Heparin, Low Molecular Weight Heparin, and Derivatives in Thrombosis, Angiogenesis, and Inflammation: Emerging Links

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ABSTRACT

The key reason behind the success of heparin in thrombosis and beyond is its polypharmacological sites of action for the prevention and treatment of multifactorial diseases that will only benefit slightly with single pharmacological mechanism–based agents. Thromboembolic disorders are driven by hypercoagulable, hyperactive platelet, proinflammatory, endothelial dysfunction, and proangiogenesis states. Heparin can effectively modulate all of those multifactorial components, as well as the interface among those components.

KEYWORDS:

In recent years, clinical data and studies have clarified the potential and shortcomings of anticoagulant therapy in the prevention and treatment of thromboembolic disorders. The discovery and introduction of heparin derivatives such as low molecular weight heparins (LMWHs) have enhanced the clinical options for the management of thromboembolic disorders while enhancing the safety of therapy. In the United States, LMWHs are currently approved for the prophylaxis and treatment of deep vein thrombosis (DVT). LMWH uses are also being expanded for additional indications for the management of unstable angina and non–Q-wave myocardial infarction.¹⁻² In addition to the approved uses, LMWHs currently are being tested for several newer indications.³⁻⁴ Because they are polypharmacological agents, these drugs are expected to find uses in several other clinical indications, such as inflammatory diseases and cancer.³⁻⁴ Additional pharmacological studies and well-designed clinical trials in which various pharmacokinetic and pharmacodynamic parameters are studied will provide additional evidence on the clinical individuality of each member of this class of novel agents.¹⁻⁴

Because heparin was discovered more than a half century ago, our knowledge of the chemical structure and molecular interactions of this fascinating polycomponent was limited at the early stages of its development. Through the efforts of a major multidisciplinary group of researchers and clinicians, it is now well recognized that heparin has multiple sites of action (Table 1) and can be used in multiple indications. In the not-too-distant future, we may witness the impact of heparin derivatives in the management of various diseases.

EMERGING LINKS AMONG THROMBOSIS, ANGIOGENESIS, AND INFLAMMATION: POTENTIAL ROLE OF HEPARIN³⁻⁴

Several lines of evidence demonstrated the interplay between the platelet/leukocyte in the activated state and the coagulation cascade. That led to the exposure of the platelet glycoprotein IIb/IIIa receptors in its active...
state, leading to platelet fibrinogen binding and amplification of platelet aggregate formation. Activated platelets also interact with leukocytes, leading to platelet–leukocyte cohesion and leukocyte activation. Hyperactive platelets also provide a surface for thrombin generation; thrombin is a potent platelet and leukocyte activator. In addition, there is significant interplay among the coagulation cascade, platelets, and the vessel wall in the promotion of thromboembolic disorders. Depending upon the venous (low shear) versus arterial (high shear) shear level, platelet/fibrin proportions and contributions vary.

Emerging links are shown among thrombosis, angiogenesis, and inflammation in vascular, cardiovascular, and inflammatory disorders (Fig. 1). For example, infection leading to the initiation of proinflammatory stimuli could be a major predisposing factor in propagation of thromboembolic disorders. Endotoxin that can be liberated from Escherichia coli and other bacteria can induce proinflammatory state, with the increase of tissue necrosis factor alpha (TNF-α) and other cytokines. That would lead to the activation of leukocytes, with increased expression of membrane L-selectin and the shedding of soluble L-selectin, which can serve as a surrogate marker of leukocyte activation. Activation of leukocytes leads to the propagation and generation of tissue factor, which initiates and amplifies a hypercoagulable state as well as the upregulation of TNF-α production. A hypercoagulable state with the generation of thrombin activates the platelets, leading to the overexpression of platelet membrane P-selectin and the shedding of soluble P-selectin, which can act as a surrogate marker of platelet activation. In addition, the proinflammatory state can induce endothelial cell (EC) insult, leading to increased EC membrane expression and shedding of soluble vascular adhesion molecule-1, intracellular adhesion molecule-1, and E-selectin.

