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# Different antithrombotic properties of factor Xa inhibitor and thrombin inhibitor in rat thrombosis models

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**Abstract**

We compared the antithrombotic properties of a factor Xa inhibitor (DX-9065a) with those of a thrombin inhibitor (melagatran) in a rat disseminated intravascular coagulation model and a rat venous thrombosis model. Rat disseminated intravascular coagulation and venous thrombosis models were produced by injection of tissue factor and platinum wire placement, respectively. DX-9065a exerted antithrombotic effects dose dependently in both models. Melagatran was also effective in the venous thrombosis model, whereas it showed an aggravation in the disseminated intravascular coagulation model at low but not high doses. In the in vitro study, DX-9065a decreased the $C_{max}$ of the thrombin generation curve in plasma irrespective of whether protein C was present or not. However, melagatran increased the $C_{max}$ at low concentrations when protein C was present. This increase was not detected in protein C-deficient plasma. These results suggest that, unlike DX-9065a, melagatran in low doses aggravates disseminated intravascular coagulation by increasing thrombin generation, which may be partly due to suppression of negative feedback by activated protein C.


*Keywords:* Disseminated intravascular coagulation model; Venous thrombosis model; Factor Xa inhibitor; Thrombin inhibitor; Protein C; Thrombin generation

## 1. Introduction

Heparin, an antithrombin dependent inhibitor, and warfarin, an oral vitamin K antagonist, are widely used in clinical practice. Variations in the efficacy and safety of these drugs (Chong, 2003; Hirsh et al., 1998) have lead to new approaches for the prevention of thrombosis, such as specific inhibitors of factor Xa and thrombin. Since factor Xa and thrombin play a central role in the blood coagulation cascade, these are attractive targets for antithrombotic therapy.

Several factor Xa inhibitors and thrombin inhibitors are currently under clinical trial. DX-9065a, a direct factor Xa inhibitor, potently and selectively inhibits factor Xa with a $K_i$ value of 41 nM (Hara et al., 1994). Melagatran, a direct thrombin inhibitor, potently and selectively inhibits thrombin with a $K_i$ value of 2 nM (Gustafsson et al., 1998). Both inhibitors have been shown to exert anticoagulant effects in several animal models of experimental thrombosis (Morishima et al., 1997; Yamashita et al., 1997; Eriksson et al., 1997; Mehta et al., 1998). DX-9065a and ximelagatran, a prodrug of melagatran, are currently under clinical development.

In addition to factor Xa and thrombin, protein C is also an important factor that regulates the coagulation pathway (Esmon, 2001; Dahlback and Villoutreix, 2003). Activated protein C has potent anticoagulant activity due to its ability to inactivate factors, such as Va and VIIIa. It exhibits antithrombotic and anti-inflammatory properties in several animal models (Griffin et al., 2002; Malm et al., 2003) and clinical studies (Bernard et al., 2001; Joyce et al., 2001). Protein C is activated by the action of the

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thrombin-thrombomodulin complex, and melagatran inhibits the activation of protein C through the inhibition of free and thrombomodulin-bound thrombins (Mattsson et al., 2001). In contrast, it is unlikely that a factor Xa inhibitor would inhibit protein C activation directly, which stimulated our interest to compare the antithrombotic properties of a factor Xa inhibitor and a thrombin inhibitor.

In the present study, we evaluated the differences in antithrombotic properties between DX-9065a and melagatran in a tissue factor-induced rat disseminated intravascular coagulation model and in a platinum wire-induced rat venous thrombosis model. Thrombin generation in the presence of these compounds was also evaluated.

