Reduction of Contact Activation Related Fibrinolytic Activity in Factor XII Deficient Patients

Further Evidence for the Role of the Contact System in Fibrinolysis In Vivo

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Abstract

In this study the contribution of activation of the contact system to activation of the fibrinolytic system in vivo was investigated in healthy volunteers and in factor XII deficient patients.

The plasminogen activating activity in plasma from healthy volunteers after infusion of desamino D-arginine vasopressin (DDAVP) was only partially blocked (for 77%) with specific antibodies to tissue-type plasminogen activator and urokinase-type plasminogen activator. The residual activity could be quenched by a monoclonal antibody that inhibits factor XII activity and was not present in patients with a factor XII deficiency. The formation of plasmin upon the DDAVP stimulus as reflected by circulating plasmin-α2-antiplasmin complexes was lower in factor XII deficient patients than in healthy volunteers. Activation of the contact system occurred after DDAVP infusion in healthy volunteers and was absent in factor XII deficient patients.

These results indicate that DDAVP induces a plasminogen activating activity that is partially dependent on activation of the contact system and that contributes to the overall fibrinolytic activity as indicated by the formation of plasmin-α2-antiplasmin complexes. This fibrinolytic activity is impaired in factor XII deficient patients which may explain the occurrence of thromboembolic complications in these patients. (*J. Clin. Invest.* 1991. 88:1155–1160.) Key words: plasminogen activator • plasminogen • plasmin • hemostasis • thrombosis

Introduction

The conversion of the fibrinolytic proenzyme plasminogen into the proteolytic enzyme plasmin may occur in vivo via several pathways (1, 2). The two most extensively studied are the tissue-type plasminogen activator (t-PA)[1] and urokinase-type plasminogen activator (u-PA) dependent pathways. The exact contribution of both pathways to the development of endogenous fibrinolytic activation under basal and activated conditions has not been precisely determined.

Activation of plasminogen in vivo can reproducibly be studied by a short lasting infusion of desamino D-arginine vasopressin (DDAVP). This substance induces increases in plasma levels of plasminogen activator activity and of both t-PA and u-PA antigen (3–5). In addition, it induces activation of plasminogen in vivo as reflected by increasing levels of plasmin-α2-antiplasmin (PAP) complex levels (Levi, M., et al., manuscript submitted for publication). Previous experiments have shown that quenching of both t-PA and u-PA by specific antibodies in plasma obtained from healthy volunteers shortly after infusion of DDAVP, does not completely abolish plasminogen activator activity (5), indicating the existence of a third pathway of plasminogen activation. This third pathway of plasminogen activation has been claimed to be dependent on activation of the contact system of blood coagulation (6). Involvement of the contact system (for reviews see Colman et al. [7] and Kaplan et al. [8]) in fibrinolysis is supported by several lines of evidence. First, patients with severe factor XII deficiency have a higher incidence of thromboembolic events, which might be attributed to an impaired fibrinolytic activity (9, 10). Second, recent observations in patients with hereditary angioedema, which is caused by a deficiency of the major inhibitor of the contact system, C1-inhibitor, have shown that increased activity of the contact system is associated with an enhanced plasmin generation (11). Finally, factor XII structurally resembles t-PA and plasminogen (12).

To determine the role of contact activation in fibrinolysis we investigated the effect of intravenous administration of DDAVP on the activation of the contact system and the fibrinolytic system in healthy subjects and individuals with mild or severe factor XII deficiency. In addition, we studied the contribution of factor XII to the plasminogen activator activity induced in plasma by DDAVP in these subjects. Our results show that DDAVP in vivo induces activation of the contact system and the generation of plasminogen activator activity and plasmin generation which is dependent in part on factor XII. This response was impaired in factor XII deficient patients.

Methods

Study subjects and DDAVP test. Six healthy volunteers (aged 21–33 yr) and in addition six subjects with a known deficiency of coagulation factor XII were studied. Of the deficient subjects, three patients (aged 21–28 yr) had factor XII activity levels below 1% (severe factor XII deficiency) while three other patients (aged 18–44 yr) had factor XII activity levels ranging from 20% to 35% (mild factor XII deficiency).

