

The Activated Clotting Time Can Be Used to Monitor the Low Molecular Weight Heparin Dalteparin After Intravenous Administration

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OBJECTIVES	This study was designed to compare the dose response of dalteparin versus unfractionated heparin (UFH) on the activated clotting time (ACT), and to determine whether the ACT can be used to monitor intravenous (IV) dalteparin during percutaneous coronary intervention (PCI).
BACKGROUND	The use of low molecular weight heparin (LMWH) during PCI has been limited by the presumed inability to monitor its anticoagulant effect using bedside assays.
METHODS	This study was performed in three phases. In vitro, ACTs were measured on volunteer (n = 10) blood samples spiked with increasing concentrations of dalteparin or UFH. To extend these observations in vivo, ACTs were then measured in patients (n = 15) who were sequentially treated with IV dalteparin and then UFH. Finally, a larger monitoring study was undertaken involving patients (n = 110) who received dalteparin 60 or 80 international U (IU)/kg alone or followed by abciximab. We measured ACT (Hemochron), activated partial thromboplastin time (aPTT), plasma anti-Xa and anti-IIa levels, tissue factor pathway inhibitor (TFPI) concentration, and plasma dalteparin concentration.
RESULTS	Dalteparin induced a significant rise in the ACT with a smaller degree of variance as compared to UFH. Five min after administration of IV dalteparin 80 IU/kg the ACT increased from 125 s (122 s, 129 s) to 184 s (176 s, 191 s) (p < 0.001). The aPTT, anti-Xa and anti-IIa activities, and TFPI concentration also demonstrated significant increases following IV dalteparin.
CONCLUSIONS	The ACT and aPTT are sensitive to IV dalteparin at clinically relevant doses. These data suggest that the ACT may be useful in monitoring the anticoagulant effect of intravenously administered dalteparin during PCI. (J Am Coll Cardiol 2003;41:394–402) © 2003 by the American College of Cardiology Foundation

Advantages of low molecular weight heparin (LMWH) over unfractionated heparin (UFH) include greater bioavailability (1), a decreased incidence of thrombocytopenia (2), and resistance to inactivation by platelet factor 4 (3). The greater bioavailability gives LMWH the potential for subcutaneous administration without the need for monitoring (4–6). This advantage has led to a significant increase in the use of LMWH in the management of unstable coronary syndromes. Therefore an increasing number of patients referred for angiography and possible percutaneous coronary intervention (PCI) are arriving at the cardiac catheterization laboratory having received subcutaneous LMWH at various intervals from the time of subcutaneous injection. Though studies have demonstrated that LMWH can be used safely during PCI (7,8), an optimal strategy for the management of the anticoagulation of these patients has not been defined.

Current management of anticoagulation with UFH in

the angioplasty setting uses the ACT to guide dosing (9,10). The ACT is a broadly used point-of-care assay, which appears to be sensitive to thrombin inhibition (11) and relatively insensitive to factor Xa inhibition (12). Because dalteparin is a LMWH that retains substantial anti-IIa activity (13), we hypothesized that the ACT would be sensitive to intravenous (IV) dalteparin at clinically relevant doses. Thus, the purpose of this study was 1) to compare the dose-response relationship between IV dalteparin and the ACT to the relationship between UFH and the ACT and 2) to determine whether the anticoagulant effects of IV dalteparin could be monitored during PCI by measuring the changes in the ACT.

METHODS

The Institutional Review Board of the Mount Sinai Hospital, New York, NY, approved all protocols for this study. This study was performed in three phases. In the first phase, the effect of increasing concentrations of dalteparin on the ACT was studied and compared with changes induced by increasing concentrations of UFH in volunteer blood samples in vitro. To extend these observations in vivo and to allow for a paired comparison, the second phase of the study involved the generation of dose-response curves by sequentially administering dalteparin and UFH to the same pa-

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Abbreviations and Acronyms

ACT	= activated clotting time
aPTT	= activated partial thromboplastin time
CVA	= cerebrovascular accident
IU	= international unit
IV	= intravenous
LMWH	= low molecular weight heparin
MI	= myocardial infarction
PCI	= percutaneous coronary intervention
TFPI	= tissue factor pathway inhibitor
UFH	= unfractionated heparin

tient. The third phase comprised a larger observational monitoring study in which patients undergoing cardiac catheterization or PCI were treated with dalteparin alone or dalteparin plus abciximab.

Comparative effects of dalteparin and UFH on the ACT in vitro. An in vitro comparative study was performed initially. This dose-response study used blood samples from 10 healthy volunteers (4 men and 6 women; mean age 39.4 ± 13.5 years). Twelve milliliters of blood were collected from each of the healthy volunteers, free of medication, via venipuncture of the antecubital vein using 21-gauge needles. Dalteparin (12,500 IU/ml) and UFH (1,000 USP U/ml) were both diluted with saline to 50 U/ml and added to aliquots of whole blood to achieve final concentrations of 0, 0.25, 0.5, 0.75, 1.0, and 1.25 U/ml. These concentrations were chosen because they yielded changes in the ACT that were within a clinically relevant range (125 to 296 s for UFH). These aliquots were immediately injected into CA510 tubes (International Technidyne, Edison, New Jersey) containing the activator Celite (diatomaceous earth) and agitated vigorously. The CA510 tubes were then placed in the Hemochron 801 (International Technidyne, Edison, New Jersey) and the ACT was performed in duplicate. Results were recorded in seconds.