## 2. Materials and methods

### 2.1. Materials

Tissue factor (Thromboplastin C Plus) and Enzygnost® TAT micro were purchased from Dade Behring (Liederbach, Germany). Ravonal® (thiopental sodium) was purchased from Tanabe Seiyaku (Osaka, Japan). Platinum wire (0.5 mm in diameter, 2.5 cm long) was bought from Sanwa Kinzoku (Saitama, Japan). Glutaraldehyde was bought from Polysciences, Inc. (Warrington, PA, USA). Human protein C was purchased from Calbiochem (La Jolla, CA, USA). Protein C-deficient human plasma was purchased from George King Bio-Medical, Inc. (Overland Park, KS, USA). Fluorogenic thrombin substrate Z-Gly-Gly-Arg-aminomethylcoumarin (Z-GGR-AMC) was bought from Bachem AG (Bubendorf, Switzerland). Anti-rat protein C antibody and recombinant human thrombomodulin were gifts from Dr Katsuhiko Nawa (Proteome Research Laboratory, Daiichi Pharmaceutical Co., LTD, Tochigi, Japan). DX-9065a, melagatran and ximelagatran were synthesized in our laboratory.

### 2.2. Animals

Nine-week-old male Wistar rats were obtained from Japan SLC (Hamamatsu, Japan). They were acclimated for 3 weeks. The animals were fed on a commercial diet (F-2, Funabashi Farms, Funabashi, Japan) and tap water was given ad libitum. All experimental procedures were performed in accordance with the in-house guidelines of the Institutional Animal Care and Use Committee of Daiichi Pharmaceutical Co., Ltd.

### 2.3. Tissue factor-induced rat disseminated intravascular coagulation model

#### 2.3.1. A Time course study

DX-9065a or melagatran was given orally to fasted rats 30 min prior to tissue factor infusion. The rats were anesthetized with thiopental sodium (Ravonal®, 100 mg/kg, i.p.). Tissue factor (0.8 U/ml) or saline was infused into the femoral vein for 1 min at a volume of 3.5 ml/kg/min. Blood was collected from the inferior vena cava at various time points (0, 3, 10, 30, 60 min) and the platelet count was measured.

#### 2.3.2. Dose–response effects

Ten minutes after tissue factor infusion, blood was collected from the inferior vena cava and the platelet count was measured. Then, the blood samples were centrifuged at $1500 \times g$ for 10 min at 4 °C. Plasma samples were stored at − 70 °C until the measurement of the concentration of thrombin anti-thrombin complex. Thrombin anti-thrombin complex concentration in rat plasma was measured using EIA kit (Enzygnost® TAT micro).

### 2.4. Rat venous thrombosis model

Rat venous thrombosis was induced with a platinum wire according to the method of Lavelle and Iomhair (1997) with a slight modification. The platinum wire, 0.5 mm in diameter and 2.5-cm long, was sharpened at one tip and bent at 60° angle 2 cm from that tip. DX-9065a or melagatran was given orally to the fasted rat. Thirty minutes after the administration of the inhibitors, the 2-cm-long platinum wire was inserted into the inferior vena cava below the renal vessel in a rat anesthetized with Ravonal® (100 mg/kg, i.p.) and left for 60 min. Then, 1 ml of glutaraldehyde in 10 mM phosphate-buffered saline (PBS, pH 7.4) was injected into the lower inferior vena cava to fix the thrombus in situ. The wire was dissected free, and the weight of the thrombus with wire was noted. Each wire was wiped free of thrombus and re-weighed to obtain the thrombus weight.

### 2.5. Phosphotungstic acid hematoxylin staining

Phosphotungstic acid hematoxylin staining was performed on tissue from all animals in the groups given 100 mg/kg of DX-9065a or 2.5 mg/kg of melagatran. Rats were sacrificed 40 min after administration of DX-9065a or melagatran in the disseminated intravascular coagulation model. The lung, liver and kidney were rapidly removed and fixed with 10% buffered formalin. Then they were embedded into paraffin, and the paraffin sections were stained with phosphotungstic acid hematoxylin.