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1. *Abbreviations used in this paper:* ANOVA, analysis of variance; DDAVP, desamino D-arginine vasopressin; PAP, plasmin-α2-antiplasmin; t-PA, tissue-type plasminogen activator; u-PA, urokinase-type plasminogen activator.

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All study subjects received DDAVP (Desmopressin, MINRIN®, Ferring AB Pharmaceuticals, Malmö, Sweden), at a concentration of 0.4 μg/kg body weight in 50 ml of saline over 15 min intravenously. They were seated 10 min before and during the entire experiment, fasted overnight, and abstained from smoking for 12 h before the experiment. The DDAVP infusion started at 9:00 a.m. The study was approved by the Institutional Review Board.

Collection of blood. Blood was collected by separate venipunctures before and at 15, 30, and 60 min after the start of the DDAVP infusion. If post-DDAVP plasma is mentioned further, the sample taken 15 min after the start of the infusion is referred to.

Blood (9 vol) for the measurement of plasminogen activator activity, t-PA antigen, u-PA antigen, factor XII activity, factor XII antigen, and prekallikrein antigen was collected in plastic syringes containing 3.2% sodium citrate (1 vol). For the measurement of PAP complexes, factor XII-C1-inhibitor complexes and kallikrein-C1-inhibitor complexes blood was collected in siliconized vacutainer tubes (Becton Dickinson, Plymouth, UK) to which Polybrene (hexadimethrine bromide; Janssen Pharmaceutica, Beerse, Belgium) and EDTA (0.05% wt/vol and 10 mM, respectively, final concentrations) were added to prevent in vitro complex formation. All blood samples were immediately immersed in melting ice and centrifuged at 4°C for 20 min at 1,600 *g*. Plasma samples were stored at −70°C until assayed.

Measurement of fibrinolytic activation. Plasminogen activator activity was assayed by an amidolytic assay (13). Briefly, 25 μl of euglobulin fraction of plasma was mixed to a final vol of 250 μl with 0.1 mol/liter Tris HCl pH 7.5, 0.1% (vol/vol) Tween 80, 0.30 mmol/liter S-2251 (Kabi Haematology, Mölndal, Sweden), 0.13 mol/liter plasminogen, and 0.12 mg/ml CNBr fragments of fibrinogen (both obtained from Kabi Haematology). The assay was performed in the absence and presence of specific antibodies against t-PA (0.25 μg/ml), u-PA (0.25 μg/ml), or factor XII (0.25 μg/ml), and mixtures of these antibodies. The antibodies used in this assay were affinity purified monospecific polyclonal rabbit antibodies against t-PA or u-PA, obtained as described previously (14), or monoclonal antibody OT-2, which was raised against factor XII and inhibits amidolytic activity of factor XIIa (Dors, D., manuscript in preparation). Antibodies were added in excess, as was revealed by similar inhibition results upon incubation with a fivefold higher or lower antibody concentration (data not shown). Results were expressed in percentage of plasminogen activator activity in the post-DDAVP plasma samples in the absence of antibodies.

t-PA antigen was measured with an ELISA (Diagnostica Stago, Asnieres-sur-Seine, France) (15). u-PA antigen was measured with a sandwich ELISA, as described before (16). Plasmin-α2-antiplasmin complex levels were determined by an RIA as previously described (Levi, M., et al., manuscript submitted). Briefly, samples to be tested were incubated with monoclonal antibody AAP-11, directed against complexed and inactivated α2-antiplasmin, coupled to Sepharose (CNBr-activated Sepharose 4B; Pharmacia Fine Chemicals, Uppsala, Sweden) by head over head rotation for 4 h at room temperature. The Sepharose beads were washed and bound complexes were detected by subsequent incubation with ^{125}I-labeled monoclonal antibody against plasmin (AP-1). After another washing procedure Sepharose bound radioactivity was measured with a multichannel gamma counter. As a standard, serial dilutions of pooled plasma in which a maximal amount of PAP complexes (1 μmol/liter) was generated by the addition of urokinase, were used. Results obtained with tested plasma samples were related to this standard and expressed as nanomoles per liter.