IN VIVO DOSE RESPONSE. To extend our observations in vivo, we compared the effects of 80 IU/kg of IV dalteparin to a standard interventional dose of IV UFH (70 U/kg). The key feature of this in vivo dose-response study was that dalteparin and UFH were given sequentially to the same patient, thereby allowing for a paired comparison. Fifteen patients (mean age 61.1 ± 10.1 years; 67% men) with a diagnosis of stable angina, asymptomatic with positive exercise stress test, unstable angina, non-Q-wave myocardial infarction (MI), or chest pain post-MI undergoing coronary angiography ($n = 5$) or PCI ($n = 10$) were enrolled after informed consent. The demographics of this study population are shown in Table 1. All patients undergoing PCI were given aspirin and clopidogrel before the procedure; abciximab was given to three of the 10 PCI patients at the discretion of the operator. Patients received a total of 80 IU/kg IV dalteparin administered in two separate boluses of 40 IU/kg each at baseline and at 5 min. After a washout period of at least 12 h, a total of 70 U/kg

Table 1. Patient Demographics

	In Vivo Dose-Response (n = 15)	Monitoring Study (n = 110)
Age	61.1 ± 10.1	61.8 ± 11.6
Men, n (%)	10 (67)	72 (66)
Hypertension, n (%)	13 (87)	88 (80)
Lipidemia, n (%)	11 (73)	86 (78)
Smoking, n (%)	4 (27)	53 (48)
Diabetes, n (%)	8 (53)	58 (53)
Previous MI, n (%)	7 (47)	19 (17)
Previous CABG, n (%)	1 (7)	10 (9)
Previous PCI, n (%)	9 (60)	44 (40)
Left anterior descending, n (%)	1 (7)	36 (33)
Circumflex, n (%)	2 (14)	22 (20)
Right coronary artery, n (%)	3 (20)	32 (29)
Saphenous vein graft	0 (0)	2 (2)
In-stent restenosis, n (%)	0 (0)	1 (1)

Data presented as mean $\pm$ SD or number (percentage) of patients.

CABG = coronary artery bypass graft; MI = myocardial infarction; PCI = percutaneous coronary intervention.

IV UFH was administered as a split bolus of 40 U/kg and 30 U/kg at baseline and at 5 min, respectively. Blood samples were drawn at baseline, and 5 and 10 min after IV drug administration from the arterial access sheath (dalteparin) or antecubital vein (UFH) into vacutainers containing one volume 3.8% sodium citrate (Becton Dickinson, Franklin Lakes, New Jersey). The anticoagulant effects of both IV dalteparin and UFH were assessed by measuring the ACT, activated partial thromboplastin time (aPTT), anti-Xa levels, and anti-IIa levels.

Monitoring study. On the basis of the observations from the in vitro and in vivo dose-response studies, we undertook an observational monitoring study in which patients were given either 60 or 80 IU/kg IV dalteparin. In this phase of the study, there was no UFH comparator arm.

PATIENT POPULATION. The study population was derived from patients with a diagnosis of stable angina, asymptomatic with positive exercise stress test, unstable angina, non-Q-wave MI, or chest pain post-MI undergoing cardiac catheterization at Mount Sinai Hospital, New York, New York. A total of 110 patients were enrolled over an eight-month period (4/00 to 12/00). Of the 110 patients, 24 (22%) underwent coronary angiography and 86 (78%) underwent PCI. The demographics of the monitoring study population are shown in Table 1. A diagram of patient flow is shown in Figure 1. Informed consent was obtained from each patient before enrollment.

Exclusion criteria for the in vivo dose response and monitoring studies included MI within 24 h, active internal bleeding, recent (within six weeks) gastrointestinal or genitourinary bleeding of clinical significance, history of cerebrovascular accident (CVA) within two years or CVA with a residual neurologic deficit, bleeding diathesis, administration of oral anticoagulant within seven days unless prothrombin time was <1.2 times control, thrombocytopenia ($<100,000/\mu\text{l}$), recent (within six weeks) major surgery or