### 2.6. Measurement of thrombin generation

Measurement of thrombin generation was performed with a 96-well microtiter plate according to the method of Hemker et al. (2000). In this assay, the following types of plasma were used: protein C-deficient human plasma, human protein C-supplemented protein C-deficient human plasma (the concentration of protein C was 65 nM), rat

normal plasma and protein C-decreased rat plasma which was prepared by the adsorption of protein C from rat normal plasma with anti-rat protein C antibody-conjugated beads. Fifty microliters of inhibitor was added to 50 μl of plasma. Then, 40 μl of fluorogenic thrombin substrate Z-GGR-AMC with phospholipid (phosphatidylcoline:phosphatidylserine=3:1) was added. After pre-incubation for 3 min at 37 °C, 60 μl of tissue factor with recombinant human thrombomodulin and $CaCl_2$ was added to trigger the coagulation reaction. The final concentrations of Z-GGR-AMC, phosphatidylcoline: phosphatidylserine, tissue factor, recombinant human thrombomodulin and $CaCl_2$ were 680 μM, 50 μM, 18 μU/ml, 10 nM, and 16.7 mM, respectively. The fluorescence was measured every 45 sec for 90 min at 37 °C (ex 390 nm, em 460 nm). The concentration of thrombin was calculated by comparison with a standard curve for human α-thrombin. The results represent the $C_{max}$ of the thrombin generation curve.

### 2.7. Statistical analysis

All data represent means±S.E.M.. The statistical significance of the data was analyzed by *t*-test and Dunnett's multiple comparison method. The statistical significance of the kinetic profile was analyzed by repeated-measures analysis of variance (ANOVA).

## 3. Results

### 3.1. Antithrombotic properties of DX-9065a or melagatran in a tissue factor-induced rat disseminated intravascular coagulation model

The antithrombotic properties of DX-9065a or melagatran in a tissue factor-induced rat disseminated intravascular coagulation model were evaluated. When tissue factor was infused into the rat femoral vein, it significantly reduced the platelet count from $63.7\pm1.65\times10^4$ to $17.5\pm3.91\times10^4$ platelets/μl ($P<0.001$) during the first 3 min. Then, the platelet count gradually increased but did not recover to basal levels. DX-9065a significantly inhibited the reduction in platelet count at a dose of 100 mg/kg. Melagatran also significantly inhibited the reduction in platelet count at a dose of 40 mg/kg. In contrast, a low dose of melagatran (0.5 mg/kg) reduced the platelet count more than the control until 60 min after the infusion of tissue factor (Fig. 1).

The effects of DX-9065a or melagatran on the platelet count and the concentration of thrombin anti-thrombin complex 10 min after tissue factor infusion in a rat disseminated intravascular coagulation model are shown in Fig. 2. When tissue factor was infused into the rat femoral vein, it significantly decreased the platelet count from $51.6\pm0.55\times10^4$ to $37.9\pm1.4\times10^4$ platelets/μl ($P<0.001$) (Fig. 2A) and significantly increased the concentration of thrombin anti-thrombin complex in rat plasma to 517±31 ng/ml compared to that in the sham group (2.8±0.91 ng/ml, $P<0.01$) (Fig. 2B). DX-9065a dose dependently inhibited the consumption of platelets by 68% and inhibited the increase in thrombin anti-thrombin complex concentration by 57% at a dose of 100 mg/kg in the rat disseminated intravascular coagulation model (Fig. 2).

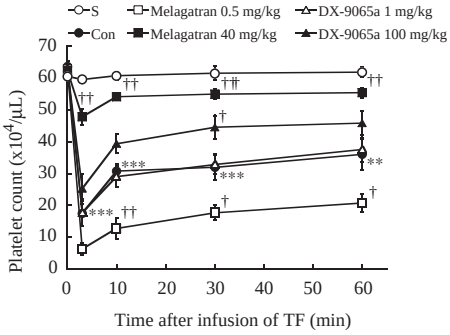


Fig. 1. Time course of changes in platelet count in a tissue factor-induced rat disseminated intravascular coagulation model. Data represent means±S.E.M. ($n=4$ for all groups). The statistical significance of the kinetic profile was analyzed by a repeated-measures analysis of variance (ANOVA). ***$P<0.001$, **$P<0.01$ vs. sham (S) group (*t*-test). †††$P<0.001$, ††$P<0.01$, †$P<0.05$ vs. control (Con) group (Dunnett's test).

