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Congenital prothrombin defects: they are not only associated with bleeding but also with thrombosis: a new classification is needed

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ABSTRACT

Objective: Congenital prothrombin deficiency is one of the rarest clotting disorders. It is commonly subdivided in Type I defects or cases of 'true' prothrombin deficiency characterized by a concomitant decrease in FII activity and antigen and in Type II or dysprothrombinemias, in which FII activity is low but FII antigen is normal or near normal. A bleeding tendency, often a severe one, is the hallmark of the two-defects even though the bleeding is usually less severe in the Type 2 defects or dysprothrombinemias.

Patients and Methods: An extensive search of published cases of prothrombin deficiency was carried out in Pubmed and Scopus. The search started in 2012, after the publication of the first family with dysprothrombinemia and venous thrombosis. A few additional families were found.

Results: Recent studies have demonstrated that the Type 2 defects are heterogeneous. Several heterozygous mutations involving the Arg596 residue of exon 14 have been demonstrated not be associated with a bleeding tendency but, surprisingly, with venous thromboses. Mutations in close areas of prothrombin have failed to show the same pattern.

Conclusions: These observations have required a reclassification of prothrombin defects. To the Type I and Type II defects, a Type III has to be added characterized by the absence of bleeding and the presence of venous thrombosis. It is not clear yet if this special variant of Type II defect is limited to the Arg596 mutations or if other residues may be involved.

KEYWORDS

Prothrombin deficiency;
dysprothrombinemia;
thrombosis; bleeding

Introduction

Prothrombin deficiency is one of the rarest coagulation disorders with a prevalence of about 1:1.500.000 [1–3]. The defect is usually classified in Type I in which there is a concomitant decrease of prothrombin activity and antigen and in Type II in which prothrombin activity is low, whereas antigen is normal or near normal. The first type is also known as 'True' deficiency or hypoprothrombinemia, whereas the latter is known as dysprothrombinemia. Association of hypoprothrombinemia with dysprothrombinemia (hypo-dysforms) has also been described. The same is true for the combination between two dysprothrombinemias (dys-dysforms) [1–3].

Altogether about 150 cases due to several mutations have been described so far. The defect has been described all over the world but it seems relatively more frequent in Latin countries and especially in Puerto Rico [1,4,5]. In this small island, isolation has allowed the diffusion of the autosomal recessive defect. It is estimated that, in that island, it may represent the third most frequent clotting defect after the hemophilias and von Willebrand Disease [4]. Prothrombin deficiency has been associated with a bleeding tendency, often a severe one. Complete absence of prothrombin is maintained to be incompatible with life [1–3].

Even homozygous with severe bleeding have prothrombin levels around 1% of normal. Usually compound heterozygotes between two Type 1 defects with hypoprothrombinemia have levels between 1 and 5% of normal and are always symptomatic. Compound heterozygotes between a Type 1 and a Type 2 defect have usually activity of 5–10% of normal but antigen may be 50–60% of normal [1–3,5]. They also may present an important bleeding tendency. Heterozygotes who have prothrombin activity around 40–55% of normal do not show usually spontaneous bleeding but they may bleed in case of surgery, deliveries or tooth extractions [1–3].

Till a few years ago no thrombosis has ever been described in patients with prothrombin deficiency and the condition was always considered a bleeding disorder, often a severe one. This assumption was demonstrated wrong in 2012 when a family with dysprothrombinemia with venous thrombosis was reported in Japan [6]. A few similar families were reported during the following years in Serbia, India, Italy [7–10]. All these patients had a mutation of amino acid Arg596. It was substituted by a leucine, a glutamine as a tryptophan. Another important feature was the absence of bleeding and the fact that the patients were all heterozygotes [6–10].

These new findings have clearly demonstrated that a new classification of prothrombin defects was needed.

Updated classification

Today we can classify prothrombin defects in three groups, namely: (1) true prothrombin deficiency (2) dysprothrombinemias with variable bleeding tendency and (3) dysprothrombinemia without bleeding but with thrombosis.

