one of the latter required a caesarean section. One patient took oral contraceptives for about two years. Medication was stopped because of the appearance of a deep vein thrombosis.

In the control group, two women took oral contraception for about one year without complication.

Patients and normal counterparts were informed of the scope of the study and gave their consent. The study was carried out according to the Helsinki agreement.

3. Results

FXII activity level in the patients varied between 35 and 60% of normal (mean 48.5%). FXII activity level in the unaffected family members varied from 70 to 155% (mean 96.5%).

FXII antigen was found to be around 50–60% of normal in the heterozygotes and around 100% of normal in the controls.

One patient and one control subject were found to be heterozygous for FV Leiden whereas no GtoA20210 prothrombin polymorphism was found.

The heterozygotes with FXII deficiency showed one myocardial infarction, two venous thrombosis and no stroke (Table 1). The basal FXII activity levels in these patients were: 50, 55 and 58% of normal (mean 54.3%).

The unaffected family members showed two myocardial infarctions, one deep vein thrombosis and one stroke (Transitory Ischemic Attack). The basal FXII activity levels in these controls were: 95,105,100 and 102% of normal (mean 100.2%).

The patient with myocardial infarction was a smoker whereas the two deep vein thrombosis occurred in a woman who took oral contraceptives for about two years and in a male patient after a trauma and consequent immobilization.

Among the normal counterparts both subjects who presented with M.I. were smokers and one of them had also a slight hypercholesterolemia. The venous thrombosis of the left leg occurred at the end of pregnancy. The stroke observed in the control group occurred in a subject with moderate hypertension. It was a TIA since no sequel was evident and a Brain CAT was negative.

Antithrombin III, Protein C and Protein S were evaluated in both the patients and the controls who showed thrombotic events and found normal in every case.

Among the patients, pregnancies and deliveries were uneventful in all cases. In the control groups, one delivery was carried out by caesarean section and another woman presented with a deep vein thrombosis of the left leg at the end of pregnancy. She was treated with local measures and with subcutaneous unfractionated heparin.

Approximate periods of immobilization were 50 days and 57 days for the heterozygotes group and for the normal counterparts, respectively (Table 2). The AB0 group status was available for about 60% of the patients and of the normal counterparts and found to be similar. Specifically, the non 0 groups were similarly distributed among patients and unaffected family members.

4. Discussion

It seems settled by now that Factor XII deficiency neither causes nor prevents thrombosis.

Recent studies have demonstrated that there is no difference in the

Table 2
Congenital and acquired prothrombin conditions found during the observation period. Het = heterozygote.

	Congenital defect	Pregnancies	Oral contraception	Surgical procedures	Approximate immobilization (days)	Comments
Heterozygous FXII deficiency	Het. FV Leiden a)	7 b)	1 c)	4 d)	50	a) One patient. b) Seven pregnancy in five women. Uneventful. c) Patient took oral contraception for two years. d) Venous thrombosis occurred after a trauma and immobilization.
Unaffected family members	Het FV Leiden a)	6 b)	2 c)	5 d)	57	a) One control b) Six pregnancies in five women. One pregnancy required caesarean section. c) Oral contraceptives for about one year without complications. d) Uneventful.

prevalence of thrombosis among patients with severe FXII deficiency and general controls or unaffected family members counterparts [8,11,19].

However, moderate FXII deficiency has been claimed to play a role as a cause of thrombosis [12,13,14]. This was mainly based on the observation that mortality in patients with FXII deficiency follows a U curve [15].

Patients with less than 10% level of FXII show a mortality rate similar to that shown by patients with 90% FXII level. Mortality rates, according to this study, progressively increases as FXII decreases to a 10–15% level but then they would decrease and be similar to that of subjects with normal FXII level even if patients had FXII levels of less than 10% of normal [15].

This is a puzzling biological phenomenon which would indicate that heterozygous patients show a worse mortality rate than homozygous or compound heterozygous patients.

It is difficult to conceive a switch of the trend opposite to what is commonly seen for blood coagulation proteins. The lower the factor, the more severe the clinical manifestations. A possible exception to this rule may be represented by FVII deficiency in which there is, sometimes, no strict correlation between FVII level and bleeding tendency [20].

Since thrombotic events play an important role in mortality rates, the observation of the U curve phenomenon could have a great clinical importance.

We would be obliged to be more concerned about a mild or moderate deficiency than about a severe one.

The present study supports the finding by Wu et al. in the Chinese population.

These Authors had shown that the link between heterozygous FXII deficiency and thrombosis is weak if any. Furthermore, the controls, in the Chinese study were not unaffected family members.

That paper confronted in fact the presence of thrombosis in heterozygous patients with FXII deficiency with that seen in normal, unrelated controls [21].

No systematic evaluation of the 46 C to T polymorphism has been carried out in our group of known heterozygous or in the normal counterparts. However since the FXII levels found in the carries of this polymorphism are only slightly higher than those, not that found in our proven heterozygous patients the results may apply to them too [14]. Furthermore, association between thrombosis and this polymorphism is only episodic. No controlled study has even been carried out.

Taken all together, the clinical studies on the effect of FXII deficiency and thrombosis indicate that the defect neither causes nor protects from thrombotic events. Furthermore, it may be stated that the U curve phenomenon may apply to mortality rates but not to thrombotic events. The report of thrombosis in a patient with FXII deficiency, as with any other clotting factor, is the result of the common, understandable impulse to attribute a causal link between the two facts. One should always remember that association between two events does not necessarily indicate the presence of a causal relation. Epidemiological and long observational studies have demonstrated that this is not the case for FXII deficiency [8,11,19].

These clinical observations are in contrast with experimental studies in mice which showed that a decrease of FXII level could diminish the development or the progression of thrombosis [22–26].

The authors declare that they have no conflict of interest.

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