In contracts, melagatran aggravated the disseminated intravascular coagulation markers at doses of 0.5 and 2.5 mg/kg. Melagatran significantly enhanced the decrease in the platelet count from $41.2\pm1.48\times10^4$ to $14.5\pm1.42\times10^4$ ($P<0.001$) and to $9.8\pm0.67\times10^4$ platelets/μl ($P<0.001$) at doses of 0.5 and 2.5 mg/kg, respectively (Fig. 2C). Fig 2D represents the effect of melagatran on the concentration of thrombin anti-thrombin complex in rat plasma. Melagatran significantly increased the plasma thrombin anti-thrombin complex concentration from 298±49 to 1147±90 ($P<0.001$) and to 2558±91 ng/ml ($P<0.001$) at doses of 0.5 and 2.5 mg/kg, respectively.

To confirm whether the effect of melagatran at 2.5 mg/kg was accompanied by fibrin deposition, histological studies were performed in the lung, liver and kidney. Fibrin thrombi were found in the lung in the melagatran-treated group at a dose of 2.5 mg/kg (Fig. 3B), but not detected in the control group and the DX-9065a-treated group at a dose of 100 mg/kg (Fig. 3A and C). Fibrin thrombi were not detected in the liver or kidney of any groups.

### 3.2. Antithrombotic effects of DX-9065a or melagatran in a rat venous thrombosis model

The antithrombotic properties of DX-9065a or melagatran in a rat venous thrombosis model were evaluated. Placement of a platinum wire in the inferior vena cava induced thrombus formation. In the control group the thrombus weight was 2.24±0.35 mg (Fig. 4A) or 2.56±0.56 mg (Fig. 4B). DX-9065a significantly reduced it to 0.39±0.12 mg ($P<0.01$) at a dose of 100 mg/kg (Fig. 4A). Melagatran also significantly reduced the thrombus

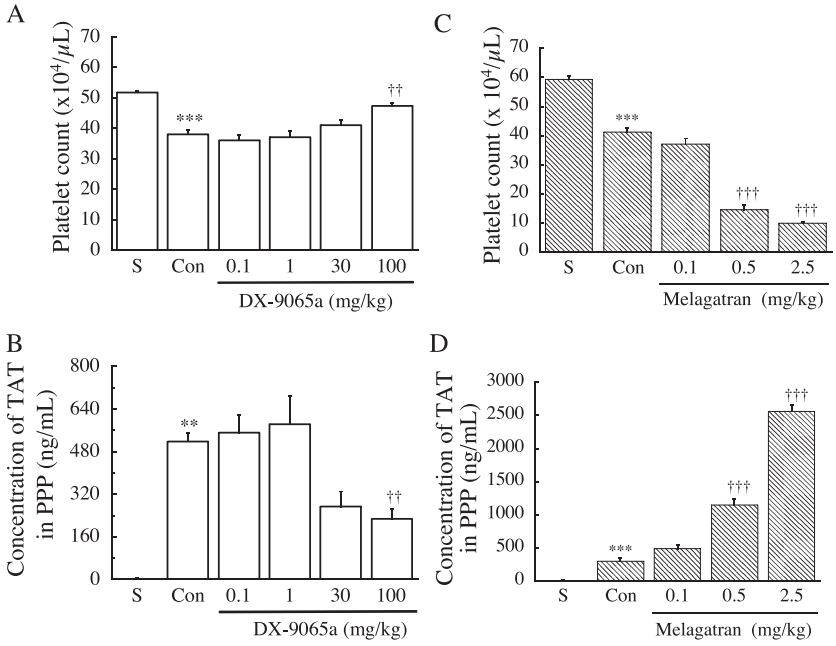


Fig. 2. Effects of DX-9065a (A, B) or melagatran (C, D) on the platelet count and the concentration of thrombin anti-thrombin complex 10 min after tissue factor infusion in a rat disseminated intravascular coagulation model. Data represent means±S.E.M. ($n=6$ for DX-9065a treatment groups, $n=5–6$ for melagatran treatment groups). ***$P<0.001$, **$P<0.01$ vs. sham (S) group ($t$-test). †††$P<0.001$, ††$P<0.01$, †$P<0.05$ vs. control (Con) group (Dunnett's test).

weight to 0.10±0.03 mg ($P<0.001$) at a dose of 20 mg/kg (Fig. 4B). Both inhibitors attenuated thrombus formation in a dose dependent manner.