Measurement of activation of the contact system. Factor XII activity was determined by a standard clotting assay. Factor XII antigen and prekallikrein levels were measured by RIA's as described previously (17). Briefly, plasma samples were incubated with either anti-factor XII monoclonal antibodies (F3) or anti-prekallikrein monoclonal antibodies (K15) coupled to Sepharose. Bound factor XII or prekallikrein was detected by subsequent incubation with ^{125}I-labeled polyclonal antibodies against factor XII or prekallikrein. Serial dilutions of pooled fresh plasma were used as standard and results obtained with test samples were expressed as a percentage of the amount of factor XII or prekallikrein in this plasma. These antigen assays measure factor XII or prekallikrein irrespective of its molecular forms, i.e., native protein or activated protein in complex with C1-inhibitor.

Factor XIIa-C1-inhibitor complexes and kallikrein-C1-inhibitor complexes were assayed by RIA as described previously (17). Briefly, a monoclonal antibody that specifically binds complexed C1-inhibitor (KOK-12) was coupled to Sepharose and incubated with plasma samples. Bound factor XII- or kallikrein-C1-inhibitor complexes were quantitated by subsequent incubation with either ^{125}I-labeled polyclonal anti-factor XII or ^{125}I-labeled anti-kallikrein antibodies, respectively. Results obtained with tested plasma samples were expressed in milliunits, as calculated by reference to a standard curve that consisted of dextran sulphate-activated pooled plasma (which contains 1 U of complexes per ml).

Statistical analysis. Means of multiple groups were compared with analysis of variance using the Newman-Keuls' test to correct for multiple comparison. Student's *t* test was used to compare means between paired groups. All calculations were done using the Number Cruncher Statistical Package (Number Cruncher Statistical System, Kaysville, UT). *P* values below 0.05 were considered statistically significant.

Results

Plasminogen activator activity induced by DDAVP in normal subjects. Plasma levels of plasminogen activating activity, t-PA antigen and u-PA antigen before and 15 min after the infusion of DDAVP in healthy volunteers are given in Table I. As expected, the infusion of DDAVP in healthy volunteers resulted in a significant nearly sevenfold increase in fibrinolytic activity as reflected by the increase in plasminogen activating activity. Plasma levels of t-PA antigen and u-PA antigen increased 3.7-fold and 1.7-fold, respectively.

Fig. 1 shows the effect of the addition of anti-t-PA, anti-u-PA, anti-factor XII, or mixtures of these antibodies on plasminogen activating activity in post-DDAVP plasma of healthy volunteers. Addition of anti-t-PA antibody resulted in an inhibition of plasminogen activating activity of 59% (SEM 4.4) whereas addition of anti-u-PA antibody reduced plasminogen activating activity with 24% (SEM 4.9). The combination of anti-t-PA and anti-u-PA antibodies did not completely inhibit plasminogen activating activity (mean inhibition 77%, SEM

Table I. Plasminogen Activation upon DDAVP Infusion

	Normal subjects		Factor XII < 50%		Factor XII < 1%	
	Before DDAVP	After DDAVP	Before DDAVP	After DDAVP	Before DDAVP	After DDAVP
Plasminogen activating activity	100	678 (52)	100	640 (70)	100	598 (42)
t-PA antigen	5.2 (1.3)	19.3 (1.2)	4.9 (0.9)	17.4 (1.4)	4.5 (1.2)	17.8 (1.2)
u-PA antigen	3.1 (0.3)	5.4 (0.4)	2.9 (0.4)	5.5 (0.6)	3.3 (0.5)	5.9 (0.5)

Plasminogen activating activity (percentage of preinfusion level), t-PA antigen (ng/ml), and u-PA antigen (ng/ml) in normal subjects (n = 6), patients with mild (n = 3) and severe factor XII deficiency (n = 3) before and 15 min after the start of an infusion with DDAVP. Mean values (and SEM) are given. Differences between the three study groups in levels before and after DDAVP were not significant (Student's *t* test).