Heparin is a glycosaminoglycan formed from sulfated oligosaccharides; it varies in the length of polymeric units and therefore has different molecular weights. LMWH is made by partial hydrolysis or enzymatic degradation of unfractionated heparin (UFH). Heparin and LMWH prevent the process of blood coagulation and have a natural antithrombin effect. In recent years, several studies have shown that heparin and LMWH have an obvious anti-inflammatory activity in addition to their traditional anticoagulant effects. In animal models, heparin disaccharides inhibited TNF-α production by macrophages and decreased immune inflammation. Heparin accelerated the healing process of mucosa in colitis in several clinical studies and had anti-inflammatory effects. Therefore, administration of

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**Table 1** Heparin Mechanisms of Action

<table>
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<tr>
<th>Mechanism</th>
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<tr>
<td>AT dependent (FXa, FIIa, FVIIa, etc.)</td>
<td>Anticoagulant, antithrombotic</td>
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<tr>
<td>AT independent (TFPI)</td>
<td>FXa, TF/FVIIa: antithrombotic</td>
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<td>Angiogenesis, metastasis, and inflammation</td>
<td>AT and TFPI-independent:</td>
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<td>Inhibition of matrix-degrading enzymes, proteases, heparinas, inflammatory and cancer cell invasion</td>
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<td>Heparin and selectin modulation: metastasis, inflammation, VTE</td>
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<td>Other mechanisms?</td>
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AT, antithrombin; F, factor; TFPI, tissue factor pathway inhibitor; TF, tissue factor; VTE, venous thromboembolism.

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**Figure 1** Emerging links among thrombosis, angiogenesis, and inflammation in vascular, cardiovascular, and inflammatory diseases.
heparin can afford both non-anticoagulant (anti-inflammatory and antiangiogenesis) and anticoagulant effects (Fig. 2).

Heparin and Venous Thromboembolism

The large majority of patients with venous thromboembolism (VTE) currently are treated with full doses of UFH or LMWH, followed by at least 3 months of oral anticoagulant therapy. Selected patients with critical manifestations of pulmonary embolism (PE) are administered thrombolytic drugs, whereas intravenous cava filters are confined to patients with either DVT or PE who present with contraindications to conventional anticoagulation.

Although considerable progress has been made in the treatment of venous thromboembolic disorders, many unanswered questions remain, which are awaiting proper solution. Furthermore, new opportunities are emerging, which have the potential to modify the therapeutic scenario substantially in the near future. Among the topics that are worth exploring are home treatment of selected patients with DVT, the treatment of cancer patients with venous thrombosis, the renewed interest for thrombolytic drugs in patients with PE, the optimal duration of oral anticoagulant therapy, and the potential of new drug categories for the initial treatment and secondary prevention of VTE.

TREATMENT OF CANCER PATIENTS WITH VENOUS THROMBOSIS

Patients with DVT who also have cancer have a higher risk of recurrent thromboembolism and major bleeding during anticoagulation. In a recent prospective cohort study in a wide series of patients with venous thrombosis with or without cancer, the 12-month cumulative incidence of both recurrent thromboembolism and major bleeding during anticoagulation was significantly higher in patients with cancer than in those without cancer. Recurrence and bleeding were both related to cancer severity, occurred predominantly during the first month of anticoagulant therapy, but could not be explained by sub- or over-anticoagulation. Possibilities for improvement using the current paradigms of anticoagulation, therefore, seem limited, and new treatment strategies should be developed.

The long-term use of LMWH recently has been compared with warfarin for the initial treatment and secondary prevention of VTE in cancer patients with venous thrombosis, favoring LMWH. No significant difference between the dalteparin group and the oral anticoagulant group was detected in the rate of major bleeding or any bleeding. These results are supported by other investigation. Taken together, these two studies show clearly that in patients with cancer and acute VTE, LMWH is more effective than an oral anticoagulant in reducing the risk of recurrent thromboembolism without increasing the risk of bleeding.

TREATMENT OF PE

Patients with noncritical manifestations of PE have long been treated with UFH in therapeutic doses. To test the hypothesis that LMWH treatment can be extended to cover the entire spectrum of patients presenting with acute VTE (thus including also patients with noncritical PE), two multicenter clinical trials have been performed in the second half of the 1990s. In the first investigation, all consecutive...
patients with acute thromboembolism were enrolled in the study irrespective of the modality of clinical presentation; in contrast, in the second investigation, only patients with symptomatic PE were eligible for the study. In both studies, the investigated LMWH (reviparin and tinzaparin, respectively) proved to be at least as effective and safe as UFH, suggesting that under some circumstances also noncritical patients with symptomatic PE might be treated at home with LMWHs. In addition, a recent meta-analysis of all available comparative clinical trials has confirmed firmly that for the treatment of PE, LMWH is as effective and safe as UFH.23