### 3.3. Comparison of the antithrombotic effects of the inhibitors in a disseminated intravascular coagulation model and a venous thrombosis model

The antithrombotic effects of DX-9065a and melagatran in the two rat thrombosis models are shown in Fig. 5. The antithrombotic effect is expressed as the percent inhibition of platelet consumption in the disseminated intravascular coagulation model or percent inhibition of thrombus formation in the venous thrombosis model. DX-9065a demonstrated dose-dependent antithrombotic effects in both rat thrombosis models (Fig. 5A). Melagatran also showed dose-dependent antithrombotic effects in the rat venous thrombosis model but in the disseminated intravascular coagulation model, melagatran produced contrasting dual responses, increasing platelet consumption at low-dose (0.5, 2.5 mg/kg) and attenuating the increase at a higher dose (20 mg/kg) (Fig. 5B).

### 3.4. Effects of DX-9065a or melagatran on thrombin generation

Fig. 6 represents the effects of DX-9065a or melagatran on thrombin generation in plasma. In human plasma, the $C_{max}$ was significantly decreased when protein C was added to the protein C-deficient plasma in the absence of compounds. DX-9065a decreased the $C_{max}$ both in protein C-deficient and protein C-supplemented protein C-deficient plasma (Fig. 6A). Melagatran also decreased it in protein C-deficient plasma. However, in protein C-supplemented protein C-deficient plasma, $C_{max}$ was increased from 380.7±5.6 to 460.9±7.0 ($P<0.001$) and to 451.9±8.1 ($P<0.001$) nM by melagatran at concentrations of 5 and 10 nM, respectively. This increase was not detected in the protein C-deficient plasma (Fig. 6B). In rat normal plasma, melagatran also increased the $C_{max}$ of the thrombin

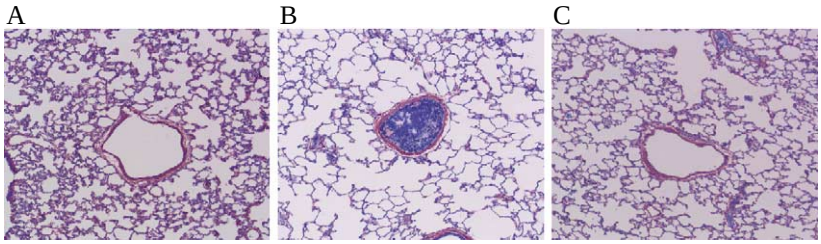


Fig. 3. Microphotographs of lungs in a rat disseminated intravascular coagulation model. (A) Control, (B) melagatran 2.5 mg/kg, (C) DX-9065a 100 mg/kg. Fibrin thrombi were shown in B by phosphotungstic acid hematoxylin stain. (Magnification of panels A, B and C: 20×).

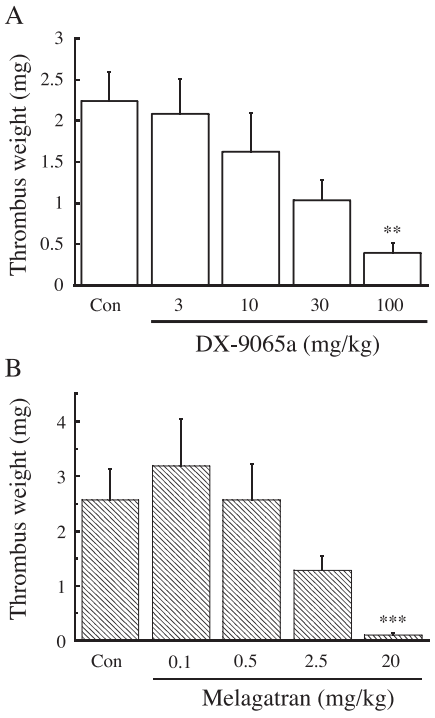


Fig. 4. Effects of DX-9065a (A) or melagatran (B) on the thrombus weight in a rat venous thrombosis model. Data represent means±S.E.M. ($n=6–7$ for all groups). ***$P<0.001$, **$P<0.01$ vs. control (Con) group (Dunnett's test).

generation curve from 35.7±3.5 to 63.3±1.8 ($P<0.01$) and to 62.9±6.9 ($P<0.01$) nM at 2 and 10 nM, respectively, but not in protein C-decreased rat plasma (Fig. 6D). In contracts, DX-9065a decreased it both in normal and protein C-deficient rat plasma (Fig. 6C).