True prothrombin deficiency

The condition is characterized by a concomitant decrease of FII activity and FII antigen. Usually lower than 5% of normal. The condition is always associated with an important bleeding tendency. Even heterozygotes with FII levels of about 40–50% of normal may sometimes present undue bleeding during delivery, surgical procedures or tooth extractions [1–3].

Dysprothrombinemias with variable bleeding tendency

Dysprothrombinemias are defects characterized by variably activity, usually around 5–10% of normal and normal or near normal FII antigen. Occasionally, FII activity may be lower than that.

The bleeding tendency is variable but surely, less severe than that of the cases of ‘true’ deficiency. In some cases bleedings may be mild.

Dysprothrombinemias with thrombosis

Our knowledge about the dysforms have changed during the last few years. Actually the first data about the existence of a severe procoagulant state associated with a mild bleeding tendency dated back to 2002 [11]. In that year Akhavan et al. reported an 11 years old girl from Iran who had only a mild-to-moderate bleeding tendency despite having prothrombin level of less than 1% but an antigen level of 61%. On the suspicion of a dysfunctional protein further studies showed that the underlying mutation was Arg67His substitution (chymotrypsinogen numbering that is equivalent to the Arg382His mutation). Furthermore, it was demonstrated a decreased activation of protein C, a decreased binding to thrombomodulin, a severe impairment of thrombin inhibition by heparin cofactor II and, finally, a decrease of prothrombin activation by the FXa–FVa complex. The complex defect caused by the Arg67His mutation was cautiously concluded to affect both the coagulant and the anticoagulant activities of thrombin. It was thought that there was a balance between these two opposing forces [11]. This interesting observation remained silent for several years and the prothrombin

defects were always considered to be responsible of a bleeding tendency. Several dysprothrombinemias were reported but no thrombosis event was ever mentioned [2,3,12–17].

Then, in 2012, a female patient from Japan who had dysprothrombinemia was reported; she had no bleeding but a venous thrombosis [6]. The proposita was 12 years old and had her first thrombosis at the age of 11. The patient was demonstrated to have a prothrombin abnormality due to an Arg596Leu mutation that possessed a gain of function. The gain of function consisted in an increased resistance toward antithrombin. Immediately, after this first report, other similar cases were reported involving always the AA Arg596 but substituted by Gln or Trp, instead of Leu as in the original case [8–10].

The main features of these dysprothrombinemias are gathered in Table 1. The most important common aspect presented by this group of patients, regardless of the country of origin, Japan, Serbia, Italy, was the occurrence of venous thrombosis at a young age (11–27 years). In addition, they were also all heterozygotes and had no bleeding tendency. At about the same time a patient from India was reported (prothrombin Amrita) with a dysprothrombinemia due also to an Arg596Gln mutation [7]. However, this case shows some differences from the previous group, despite the same mutation, since the venous thrombosis occurred at the age of 60. Furthermore, there was no family story [7].

Recently, Ding et al. [18] reported a family in which there are two patients with an Arg382His mutation and an important bleeding tendency, one of whom had a post-partum deep vein thrombosis (DVT) and pulmonary embolism (PE) (Table 2).

The interesting peculiarity of this new family is that the proposita and her sister are homozygous for the defect (FII activity 1%; FII antigen, normal). All the other previous patients were heterozygous. The proposita is a 30 years old Chinese who had a DVT and PE 28 days post-partum and was treated with low molecular weight heparins and fresh frozen plasma. It is doubtful that this patient may be similar to the previous cases because the patient had received during pregnancy, because of bleeding, 600 Units of a prothrombin complex concentrates (PCC) every week, beginning on the sixth week of gestation for an approximate total of 20,400 Units. This prolonged replacement therapy could have created on the long run a thrombophilic state with consequent thrombosis. The patient never had other thrombosis. Furthermore, her homozygous sister never had thrombosis.