Recent studies have put into question the systematic use of anticoagulants alone in the initial treatment of patients with submassive PE. Given that the risk of an unfavorable outcome definitely seems higher in patients with right ventricular dysfunction, as shown by echocardiography, the use of thrombolytic drugs (ie, drugs that have the potential to promptly restore the patency of the pulmonary arterial vessels), might improve the outcome of patients with PE. Recently, two meta-analyses of comparative studies between thrombolysis and heparin in the treatment of acute PE have been published.26,27 The results of these meta-analyses consistently show that patients treated with thrombolytic drugs have a more favorable outcome in terms of prevention of short-term recurrent episodes of PE than those treated with heparin alone. The difference becomes statistically significant when a composite end point consisting of death/recurrence is calculated.26 However, patients treated with thrombolytic drugs definitely exhibit a higher hemorrhagic risk.26,27

In a recent prospective controlled study, 256 patients with submassive PE and a contemporary right ventricular dysfunction were randomly assigned to receive heparin plus alteplase or heparin plus placebo.28 The results of this study have the potential to expand the use of thrombolysis in patients with acute PE, at least in those with right ventricular dysfunction. In addition, whether the prognostic value of echocardiographically assessed right ventricular dysfunction applies to noncritical patients with PE still remains to be demonstrated convincingly.29

THE OPTIMAL DURATION OF ANTICOAGULANT TREATMENT

Available Information from Recent Clinical Trials

Prospective cohort30–32 and population-based studies33 performed in recent years have shown that the incidence of recurrent VTE in patients receiving a short (3- to 6-month) course of anticoagulation is higher than previously believed. Schulman et al34 performed a multicenter trial comparing 6 weeks of oral anticoagulant treatment with 6 months of such therapy in 897 patients who had had a first episode of VTE. After 2 years of follow-up, there were 80 recurrences among the 443 patients randomly assigned to the 6-week group (18.1%), and 43 recurrences among the 454 patients randomly assigned to the 6-month group (9.5%). Therefore, this trial showed a substantial reduction in the risk for recurrent thromboembolism among patients who were administered 6 months of anticoagulation. Other recent studies have addressed the potential for prolonged anticoagulation in selected categories of patients. Kearon et al35 randomly assigned consecutive patients to receive 3 months or 2 years of oral anticoagulant therapy following an episode of acute VTE. A prespecified interim analysis led to early termination of the trial after 162 patients had been enrolled for an average of 10 months. Of 83 patients assigned to continue to receive placebo, 17 had a recurrent episode of VTE (27% per patient-year), compared with one of 79 patients assigned to receive warfarin (1.3% patient-year). There was a nonsignificant trend toward a higher risk of (nonfatal) major bleeding in patients assigned to warfarin compared with those assigned to the placebo group.

In a recent Italian multicenter study, 267 patients with a first episode of idiopathic proximal DVT who had completed 3 months of anticoagulant treatment were randomly assigned either to withdraw anticoagulation or to continue for an additional 9 months.36 Of the 134 patients assigned to extended anticoagulation, 21 had a recurrence of VTE (5.0% per patient-year; average follow-up, 37.8 months), compared with 21 of the 133 patients randomized to withdrawal of anticoagulation (5.1% per patient-year; average follow-up, 37.2 months). Four patients had nonfatal major bleeding during extended anticoagulant treatment (3.0%). The results of this study suggest that extending to the course of anticoagulant treatment in patients with idiopathic proximal DVT from 3 months to 1 year is not associated with long-term clinical benefit. These conclusions have been supported by those of a French multicenter clinical trial, which addressed the comparison between 3 and 6 months of anticoagulation in patients with proximal DVT, and that between 6 and 12 weeks in patients with isolated calf DVT.37 The benefit of extending the duration of oral anticoagulation beyond the currently recommended 3-month period in selected patients with clinically symptomatic PE recently has been addressed by another Italian multicenter trial.38

In a multicenter trial addressing the optimal duration of oral anticoagulant therapy after a second episode of VTE, Schulman et al39 found a considerable reduction in the risk for recurrent thromboembolism (from 21 to 3%) in patients allocated to receive 4 years
compared with 6 months of warfarin. Surprisingly, this benefit was offset by a remarkably higher incidence of major bleeding (8.6 versus 2.7%).