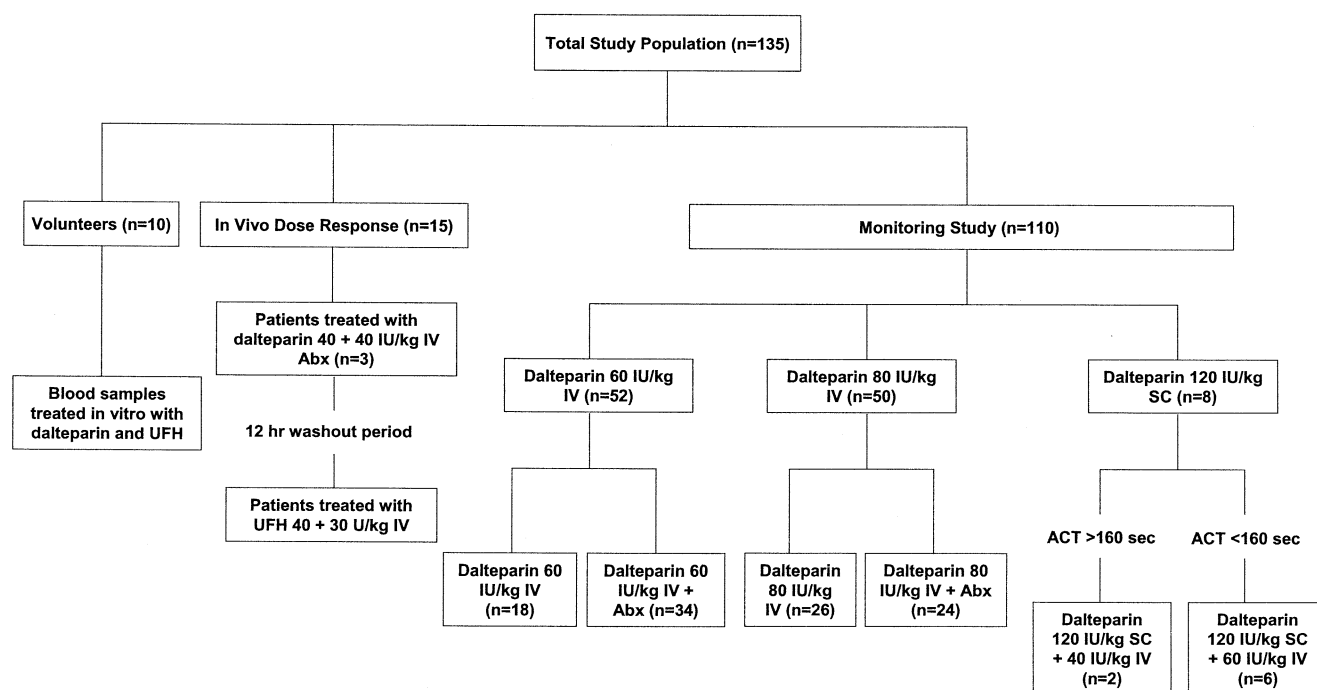


Figure 1. Patient flow for the in vitro dose response (volunteers), in vivo dose response, and monitoring studies. Abx = administration of abciximab; IV = intravenous; SC = subcutaneous.

trauma, intracranial neoplasm, arteriovenous malformation or aneurysm, severe uncontrolled hypertension $>180/100$ mm Hg on enrollment to the study, use of IV dextran before PCI, or hypersensitivity to one of the study drugs or their components. Because excretion of dalteparin is predominantly renal, patients with creatinine ≥ 2.0 were also excluded. Enrolled patients being treated with IV UFH had their infusion of UFH discontinued at least 4 h before catheterization. Patients too unstable to allow for heparin cessation on call to the cardiac catheterization laboratory were excluded. Patients with a baseline ACT ≥ 160 s were excluded because an ACT above this level was likely due to residual heparin effect that could confound the analysis of dalteparin's effect on the indices of coagulation. Patients who had received subcutaneous dalteparin ($n = 8$) before catheterization were not excluded on the basis of the initial ACT. In these patients, an ACT >160 s mandated a reduced dose (40 IU/kg) of dalteparin.

STUDY MEDICATIONS. The effects of ACT, aPTT, and other coagulation parameters were studied using two doses of IV dalteparin (60 IU/kg or 80 IU/kg), with and without abciximab. The initial 52 consecutive patients received 60 IU/kg IV dalteparin, and the subsequent 50 consecutive patients received 80 IU/kg IV dalteparin. An initial dose of 60 IU/kg was selected on the basis of previously published data (14). The dose of 80 IU/kg was chosen on the basis of a 20 IU/kg increment (40 IU/kg to 60 IU/kg) used in the aforementioned study by Kereiakes *et al.* (14).

A total of 110 patients received IV dalteparin. Of the 110 patients receiving IV dalteparin, 52 (47%) received a dose of 60 IU/kg. Thirty-four of these 52 patients (65%) also

received abciximab (0.25 mg/kg bolus and 0.125 $\mu\text{g/kg/h}$ maintenance infusion). Of the 110 patients receiving IV dalteparin, 50 (46%) received a dose of 80 IU/kg. Twenty-four of these 50 patients (48%) also received abciximab. Abciximab was given at the operator's discretion and was administered after the 10 min blood draw. All patients undergoing PCI were given aspirin and clopidogrel before the procedure.

SUBCUTANEOUS SUBSTUDY. Of the 110 patients, eight received a single subcutaneous injection of dalteparin 120 IU/kg during the noninvasive phase of their hospital course. Using data from the dose-response study, it was arbitrarily decided that immediately before PCI patients with an ACT >160 s ($n = 2$) were to receive an IV dalteparin 40 IU/kg bolus and patients with ACT <160 s ($n = 6$) were to receive an IV dalteparin 60 IU/kg bolus. Four of the eight patients received abciximab (0.25 mg/kg bolus and 0.125 $\mu\text{g/kg/h}$ maintenance infusion) 10 min after dalteparin at the operator's discretion.

BLOOD SAMPLES. Six blood samples were collected from the arterial access sheath in sodium citrate vacutainers at the following time points: baseline and 5, 10, 20, 40, and 60 min after IV dalteparin in the observational monitoring study. Patients in the subcutaneous substudy had their blood samples drawn directly from the antecubital vein at baseline and then every hour following dalteparin subcutaneous administration until the time of the procedure. Blood samples were centrifuged at 3,000 g for 15 min at 22°C. Platelet-poor plasma was collected and aliquoted into 2 ml eppendorf tubes and stored at -70°C . Frozen samples were