The enhancement of endogenous thrombin potential in the presence of protein C was not detected at the low dose of melagatran (data not shown).

## 4. Discussion

The antithrombotic properties of a factor Xa inhibitor (DX-9065a) were compared with those of a thrombin inhibitor (melagatran) in two different rat thrombosis models, a tissue factor-induced disseminated intravascular coagulation model and a venous thrombosis model. Although melagatran, an active form of ximelagatran, has low oral bioavailability (approximately 5%) compared to ximelagatran (approximately 20%) in humans (Eriksson et al., 2003), both forms were well absorbed in rats. Therefore melagatran was given orally to rats in this study.

DX-9065a dose dependently attenuated thrombus formation in the venous thrombosis model and reduced the consumption of platelets and the thrombin anti-thrombin complex concentration in plasma in the disseminated intravascular coagulation model. Melagatran also exerted a dose-dependent antithrombotic effect in the venous thrombosis model. However, in a disseminated intravascular coagulation model melagatran enhanced the consumption of platelets and increased the plasma thrombin anti-thrombin complex concentration at low doses of 0.5 and 2.5 mg/kg. This enhancement was accompanied by fibrin deposition in the lung. Ximelagatran showed the same results (data not shown). These data suggest that melagatran enhanced thrombin generation by affecting the regulation of blood coagulation.

Protein C is one of the important factors that regulate the coagulation pathway. Activated protein C has potent anti-coagulant activity and exhibits antithrombotic properties in several animal models (Griffin et al., 2002; Malm et al., 2003) and clinical studies (Bernard et al., 2001; Joyce et al., 2001). Drotrecogin alfa (recombinant human activated protein C) reduced mortality (Bernard et al., 2001) and improved disseminated intravascular coagulation symptoms (Joyce et al., 2001) in patients with severe sepsis. Therefore, we tested the effects of DX-9065a as well as melagatran on thrombin generation in protein C-deficient human plasma or human protein C-supplemented protein C-deficient human plasma. DX-9065a decreased thrombin generation in both the protein C-deficient plasma and protein C-supplemented protein C-deficient plasma. In contrast, melagatran enhanced thrombin generation at low concentrations (5 and 10 nM) in protein C-supplemented protein C-deficient

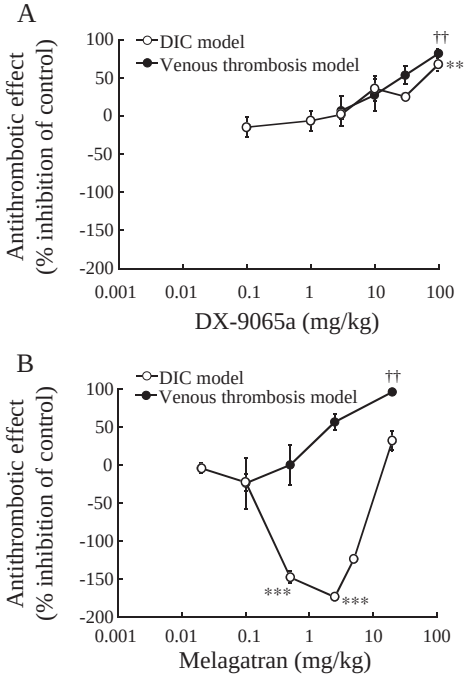


Fig. 5. Comparison of antithrombotic effects of the inhibitors (DX-9065a (A), melagatran (B)) in a disseminated intravascular coagulation model and a venous thrombosis model. The *Y*-axis shows antithrombotic effect (percent inhibition of control). The antithrombotic effects represent percent inhibition of platelet consumption in a disseminated intravascular coagulation model or percent inhibition of thrombus formation in a venous thrombosis model. The *X*-axis expresses logarithmic doses of inhibitors. Data represent means±S.E.M. ($n=6–8$ for all groups). ***$P<0.001$, **$P<0.01$, ††$P<0.01$ vs. control group (Dunnett's test).