The Arg382His mutation does not belong to the classic group which involves Arg596Leu or Gln or Trp mutations. Due to the different site of the mutation, the condition has to be considered a dysprothrombinemia with both bleeding and thrombosis secondary to

Table 1. Cases of prothrombin abnormalities due to mutations of Arg596 that are associated with a gain of function towards antithrombin and that therefore cause a thrombophilic state.

Authors, year	Age, sex	FII act	FII ant	Bleeding	Venous thrombosis (age at first episode)	Associated Risk	Mutation	Genotype	Eponym	Comments
Miyawaki et al. (2012) [6]	17, F	37.6	63.8	No	Yes (11 Years)	No	Arg596Leu	Het.	Prothrombin Yukuhashi	Patient from Japan
Djordjevic et al. (2013) [8] Fam 1	n.r., F	n.r.	n.r.	n.r.	Yes (17 years)	No	Arg596Leu	Het.	Prothrombin Belgrade	Six patients in two families
Fam 2	n.r., F	n.r.	n.r.	n.r.	Yes (16 years)	No	Arg596Leu	Het.		
Sivasundar et al. (2013) [7]	60, M	n.r.	n.r.	n.r.	Yes	n.r.	Arg596Gln	Het.	Prothrombin Amrita	No founds study
Kishimoto et al. (2016) [9]	23, F	n.r.	n.r.	No	Yes (15 years)	No	Arg596Leu	Het.	Prothrombin Belgrade	Patient from Japan
Bulato et al. (2016) [10] Fam. 1	47, M	54	80	No	Yes (38 years)	No	Arg596Trp	Het.	Prothrombin Padua	Seven patients in two families
Fam. 2	29, F	29	89	No	Yes (27years)	No	Arg596Trp	Het.		

Note: Het. = Heterozygote; n.r. = not reported. (a) Data supplied in 'Correspondence'. N. Engl Med 2012; 367:1069–1070. Only the families with the Arg596 mutations are included.

Table 2. Main features of the prothrombin defect, a dysprothrombinemias due to mutations not involving the Arg596 amino acid but still associated with thrombosis.

Authors, year	Age, sex	FII act	FII ant	Bleeding	Venous thrombosis (age at first episode)	Associated risks	Mutation	Genotype	Eponym	Comments
Ding et al. (2017) [18]	30, F	1	82.9	Yes, severe	30	Pregnancy: Replacement therapy with PCC during most of the pregnancy	Arg382His	Hom	n.r.	A sister similarly affected but without thrombosis

intensive replacement therapy even though a contributory effect of other factors cannot be excluded [18]. Thrombotic events, both arterial and venous, have been described in several coagulation factors deficiencies after or during replacement therapy [19–23].

Discussion

Un updated classification of prothrombin defects is the following: Type I or cases of 'true' prothrombin deficiency; Type II or cases with dysprothrombinemia and a variable bleeding tendency, sometimes only mild and Type III cases of dysprothrombinemia with no bleeding but instead a prothrombotic state.

The observation that a clotting factor may be associated both with bleeding and thrombosis, depending on the site of the mutation has important implications. It will be of paramount importance to know the precise area of the protein which is critical for this shift in behavior. Several dysprothrombinemias with mutations in sites of the protein upstream on downstream of the Arg596 site have been described but no association with thrombosis has ever been reported (Table 3). These dysprothrombinemias are prothrombin Salakta (Gln509Ala), prothrombin Greenville (Arg517Gln), prothrombin Perija (Gly548Ala), prothrombin Scranton (Lys599Thr) and others show mutation close to the Arg596 amino acid but apparently no venous thrombosis has ever been reported [12–17,24–26]. As far as we can tell the critical area seems to reside around Arg596, site of the mutation of the cases associated with a demonstrated occurrence of venous thrombosis.