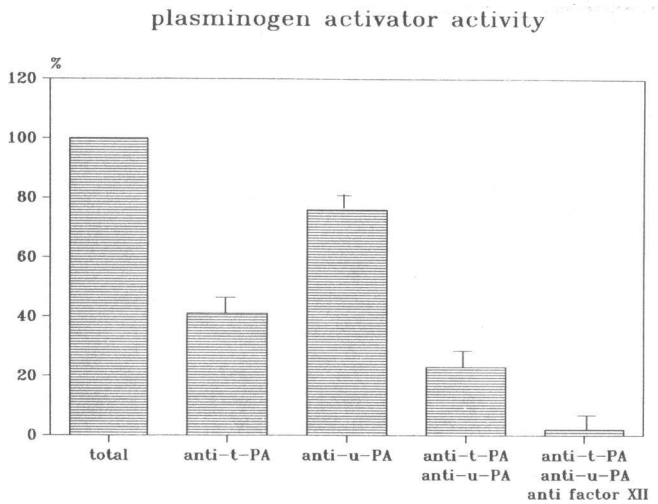

Figure 1. Inhibition of DDAVP-induced plasminogen activator activity by antibodies against t-PA, u-PA, and factor XII activity. Euglobulin fractions were prepared from six healthy donors 15 min after DDAVP infusion. Plasminogen activating activity in each sample was arbitrarily set at 100%. Each sample was preincubated with either anti-t-PA, anti-u-PA, or anti-factor XII antibodies, or combinations, and residual plasminogen activator activity was assessed. Results represent mean values and SEM.

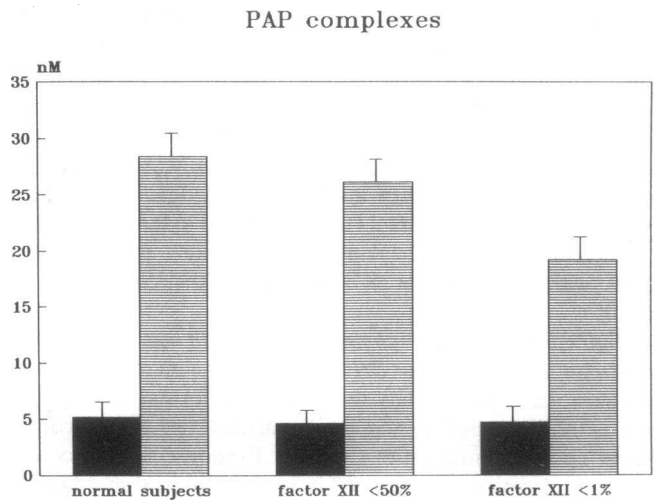

Figure 3. Plasma levels of PAP complexes induced by DDAVP in normal individuals and factor XII deficient patients. *Left bars:* plasma level before DDAVP infusion; *right bars:* plasma level 15 min after the start of the DDAVP infusion. Data represent mean values, bars indicate SEM. Normal subjects (n = 6), patients with mild (n = 3) and severe factor XII deficiency (n = 3) are compared.

2.7), indicating the presence of non-t-PA or u-PA dependent fibrinolytic activity in post-DDAVP plasma. This residual plasminogen activating activity could be quenched by addition of a monoclonal antibody against factor XII activity to the combination of anti-t-PA and anti-u-PA antibodies (mean inhibition 98%, SEM 2.0).

Plasminogen activator activity induced by DDAVP in patients with factor XII deficiency. The increase in plasminogen activating activity in plasma obtained 15 min after the start of the DDAVP infusion in severe factor XII deficient patients was less than that in the healthy subjects (678% [SEM 52] vs. 598% [SEM 42]); however, this difference did not reach statistical significance, probably due to the variation in the assay and the small number of factor XII deficient patients studied. Post-DDAVP plasminogen activating activity in plasma from patients with a mild factor XII deficiency did not differ from that in healthy subjects (Table I).