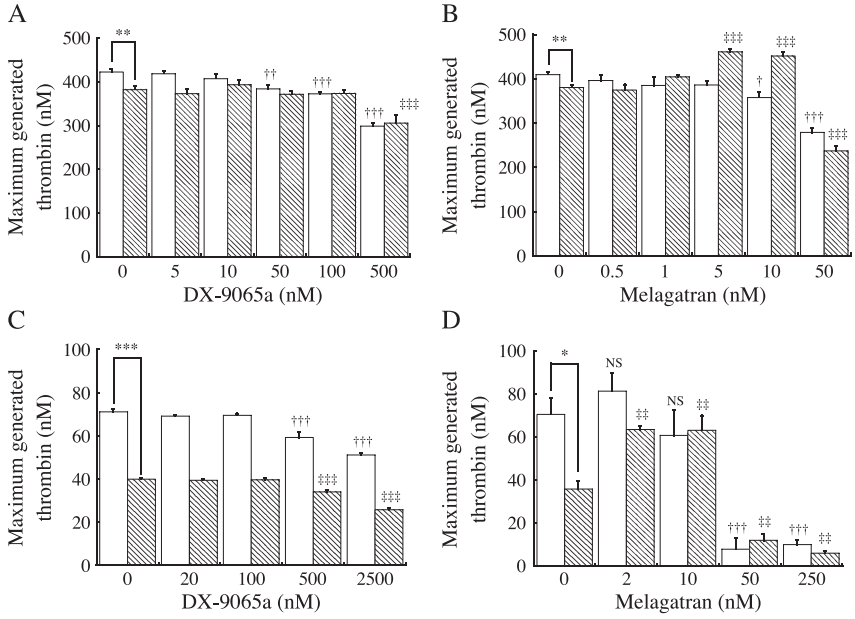


Fig. 6. Effect of DX-9065a or melagatran on thrombin generation in plasma. Thrombin generation was measured in the presence of DX-9065a (A, C) or melagatran (B, D). (A) and (B) show the effect of the inhibitors on thrombin generation in protein C-deficient human plasma (open bars) or protein C-supplemented protein C-deficient human plasma (hatched bars). (C) and (D) shows the effect of the inhibitors on thrombin generation in protein C decreased rat plasma (open bars) or rat normal plasma (hatched bars). Data represent means±S.E.M. ($n$=6 for all groups in human plasma, $n$=3–4 for all groups in rat plasma). ***$P$<0.001, **$P$<0.01, *$P$<0.05 vs. control (protein C-deficient or protein C-decreased plasma without inhibitor). †††$P$<0.001, ††$P$<0.01, †$P$<0.05 vs. control (protein C-deficient or protein C-decreased plasma without inhibitor). ‡‡‡$P$<0.001, ‡‡$P$<0.01 vs. control (protein C-supplemented protein C-deficient or normal plasma without inhibitor).

plasma. It also enhanced thrombin generation in rat plasma at concentrations of 2 and 10 nM but not in protein C-deficient human plasma or protein C-decreased rat plasma. These results suggest that the thrombin inhibitor has the potential to stimulate thrombin generation through the attenuation of the protein C pathway. This view is supported by the following reports: (1) argatroban, another direct thrombin inhibitor, enhanced thrombin generation in human plasma (Mohri et al., 1998; Mohri et al., 1999), (2) melagatran inhibited the thrombin-thrombomodulin-mediated generation of activated protein C on endothelial cells (Linder et al., 2003).