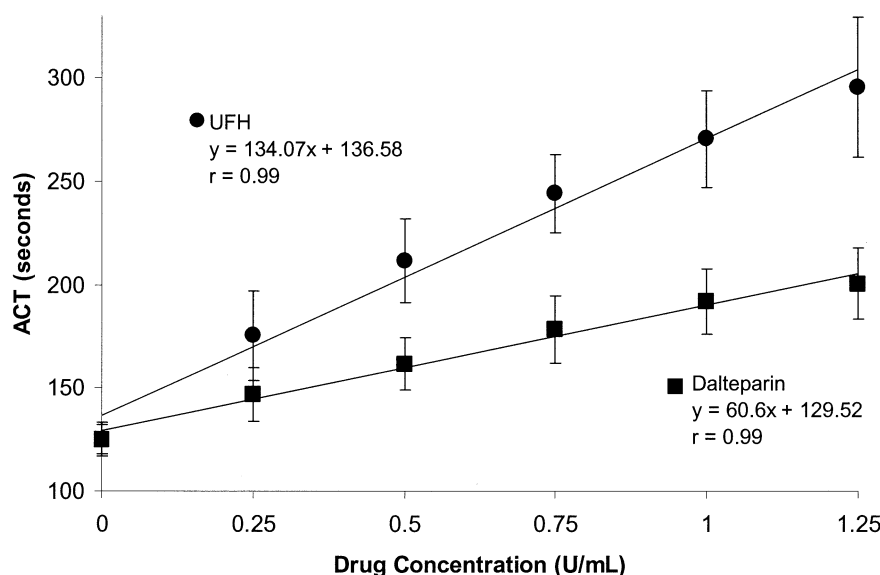


Figure 2. In vitro dose-response of the activated clotting time (ACT) as a function of increasing concentration of anticoagulant in whole blood. Samples were derived from healthy volunteers ($n = 10$) and aliquots ($n = 200$) were treated with increasing doses of either unfractionated heparin (UFH) (circles) or dalteparin (squares). Data presented as mean $\pm$ SD.

transported on dry ice to the Thrombosis and Hemostasis Research Laboratory at Loyola Medical Center (Maywood, Illinois). Arterial access sheaths were removed 2 to 4 h after completion of the procedure.

MONITORING OF THE ANTICOAGULANT EFFECT. ACT. Two milliliters of blood was collected in a FTCA510 tube (International Technidyne, Edison, New Jersey) with the activator Celite. The tube was vigorously shaken and placed in the Hemochron 8000 (International Technidyne, Edison, New Jersey). Results were recorded in seconds.

aPTT. Samples were analyzed using STA-PTT A 5, a reagent containing rabbit cerebral cephalin and silica (Diagnostica Stago, Asnieres-Sur-Seine, France) on the STA-R (Diagnostica Stago, Gennevilliers, France). Results were recorded in seconds.

ANTI-PROTEASE ASSAYS. Both the anti-Xa (15) and anti-IIa (16) assays were performed as previously described. Anti-Xa and anti-IIa activities were both adjusted to a reference baseline value of 0.0 IU/ml in all cohorts.

HEPTEST ASSAY. Heptest was performed on a fibrometer, according to Yin *et al.* (17). Heptest activity was adjusted to a reference baseline value of 0.0 IU/ml in all cohorts.

QUANTIFICATION OF TFPI. A tissue factor pathway inhibitor (TFPI) enzyme linked immunoabsorbent spectrophotometric assay was performed (18,19). Results were expressed as nanograms/milliliter.

Assessment of safety. Minor and major bleeding was defined according to the criteria used by the Thrombolysis in Myocardial Infarction (TIMI) Trial (20). Severe thrombocytopenia was defined by a platelet count below 50,000/ μ l. Mild thrombocytopenia was defined as a platelet count below 100,000/ μ l or a count below 50% of the baseline

value. The degree of thrombocytopenia was assessed post-procedurally after either dalteparin alone or combination dalteparin and abciximab therapy. Myocardial infarction was defined as an elevation in CK-MB three times the upper limit of normal. Blood for CK-MB analysis was drawn before PCI and every 8 h for 24 h.

Statistical analysis. Patient demographic and coagulation parameter data were described using mean (95% confidence interval) and mean $\pm$ SD where indicated. Analysis of variance was used to compare the effect on coagulation parameters across the six sampling time points for each dose of dalteparin, 60 IU/kg and 80 IU/kg. Normality was tested using the Shapiro-Wilkes test for small samples. All data passed normality with the exception of the ACT data from the sequential dalteparin and UFH group, which were transformed using logarithms and retested. The variability of the ACT response to dalteparin and UFH were determined by calculating the coefficient of variance (mean/SD $\times$ 100) for both drugs in the in vitro and in vivo dose-response studies. To determine whether these variations were similar, an F test was performed as previously described (21). All statistical analyses were performed using SPSS version 10.0. A two-tailed $p < 0.05$ was considered statistically significant.

RESULTS

Comparative effects of dalteparin and UFH on the ACT in vitro. Dalteparin and UFH both demonstrated a significant dose response with respect to the ACT (Fig. 2). Although a greater slope was seen with UFH, a moderate slope was present in the dalteparin-induced curve. The degree of variability in ACT at the highest concentration (1.25 U/ml) as measured by the coefficient of variance was