Fig. 2 shows the effect of the addition of anti-t-PA, anti-u-PA, or the combination of both antibodies on the plasminogen activating activity in post-DDAVP plasma of patients with mild or severe factor XII deficiency in comparison to that of

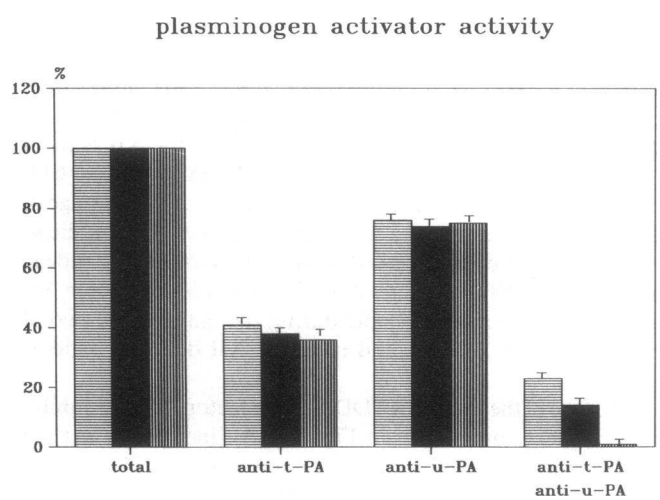

Figure 2. DDAVP-induced plasminogen activator activity in factor XII deficient patients in comparison to that in healthy individuals. Plasminogen activating activity observed in plasma obtained 15 min after the start of DDAVP infusion in each individual was arbitrarily set at 100%, results obtained in the presence of antibodies (anti-t-PA, anti-u-PA, and the combination of anti-t-PA and anti-u-PA) were related to this. *Left bars:* normal individuals (n = 6); *middle bars:* patients with mild factor XI deficiency (n = 3); and *right bars:* patients with severe factor XII deficiency (n = 3). Results represent mean values and SEM.

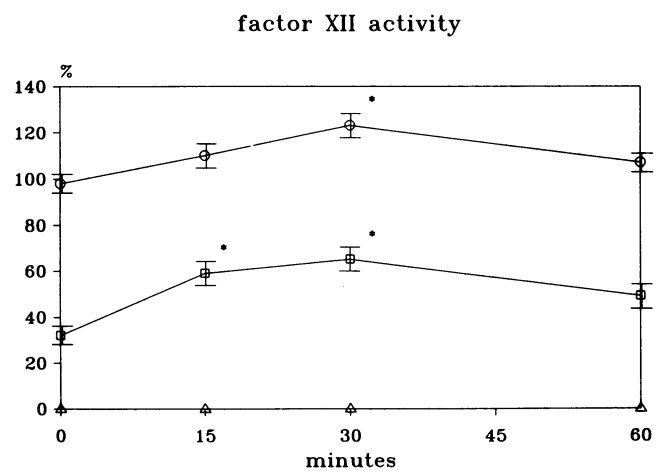

Figure 4. Functional levels of factor XII after DDAVP infusion. Time scale indicates minutes after the start of the DDAVP infusion. Data represent mean values, bars indicate SEM. Results of normal individuals (○, n = 6), subjects with mild (□, n = 3) and severe factor XII deficiency (△, n = 3) are given. Significance compared to preinfusion levels is indicated (*P < 0.05, ANOVA and Newman-Keuls' test).

healthy volunteers. There was no significant difference in the inhibition of plasminogen activating activity by either anti-t-PA or anti-u-PA antibodies between the healthy subjects and the factor XII deficient subjects. However, the residual plasminogen activating activity observed after the addition of both anti-t-PA and anti-u-PA antibodies was significantly reduced in plasma of patients with mild factor XII deficiency (14%, SEM 1.2, $P < 0.05$ Student's t test) compared to that in healthy volunteers (23%, SEM 2.1) and was completely absent in patients with severe factor XII deficiency (0%, SEM 0.3, $P < 0.05$).