Melagatran did not increase platelet consumption at a higher dose (20 mg/kg) in the disseminated intravascular coagulation model, and it decreased thrombin generation at a higher concentration (50 nM) in spite of the expectation of its stronger protein C pathway inhibition. Our data suggested that the higher concentration of melagatran was enough to inhibit thrombin activity, the generation of which was enhanced by inhibition of the protein C pathway. Another explanation is that the higher concentration of melagatran suppressed thrombin generation through the inhibition of feedback activation of factor V, factor VII, factor VIII and factor XI, which were converted to active forms by thrombin. These feedback reactions by thrombin play an important role in thrombin generation (He et al., 2001), and the inhibition of these feedback reactions by the high concentration of melagatran may have more effect on thrombin generation than activated protein C. In contrast, a low concentration of melagatran insufficiently blocks the feedback activation of coagulation factors and then the protein C pathway may have a dominant role in the regulation of thrombin generation.

In our study, melagatran increased the $C_{max}$ of the thrombin generation curve, indicating that it enhanced thrombin generation in the propagation phase. However, the enhancement of endogenous thrombin potential by melagatran was not detected. Mohri et al. (1999) reported that argatroban, a direct thrombin inhibitor, increased not only the $\boldsymbol{C}_{max}$ of the thrombin generation curve, but also the endogenous thrombin potential, using defibrinated human plasma. These data indicate the possibility that direct thrombin inhibitors induce thrombosis through an enhancement of thrombin generation under some conditions.

The $\boldsymbol{C}_{max}$ of the thrombin generation curve in our experiment is an approximate value because of non-linearity between thrombin activity and fluorescent signal (Hemker et al., 2003). However, it is possible to compare the values obtained at different concentrations of the compound. This problem was solved by monitoring the fluorescence signal and comparing it with a constant known thrombin activity in a parallel non-clotting sample (Hemker et al., 2003). This method is useful for the quantification of thrombin generation.

The plasma concentration of melagatran was estimated from the thrombin time, using treated rat plasma. The concentration of melagatran at a dose of 0.5 mg/kg was 10 nM (data not shown), which was comparable to that for the enhancing effect on thrombin generation in vitro. Melagatran

showed an antithrombotic tendency at a dose of 20 mg/kg and the plasma concentration was 3.6 μM (data not shown). Eriksson et al. (2000, 1998) reported that melagatran inhibited the consumption of platelets and the reduction in the fibrinogen concentration in plasma in the endotoxin-induced pig model. In these studies, the plasma concentrations of melagatran ranged from 450 to 850 nM, which were much higher than the concentration of melagatran causing aggravation of disseminated intravascular coagulation markers in our study. Further studies may be required to clarify the effects of melagatran in endotoxin-induced models.

Since direct thrombin inhibitors inhibit clot-bound thrombin (Finkle et al., 1998; Gast et al., 1994; Weitz et al., 1990), these are expected to have an advantage as anticoagulants compared to antithrombin-dependent inhibitors such as heparin. However most clinical trials in patients with coronary artery disease failed to show a clear advantage of thrombin inhibitors over heparin (The GUSTO IIb investigators, 1996; OASIS-2 investigators, 1999; Thrombin inhibition in Myocardial Ischaemia (TRIM) study group, 1997; Klootwijk et al., 1999), except for a few studies (Organization to Assess Strategies for Ischemic Syndromes (OASIS) Investigators, 1997). Furthermore, no clear dose-response relationship has been observed in these clinical studies. In a TRIM (thrombin inhibition in myocardial ischaemia) study, inogatran, a direct thrombin inhibitor, increased the number of ischemic events more than heparin did (Thrombin inhibition in Myocardial Ischaemia (TRIM) study group, 1997). These unexpected results might be attributed to the attenuation of protein C pathway.

In conclusion, DX-9065a demonstrated a significant dose-response relationship for its antithrombotic effects in both rat thrombosis models, whereas low-dose melagatran increased the consumption of platelets and increased the thrombin anti-thrombin complex concentration in a disseminated intravascular coagulation model. The aggravation of disseminated intravascular coagulation was accompanied by an increase in thrombin generation, which may be partly due to suppression of negative feedback of activated protein C by melagatran. Therefore, direct factor Xa inhibitors may be preferable in the treatment of thrombosis.

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