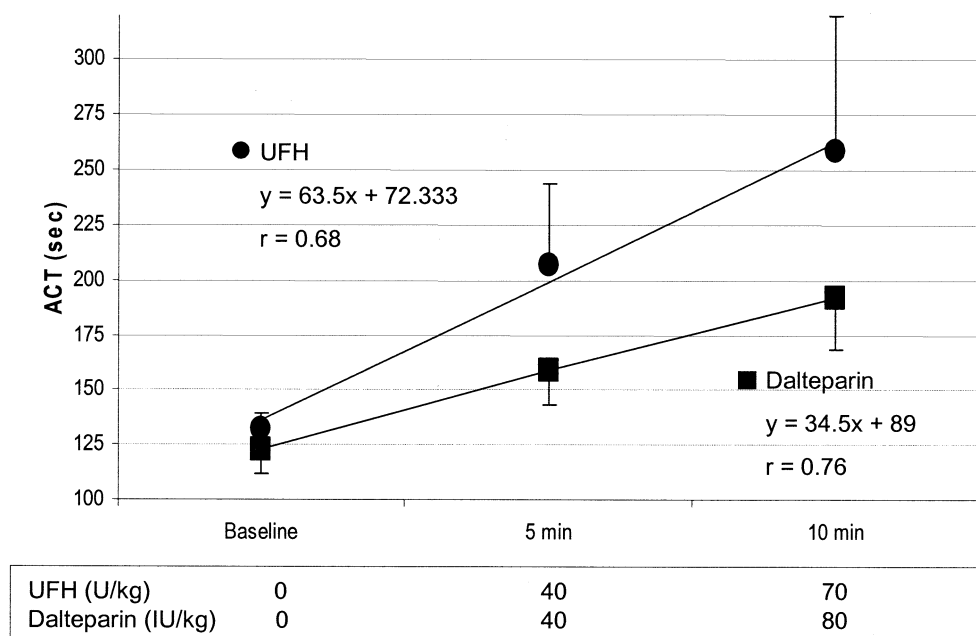


Figure 3. In vivo dose-response of the ACT as a function of the cumulative doses of UFH (circles) or dalteparin (squares). Patients (n = 15) initially received dalteparin (40 + 40 IU/kg 5 min apart, for a total of 80 IU/kg), and then, following a 12-h delay to allow elimination of dalteparin and return of the ACT to baseline, received UFH (40 + 30 U/kg 5 min apart, for a total of 70 U/kg). Data presented as mean $\pm$ SD. Abbreviations as in Figure 2.

numerically, but not statistically, lower for dalteparin than for UFH (8.5 vs. 11.4; $p = 0.26$).

Comparative effects of dalteparin and UFH on the ACT in vivo. As was seen in the in vitro experiments, a significant rise in ACT was discernible as the dose of both dalteparin and UFH was increased in vivo (Fig. 3). The ACT of patients receiving 80 IU/kg dalteparin demonstrated a significantly lower coefficient of variance as compared to patients receiving 70 U/kg UFH (12.5 vs. 23.6; $p = 0.03$).

After the boluses of dalteparin and UFH were administered, the ACT, aPTT, anti-Xa activity, and anti-IIa activity all increased (Table 2). The increases observed in the ACT after dalteparin administration from samples drawn at the 5 and 10 min time points demonstrated a significant but weak correlation to the elevations in anti-Xa ($r = 0.47$; $p = 0.01$) and anti-IIa activities ($r = 0.37$; $p = 0.048$). After UFH administration numerically weaker correlations were seen between the increases in the ACT and

the elevations in anti-Xa ($r = 0.06$; $p = 0.75$) and anti-IIa activities ($r = 0.26$; $p = 0.18$) from samples collected at the 5- and 10-min time points.

Monitoring study. RESPONSE OF COAGULATION PARAMETERS TO IV DALTEPARIN. The changes in coagulation parameters for the 102 patients in the monitoring study who received IV dalteparin without pretreatment with subcutaneous dalteparin are listed in Table 3. Following IV dalteparin 60 or 80 IU/kg, there was a significant elevation in the ACT at 5 min ($p < 0.001$) and throughout the 60-min observation period ($p < 0.001$). A rise in the ACT of at least 20% from baseline values was seen in every patient studied. In comparison to 60 IU/kg, an 80 IU/kg IV dose of dalteparin led to a significantly greater elevation in the ACT that was sustained throughout the 60-min observation period ($p < 0.001$). Similar trends were seen in the aPTT, in the plasma levels of anti-Xa and anti-IIa activity, and Heptest (Table 3). These trends were also present in the group of patients who were treated with dalteparin alone

Table 2. Changes in Coagulation Parameters Induced by Intravenous Dalteparin and UFH

Drug dose	Coagulation Parameters			
	ACT (s)	aPTT (s)	Anti-Xa (IU/ml)	Anti-IIa (IU/ml)
Dalteparin baseline	123 $\pm$ 11	28.5 $\pm$ 2.3	0.0 $\pm$ 0.0	0.0 $\pm$ 0.0
Dalteparin 40 IU/kg	157 $\pm$ 16	55.6 $\pm$ 15.0	1.0 $\pm$ 0.6	0.4 $\pm$ 0.5
Dalteparin 80 IU/kg	192 $\pm$ 24	88.6 $\pm$ 21.7	1.9 $\pm$ 0.8	1.1 $\pm$ 0.5
UFH baseline	132 $\pm$ 7	29.4 $\pm$ 2.7	0.0 $\pm$ 0.0	0.0 $\pm$ 0.0
UFH 40 U/kg	207 $\pm$ 37	106.5 $\pm$ 13.3	0.5 $\pm$ 0.3	0.6 $\pm$ 0.3
UFH 70 U/kg	259 $\pm$ 61	110.0 $\pm$ 0.0	1.1 $\pm$ 0.5	0.9 $\pm$ 0.3

Data presented as mean $\pm$ SD. Baseline anti-Xa activity and anti-IIa activity are reference values for assays used. Patients (n = 15) were treated sequentially with dalteparin and then, after a minimum washout period of 12 h, with UFH.

ACT = activated clotting time; aPTT = activated partial thromboplastin time; IU = international units; UFH = unfractionated heparin.