Induction of PAP complexes by DDAVP in healthy subjects and in patients with factor XII deficiency. The increase in plasminogen activating activity resulted in the formation of plasmin as indicated by the increase in plasma levels of PAP complexes upon the infusion of DDAVP. Response of PAP complexes in plasma upon DDAVP in healthy subjects and factor XII deficient individuals is shown in Fig. 3. PAP complexes increased from 5.2 nM (SEM 0.7) to 28.3 nM (SEM 2.3) after DDAVP infusion in healthy volunteers. In patients with severe factor XII deficiency this increase in PAP complexes upon DDAVP was significantly reduced (preinfusion level 4.7 nM [SEM 1.0] vs. postinfusion level 19.7 nM [SEM 1.8], $P < 0.05$ compared to normal subjects, Student's t test). The level of PAP complexes in plasma after DDAVP in patients with mild deficiency of factor XII was between that observed in patients with severe deficiency of factor XII and that seen in healthy subjects (Fig. 3, preinfusion level 4.9 nM [SEM 1.1] vs. postinfusion level 26.6 nM [SEM 1.7]).

Activation of the contact system induced by DDAVP. PAP complexes were significantly lower in the patients with severe factor XII deficiency upon administration of DDAVP compared to normal controls. In addition, DDAVP generated a factor XII dependent plasminogen activating activity in healthy subjects. These results might point to contact activation and subsequent additional plasmin generation induced by DDAVP. We therefore decided to study activation of the contact system during DDAVP infusion.

The effect of the infusion of DDAVP in healthy subjects and factor XII deficient patients on factor XII activity is shown in Fig. 4. A small but significant increase in factor XII activity was observed in all healthy subjects peaking at 30 min after the start of the DDAVP infusion (mean preinfusion level 98%, SEM 2.8, mean peak level 121%, SEM 3.0, $P < 0.05$ analysis of variance [ANOVA] and Newman-Keuls' test) and subsequently decreased. A similar increase was observed in the patients with a mild factor XII deficiency. Factor XII activity levels in these patients increased from 31% (SEM 6.4) to 61% (SEM 4.0). Factor XII activity was not detected at any stage of the study in patients with a severe factor XII deficiency indicating that the increases observed in the other groups were indeed due to factor XII. In contrast to functional levels, factor XII antigen levels remained stable during the entire study period both in healthy subjects and in factor XII deficient patients (Table II).

Table II. Factor XII and Prekallikrein Antigen upon DDAVP Infusion

	Normal subjects		Factor XII < 50%		Factor XII < 1%	
	Before DDAVP	After DDAVP	Before DDAVP	After DDAVP	Before DDAVP	After DDAVP
Factor XII antigen	97 (2.0)	98 (2.4)	34 (7.0)	33 (6.6)	0 (0.1)	0 (0.2)
Prekallikrein antigen	101 (0.7)	100 (0.9)	98 (1.1)	96 (1.3)	104 (1.3)	101 (1.4)

Factor XII antigen (%) and prekallikrein antigen (%) in normal subjects ($n = 6$), patients with mild ($n = 3$), and patients with severe factor XII deficiency ($n = 3$) before and 15 min after the start of an infusion with DDAVP. Mean values (and SEM) are given. There were no significant differences between pre and postinfusion values in the three study groups.