These observations have further complicated our appraisal of the clotting mechanism. It is noteworthy to indicate that all patients with mutations in this area but one, prothrombin (Leiden) [24] are Type 2 defects suggesting a role of the FII antigen in the potential occurrence of thrombosis. This may explain why no thrombotic event has ever been described in patients with true prothrombin deficiency (Type I defect) [27].

The new studies on prothrombin have modified our understanding of prothrombin defects. The exclusively bleeding tendency remains valid only for patients with 'true' deficiency [27]. Furthermore, the fate of the dysprothrombinemias results drastically changed. The dysprothrombinemias should be divided in a group with only a mild bleeding tendency, tendency (Scranton, Perija, etc.) and a group associated with venous thrombosis and no bleeding (Yukuhashi, Belgrade, Padua, etc.). The presence of prothrombin antigen level seems to play a pivotal role.

However, by itself it cannot explain the entire picture. Critical seems also the site in which the mutation occurs. The area around Arg596 seems critical

Table 3. Main features of prothrombin defects close to the Arg596 amino acid.

Eponym	Salakta	Greenville	(Naetsved)	(Palermo)	C.Christi	Perija	Yukuhashi, Belgrade, Amrita, Padua 2	Scranton	Quick	(Leiden)
References	[15]	[13]	[18]	[26]	[27]	[17]	[7–11]	[16]	[14]	[25]
Mutation	Glu509Ala	Arg560Gln	Ala563Val + Gln519Stop	Arg581Cys	Gly584Stop + Arg475Cys	Gly591Ala	Arg596Leu Arg596Gln Arg596Trp	Lys599Thr	Gly601Val + Arg425Cys	Trp612Stop + IVS2-25C G
Genotype	Hom	Het	C. Het	Hom	Comp.Het	Hom	All.Het	Het	Comp.Het	Comp.Het
Bleeding	No	No	Mild	Severe	No	Mild	No	No	Severe	Severe
Thrombosis	No	No	No	No	No	No	Yes	No	No	No
Country	Tunisia	U.S.A.	Denmark	Italy	Latin America	Venezuela	Japan, Serbia, India, Italy	U.S.A.	Latin America	Holland
Type of defect	II	II	II	II	II	II	II	II	II	I
Amino acid	509	550	563	581	541	591	596	599	601	612
		Arg	Ala	Arg	Gln	Gly	Arg	Lys	Gly	Trp

Note: No thrombotic event has been reported in any of these patients who are mainly affected by dysprothrombinemias or Type II defects. Only a defect due to a compound heterozygosis between Trp617Stop and a splicing defect is a Type I defect. Amino acid residues numbering includes the 43 residues of the pre-pro-sequence. Eponyms between parentheses have been tentatively supplied, for the sake of completion, by the authors of the present paper.

for the occurrence of thrombosis. The extension of that area is unknown. Unfortunately, we do not know if patients with dysprothrombinemias due to mutations in areas close to the Arg596 amino acid had thrombotic events after the publication of the original reports.

The 'thrombotic' area probably involves only a few AA and it remains for future research to establish where the breaking points exist, if any. In fact, dysprothrombinemias both downstream and upstream of the Arg596 have been reported with no apparent association with thrombosis.

These developments on prothrombin are in agreement with recent studies involving FIX. A mutation (Arg338Lys) in exon 14 of FIX (FIX Padua) has been associated with a thrombophilic state due to highly increased FIX activity [28]. Here, again a factor which is involved in a severe bleeding condition, hemophilia B, may turn to be thrombophilic. The fact that some studies have demonstrated the rarity of this FIX mutation does not diminish the importance of the finding [29,30] as shown by the attempts to utilize this mutation to prepare high potency concentrates [31,32].

Reviews on prothrombin defects have to be rewritten since even the most recent ones had no opportunity to deal with these new developments [2,3].

Today, only Factor X defects remain thrombosis free [27]. FII and FIX defects could be defined as thrombohemorrhagic conditions or, better, as hemorrhagic-thrombotic disorders.

Disclosure statement

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