Table 3. Indices of Coagulation After Intravenous Dalteparin and Abciximab

Assay	Dose (IU/kg)	Pre-Abciximab			Post-Abciximab*		
		Baseline	5 min	10 min	20 min	40 min	60 min
ACT (s)	60	128 (123, 132)	175 (167, 182)	177 (171, 183)	190 (182, 198)	186 (173, 198)	175 (165, 184)
	80	125 (122, 129)	184 (176, 191)	187 (180, 193)	189 (182, 195)	186 (179, 193)	180 (174, 187)
aPTT (s)	60	31.0 (27.5, 34.5)	63.6 (57.0, 70.3)	63.7 (57.7, 69.7)	70.4 (61.8, 79.1)	66.2 (57.3, 75.1)	59.0 (52.9, 65.1)
	80	29.0 (27.7, 30.3)	78.6 (71.7, 85.4)	78.4 (72.7, 84.1)	83.3 (76.4, 90.2)	82.7 (75.3, 90.1)	77.5 (70.1, 84.8)
Anti-Xa (IU/ml)	60	0.0	1.15 (1.03, 1.27)	1.14 (1.04, 1.24)	1.00 (0.91, 1.11)	0.86 (0.75, 0.97)	0.71 (0.60, 0.82)
	80	0.0	1.55 (1.43, 1.68)	1.48 (1.38, 1.58)	1.45 (1.31, 1.58)	1.24 (1.08, 1.39)	1.03 (0.86, 1.21)
Anti-IIa (IU/ml)	60	0.0	0.65 (0.57, 0.71)	0.65 (0.59, 0.71)	0.58 (0.51, 0.64)	0.49 (0.40, 0.59)	0.38 (0.32, 0.44)
	80	0.0	0.81 (0.76, 0.87)	0.77 (0.72, 0.82)	0.73 (0.67, 0.79)	0.62 (0.55, 0.69)	0.48 (0.40, 0.57)
Heptest (IU/ml)	60	0.0	1.39 (1.26, 1.51)	1.34 (1.25, 1.44)	1.23 (1.13, 1.33)	1.04 (0.94, 1.14)	0.89 (0.77, 1.01)
	80	0.0	1.74 (1.59, 1.88)	1.69 (1.57, 1.82)	1.66 (1.52, 1.81)	1.42 (1.27, 1.56)	1.19 (1.02, 1.35)
TFPI (ng/ml)	60	57 (52, 63)	411 (372, 450)	465 (433, 497)	449 (417, 481)	396 (360, 432)	314 (278, 350)
	80	46 (41, 51)	341 (306, 377)	356 (328, 384)	356 (322, 390)	308 (275, 340)	273 (234, 312)

Values are presented as mean (95% CI). *34 of the 52 patients treated with 60 IU/kg, and 24 of the 50 patients treated with 80 IU/kg received abciximab. $p < 0.001$ for all timepoints compared to baseline.

CI = confidence interval; TFPI = tissue factor pathway inhibitor. Other abbreviations as in Table 2.

(Fig. 4). Note that in this group of patients, a numerical decline in the ACT at 60 min was noted in comparison to the peak levels at 10 min (156 vs. 167 s for 60 IU/kg dose, $p = 0.09$; 175 vs. 187 s for the 80 IU/kg dose, $p = 0.10$).

The ACT and aPTT values generated from samples drawn at baseline and 5 min post-dalteparin were correlated ($r = 0.82$; $p < 0.001$). Elevations in anti-Xa and anti-IIa activity were weakly correlated with changes in the ACT ($r = 0.26$; $p = 0.012$ and $r = 0.31$; $p = 0.002$, respectively).