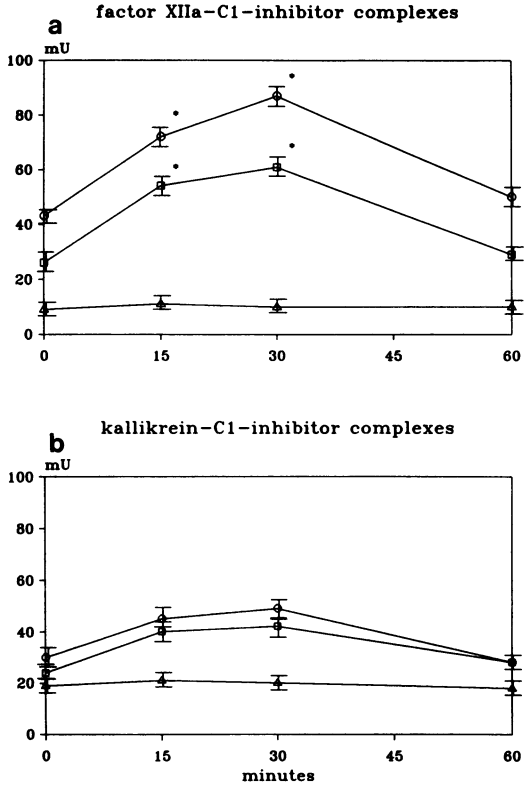

Figure 5. Activation of the contact system upon intravenous administration of DDAVP to normal individuals (○, $n = 6$) and to patients with mild (□, $n = 3$) and severe (△, $n = 3$) factor XII deficiency. *Upper panel:* factor XIIa-C1-inhibitor complexes. *Lower panel:* kallikrein-C1-inhibitor complexes. Data represent mean values, bars indicate SEM. On x axis time (in minutes) after start of the DDAVP infusion is given. Significance compared to preinfusion levels is indicated (*$P < 0.05$, ANOVA and Newman-Keuls' test).

Results of the infusion of DDAVP on factor XIIa-C1-inhibitor complexes are shown in Fig. 5 *a*. An increase in plasma levels of factor XIIa-C1-inhibitor complexes was observed in all healthy subjects and in patients with a mild factor XII deficiency. Mean preinfusion plasma level of factor XIIa-C1-inhibitor complexes in healthy volunteers was 43 mU (SEM 5.1) and increased upon infusion of DDAVP to a mean maximal level of 87 mU (SEM 8.3, $P < 0.05$, ANOVA and Newman-Keuls' test) at 30 min after the start of the infusion. Patients with a mild deficiency of factor XII had lower preinfusion levels (mean 26 mU, SEM 4.7) which increased to 61 mU (SEM 7.0, $P < 0.05$, ANOVA and Newman-Keuls' test) at 30 min. In the patients with severe factor XII deficiency no factor XIIa-C1-in-

hibitor complexes were detected in any of the blood samples studied (i.e., levels were < 10 mU/ml).

Mean kallikrein-C1-inhibitor complex levels increased from 30 mU (SEM 4.6) to 49 mU (SEM 7.1) at 30 min in normal subjects and from 24 mU (SEM 5.3) to 42 mU (SEM 7.3) at 30 min in patients with a mild factor XII deficiency (Fig. 5 *b*). Both increases were not statistically significant. Plasma levels of kallikrein-C1-inhibitor complexes in subjects with severe factor XII deficiency were the lowest compared to the other groups and they remained stable during the DDAVP infusion. Plasma levels of prekallikrein showed no change during the study in all groups (Table II).

Discussion

Deficiency of factor XII results in a prolongation of coagulation tests in vitro, which may suggest the association with a bleeding tendency in affected individuals. However, in vivo these individuals rather seem to have a higher incidence of thromboembolism (9, 10), as was illustrated by the clinical course of the index case, Mr. Hageman, who died of pulmonary embolism (18, 19). The data presented here show that activation of the contact system contributes to plasminogen activation, induced in vivo by intravenous administration of DDAVP. Therefore, we propose that the thromboembolic events that are observed in factor XII deficient individuals are due to impaired fibrinolysis.