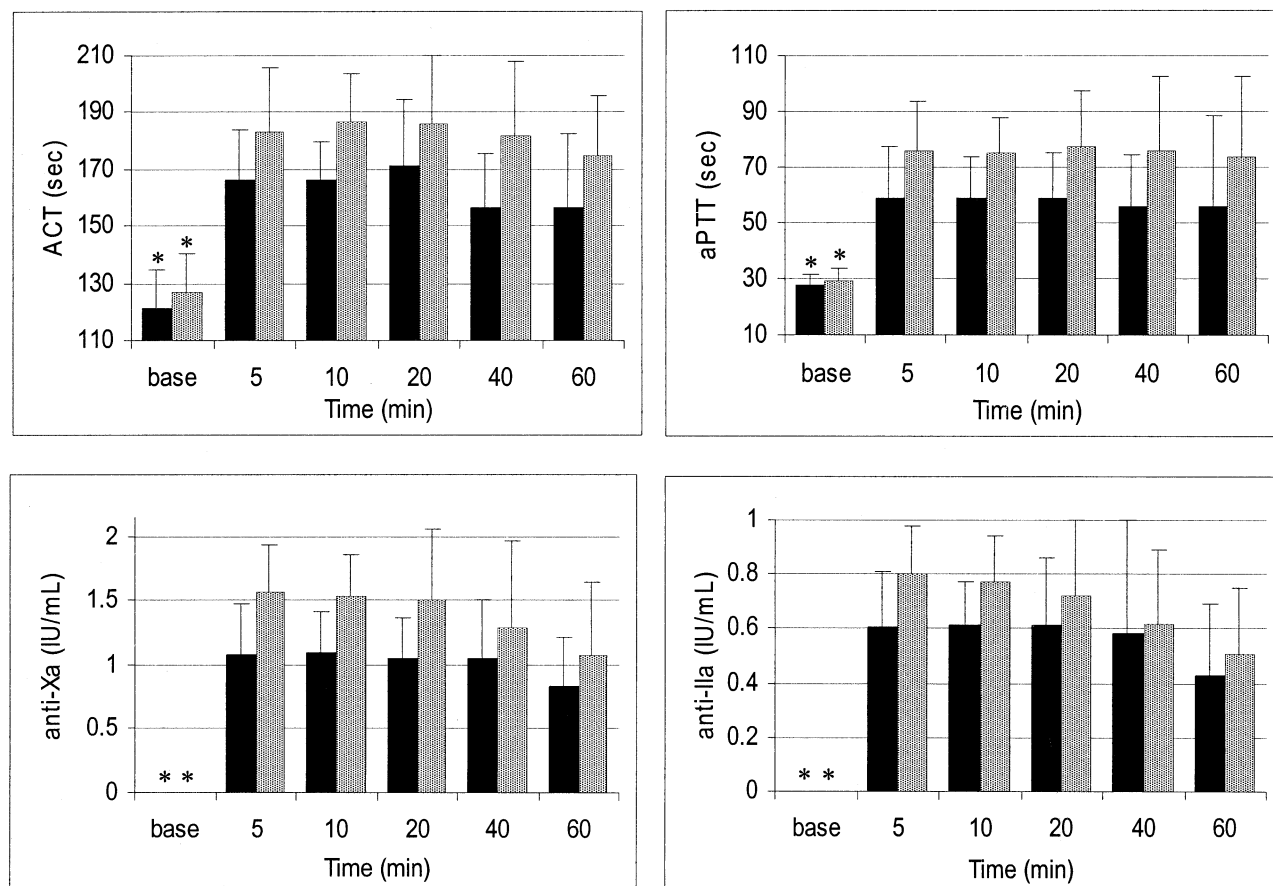


Figure 4. Indices of coagulation following administration of intravenous (IV) dalteparin 60 IU/kg ($n = 18$) and IV dalteparin 80 IU/kg ($n = 26$). Data presented as mean (95% confidence interval). * $p < 0.001$ for all time points compared to baseline. **Black bars** = dalteparin 60 IU/kg; **gray bars** = dalteparin 80 IU/kg.

RESPONSE TO SUBCUTANEOUS DALTEPARIN ADMINISTRATION. Eight patients in the study received subcutaneous dalteparin 120 IU/kg at least 2 h before PCI. There was an increase in ACT from a baseline of 126 ± 10 s to 146 ± 11 s at 1 h, with a peak value of 151 ± 13 s at 2 h ($p < 0.001$). Similarly, aPTT rose from a baseline of 28.2 ± 2.5 s to 36.6 ± 6.1 s at 1 h ($p < 0.001$) and to 38.7 ± 5.8 s at 2 h ($p < 0.001$). Anti-Xa activity increased to 0.56 ± 0.24 IU/ml at 1 h and 0.70 ± 0.18 IU/ml at 2 h ($p < 0.001$).

These patients received IV dalteparin 40 IU/kg ($n = 2$) or 60 IU/kg ($n = 6$) during PCI depending on the ACT measured immediately after sheath insertion. In those patients receiving IV dalteparin 40 IU/kg, 5 min after the IV bolus the ACT increased from 160 ± 1 s to 220 ± 10 s; aPTT increased from 42.2 ± 13.3 s to 71.6 ± 10.7 s and anti-Xa activity increased from 0.79 ± 0.4 IU/ml to 1.21 ± 0.4 IU/ml. In those patients receiving IV dalteparin 60 IU/kg at 5 min after the IV bolus, the ACT increased from 141 ± 13 s to 172 ± 20 s ($p < 0.001$); aPTT increased from 36.7 ± 2.6 s to 76.3 ± 12.2 s ($p < 0.001$) and anti-Xa activity increased from 0.71 ± 0.26 IU/ml to 1.79 ± 0.64 IU/ml ($p = 0.006$).

Safety outcomes. DEATH, MI, AND URGENT REVASCULARIZATION. There was no death or urgent revascularization during the hospital course of the study population. Two of the 96 PCI patients had MI (2.1%).

BLEEDING RATES IN PATIENTS UNDERGOING PCI. One patient in the IV dalteparin 80 IU/kg and abciximab cohort suffered major bleeding requiring transfusion, yielding a major bleeding rate of 1.0% (1/96). Two patients in the IV dalteparin 80 IU/kg and abciximab cohort and one each in the in vivo dose-response study and subcutaneous dalteparin 120 IU/kg cohort had minor bleeding, yielding a minor bleeding rate of 4.2% (4/96). No patients with minor bleeding required blood transfusion.

EPISODE OF THROMBOCYTOPENIA. One patient in the IV dalteparin 80 IU/kg and abciximab cohort experienced an episode of severe thrombocytopenia with no sequelae. There were no episodes of mild thrombocytopenia.

DISCUSSION

This is the first study to prospectively analyze the effects of IV dalteparin on the ACT. Our findings suggest that the ACT may constitute a reliable assay for monitoring dalteparin during PCI for the following reasons. First, the elevation is significant and rapidly detectable; within 5 min of receiving IV dalteparin 80 IU/kg, the ACT increased on average 59 s (Table 3). Second, there is a highly significant dose-response relationship in vitro ($r = 0.99$) (Fig. 2) and in vivo ($r = 0.76$) (Fig. 3). A dose response is also demonstrated by the fact that patients receiving IV dalteparin 80 IU/kg experienced a significantly greater rise in their ACT in comparison with those receiving 60 IU/kg. Third, in comparison to the rise in the ACT induced by UFH, there

appears to be less variance in the dalteparin-induced elevations in the ACT both in vitro and in vivo. Fourth, the increase in ACT was sustained for a period of time relevant to current interventional practice (40 min, with a decline in values at approximately 60 min for patients treated with dalteparin alone). This raises the possibility that a decline in the ACT to a targeted level (for example, <150 s) could potentially be used to determine the timing of sheath removal. Finally, these observations are consistent with previous reports. In a study by Kereiakes *et al.* (14), patients treated with either 40 IU/kg or 60 IU/kg of dalteparin IV achieved mean ACTs 30 min after administration of 166 ± 28 s and 180 ± 38 s, respectively. Similarly, in a study using IV enoxaparin during PCI the mean ACT increased from 130 ± 19 to 188 ± 29 s 5 min after administration (7). These previously reported data support the notion that the ACT may be useful in monitoring the effects of intravenously administered LMWH.