Activation of the contact system by DDAVP as demonstrated by an increase in factor XIIa-C1-inhibitor complexes presumably was responsible for the observed non-t-PA and non-u-PA related plasminogen activator activity in post-DDAVP plasma for two reasons. First, this residual plasminogen activator activity could be quenched with a monoclonal antibody that specifically blocks factor XII activity. Second, this non-t-PA and non-u-PA dependent plasminogen activator activity was reduced in patients with half-normal levels of factor XII and completely absent in patients with no detectable factor XII. The relevance of this factor XII dependent plasminogen activator activity in vivo was demonstrated by the observation that activation of plasminogen in vivo as reflected by the increase in PAP complexes was significantly reduced in severely factor XII deficient patients compared to healthy controls. This difference between PAP complex levels upon DDAVP was not due to a diminished t-PA or u-PA increase in the factor XII deficient patients (Table I). Therefore, these findings imply that activation of the contact system contribute to activation of the fibrinolytic system in vivo.

Earlier observations revealed the absence of prothrombin activation (no generation of prothrombin fragment F_{1+2}) and fibrinogen conversion to fibrin (no generation of fibrinopeptide A) in vivo upon administration of DDAVP, suggesting that the activation of the contact system observed in this situation does not result in significant activation as previously described (Levi, M., et al., manuscript submitted). Hence, these results support the hypothesis that the function of the contact system in vivo is a profibrinolytic rather than a procoagulant one.

Several studies have shown an increase in factor XII activity upon DDAVP (20, 21) with the exception of one report (22). In our study we also observed an increase in the factor XII activity upon the administration of DDAVP to normal subjects; however, the total factor XII antigen level remained stable. In addition, we observed significant increases in plasma levels of factor XIIa-C1-inhibitor complexes, indicating factor XII activation in vivo (17, 23). Presumably, the increase in factor XII activity noted after DDAVP was due to the fact that activated factor XII is more easily detected in the functional assay than native factor XII. Thus, DDAVP induced activation of native factor XII already present in the circulation rather than an increase in availability of factor XII. The mechanism of this activation induced by DDAVP is not fully understood. It has been shown that trace amounts of plasmin can activate factor XII (24, 25). Therefore, plasmin formation by the t-PA and u-PA release upon infusion of DDAVP may initiate contact system activation which then itself may further enhance fibrinolytic activation.

Several mechanisms by which the contact system may induce plasminogen activation can be envisaged. Active factor XII is able to directly activate plasminogen (26). However, at plasma concentrations the direct effect of factor XII on the activation of plasminogen is presumed to be very small (6, 27). Since, however, plasma kallikrein and factor XIa also are able to directly convert plasminogen into plasmin (7, 27), a combined effect of factor XIIa, kallikrein, and factor XIa may potentially cause contact activation dependent fibrinolytic activity. Other studies, such as those in which dextran sulphate–activated euglobulins are studied, have shown that kallikrein is able to cleave and thereby activate single chain u-PA (28). It has been proposed that via this mechanism the contact system contributes to plasminogen activator activity. However, such a mechanism cannot explain the contact system mediated fibrinolytic activation that we describe here, since the factor XII dependent fibrinolytic activity was still observed after specific inhibition of u-PA. Thus, the contact system dependent plasminogen activator activity as observed in this study apparently occurred via a factor XII dependent pathway that was independent from u-PA. The molecular basis for this pathway remains to be established, although several studies have proposed the involvement of a yet unidentified plasminogen activator that is dependent on factor XII (6, 29).

Activation of the contact system results in the release of bradykinin from high molecular weight kininogen (7). Bradykinin potentially may affect fibrinolytic activation by inducing plasminogen activator release from the vessel wall (30, 31). However, it is unlikely that this mechanism significantly contributed to the activation of the fibrinolytic system since patients with severe factor XII deficiency had no detectable contact system activation upon DDAVP (Fig. 5), whereas they showed identical t-PA and u-PA release compared to healthy subjects.

In conclusion, infusion of DDAVP induces plasminogen activator activity that is related to activation of the contact system and, in particular, factor XII, and that occurs in addition to an increase in t-PA and u-PA activity. Therefore, we suggest that the contact system should be considered as a fibrinolytic rather than a blood coagulation system and that the association between thromboembolic disease and deficiencies of the contact system are due to an impaired fibrinolytic capacity in these patients.

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