To determine whether the elevation seen in the ACT following IV dalteparin occurred in isolation or in conjunction with changes in other indices of anticoagulation, the aPTT, plasma anti-Xa and anti-IIa activities, and TFPI concentration were also measured. The aPTT rose significantly after IV dalteparin administration and to a level traditionally regarded as therapeutic in the context of medical management for unstable angina (1.5- to 2-fold above baseline) (22). The aPTT demonstrated a significant correlation ($r = 0.82$) with the ACT, supporting the notion that the elevated ACT values post-dalteparin reflected an anticoagulated state. Studies have shown both the ACT and aPTT to be prolonged by direct thrombin inhibitors (11,23) and to be relatively insensitive to Factor Xa inhibition (12,24). These observations suggest that the rise in the ACT documented in the present study was more likely due to dalteparin's anti-IIa activity than to its anti-Xa activity. A theoretic advantage of factor IIa inhibition, versus factor Xa inhibition, is that inhibition of thrombin may prevent feedback activation of factors V and VIII (25,26).

Our findings of poor correlations between the ACT and anti-Xa and anti-IIa activities indicate that dalteparin-induced changes in the ACT cannot be accounted for solely on the basis of dalteparin's anti-Xa and anti-IIa effects. Despite the poor correlation in this study between the ACT and plasma anti-Xa and anti-IIa activities, as well as ambiguity in the literature regarding a correlation between the ACT and anti-Xa activity in UFH-treated patients (27,28), monitoring of UFH with the ACT to achieve target levels between 200 and 300 s has become a generally accepted standard in interventional practice (29). Furthermore, unlike anti-Xa and anti-IIa levels, the ACT has been correlated to clinical outcomes (10,30). One explanation for the observation that the ACT appears to correlate better to clinical outcomes than to specific levels of factor Xa or IIa inhibition is that the ACT is influenced by a variety of factors that collectively determine the blood's propensity to thrombose. For example, antiplatelet agents have been

noted to increase the ACT (31). In addition, both dalteparin and UFH affect the coagulation mechanism at multiple levels. Antithrombin (whether potentiated by LMWH or UFH) not only exerts anti-Xa and anti-IIa effects, but also has been shown to inhibit factors XIIa (32), XIa (33,34), and IXa (35,36) of the intrinsic coagulation pathway. These agents also induce the release of TFPI (37) and may alter the inducibility of tissue factor on monocytes, thereby providing additional mechanisms to retard thrombus formation. Seen in this light, it is hardly surprising that weak correlations were found between the ACT and specific anti-Xa and anti-IIa levels.

Others have proposed that rather than using the ACT, the effects of LMWH in patients undergoing PCI would be better gauged by measuring anti-Xa levels (28,38). However, these observations pertain to LMWH administered subcutaneously and to a specific formulation, enoxaparin, that has a diminished anti-IIa effect in comparison to dalteparin (13). Moreover, measurement of anti-Xa activity would assess the action of LMWH on an isolated (albeit important) step of the cascade, but would not take into account the multiple levels at which a LMWH acts (factors XIIa, XIa, IXa, and TFPI as noted earlier). Additionally, to our knowledge previous clinical studies have demonstrated only weak correlations between anti-Xa levels and thrombus formation (39,40), and reported conflicting data on the relationship between anti-Xa levels and the incidence of hemorrhage (40–42). In contrast, a recent study by Chew *et al.* demonstrated a significant relationship between the ACT and ischemic/bleeding events after PCI in patients receiving UFH (10). Furthermore, anti-Xa assays used for monitoring display interassay variability depending on assay technique (43) or type (44) and are not readily available as a bedside device. Thus, it should not be assumed *a priori* that anti-Xa activity monitoring would be superior to monitoring the ACT in patients receiving intravenous LMWH.

Point-of-care monitoring of the effects of LMWH may be particularly important in patients with morbid obesity or renal dysfunction (26,45,46), two populations in whom rigorous dosing studies have not been reported (46). Additional benefits of monitoring may include detection of errors in IV administration (47) and the potential for dose adjustment in patients receiving antithrombotic, fibrinolytic, or antiplatelet therapy before PCI.

Study limitations. There are several limitations associated with the study. First, ACTs were measured using the Hemochron system, and thus our data may not be readily extrapolated to the Hemotech system (Medtronic, Parker, Colorado) used in many cardiac catheterization laboratories. Second, the study design was nonrandomized and open label. Third, there was no comparator arm with heparin alone in the monitoring study. Fourth, abciximab was the lone adjunctive glycoprotein IIb/IIIa inhibitor used, and therefore the results may not apply to patients receiving eptifibatide or tirofiban. Fifth, our findings may not be

generalized to all LMWHs; the effect of dalteparin on the ACT, aPTT, and other tests may differ from other LMWH agents because of differences in the ratios of anti-Xa:anti-IIa activity. The lower anti-Xa:anti-IIa ratio for dalteparin compared to enoxaparin (2.7 vs. 3.8), for example, may result in greater sensitivity of anti-IIa dependent tests for dalteparin than for enoxaparin. Finally, blood samples were collected for only 60 min after the initial dalteparin bolus, thereby limiting the study temporally. A longer period of sampling may have helped to delineate the duration of the anticoagulated state induced by dalteparin.

In conclusion, the ACT and aPTT are sensitive to IV dalteparin at clinically relevant doses. These data suggest that the ACT may have the potential to be integrated into an IV dalteparin monitoring strategy. The ability to monitor dalteparin may facilitate the use of LMWH in PCI and in other invasive procedures such as coronary artery bypass surgery.

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