In a recent multicenter, double-blind, randomized trial, Ridker et al. demonstrated convincingly that low-intensity warfarin prophylaxis, using a targeted international normalized ratio (INR) of 1.5 to 2.0, is superior to placebo in preventing recurrent VTE in patients with idiopathic VTE who have been treated previously for at least 3 months with warfarin at the conventional level of intensity. Kearon et al. reported simultaneously the results of a different randomized, double-blind trial of similar size that found that low-intensity warfarin (INR, 1.5 to 1.9) was significantly less effective than conventional-intensity warfarin (INR, 2.0 to 3.0) for extended prevention of recurrent VTE, without significant differences in the rate of bleeding complications.

HEPARIN AND ASTHMA

The significant reduction in symptoms at 10 minutes might be related to the ability of heparin to prevent the release of histamine from mast cells. Heparin may interfere with stimulation of mast cell mediator secretion by blocking internal calcium release. Later, heparin may reduce eosinophil recruitment through different mechanisms: by preventing mast cell mediator release, heparin could indirectly downregulate adhesion molecules on ECs, and thus limit eosinophil migration into the nasal mucosa. Furthermore, the heavily anionic heparin may inactivate platelet-activating factor, a cationic protein with a potent chemotactic activity for human eosinophils. Intranasal heparin attenuated the nasal response to an allergic challenge in atopic rhinitic subjects, and no adverse reaction was noted. More studies are needed to explain completely the mechanisms by which heparin produces its anti-inflammatory activity; this will allow us to optimize heparin use in allergic diseases, such as rhinitis and asthma.

The anti-inflammatory activity of heparin has been reinforced by positive, although small, clinical trials in patients suffering from a range of inflammatory diseases, including rheumatoid arthritis and bronchial asthma. In addition, several clinical studies have recently demonstrated the anti-inflammatory activity of heparin in the treatment of inflammatory bowel disease (IBD) at doses that do not produce antithrombotic complications. Given that it is now well recognized that different portions of the heparin molecule exhibit anti-inflammatory activity, and that a pentasaccharide sequence retains the ability to inhibit antithrombin III, it is possible that the anti-inflammatory actions of heparin are distinct from its anticoagulant activity.

HEPARIN/LMWH AND INFLAMMATORY BOWEL DISEASES

Under various experimental and clinical conditions, heparin was found to reduce actively the process of leukocyte recruitment into the site of injury or of application of inflammatory stimuli. Salas et al. provide evidence for the first in vivo mechanism responsible for the antimitogenic action of heparin. In fact, intravital microscopy techniques have allowed direct observation of inflamed microvascular beds with definition of the paradigm of white blood cell extravasation. Leukocyte interaction with the endothelium of an inflamed post-capillary venule is initially intermittent and dynamic (cell rolling); it then becomes static (firm adhesion), and is finally followed by diapedesis. Using the potent cytokine TNF-α to promote this cascade of events in vivo, Salas et al. reported that heparin downregulated TNF-α–induced leukocyte rolling, adhesion, and migration into gut tissue without affecting changes in vascular permeability. These data extend and confirm previous studies in which heparin reduced leukocyte adhesion to vascular ECs in vitro and recruitment of inflammatory cells into other tissues during an experimental inflammatory reaction.

Several uncontrolled studies have suggested that heparin may be potentially therapeutic in the clinical management of both ulcerative colitis and Crohn disease. Although these studies included only a limited number of patients, they demonstrated apparent beneficial effects of heparin with no associated hemorrhagic complications. Heparin has been shown to suppress selected neutrophil functions, such as superoxide generation and chemotaxis in vitro, to reduce eosinophil migration, and to diminish vascular permeability. Among the mechanisms that may account for the anti-inflammatory actions of heparin, binding of this glycosaminoglycan to adhesion molecules expressed on the surface of activated ECs and/or leukocytes has been proposed. Recent in vitro studies have demonstrated the ability of heparin to bind effectively to endothelial P-selectin (but not E-selectin), as well as L-selectin and CD11b/CD18 expressed on neutrophils. Taken together, these reports suggest that the therapeutic actions of heparin observed in patients with IBD may involve attenuation of inflammatory processes as well as a hypercoagulable state associated with clinical exacerbation of IBD; these effects may promote mucosal repair.

Available uncontrolled data show that heparin may be effective in steroid-resistant ulcerative colitis, with a percentage of complete clinical remission of more than 70% after an average of 4 to 6 weeks of therapy. The administration of heparin currently is not justified by the very limited available data. LMWH was used in a single trial in patients with steroid-refractory ulcerative colitis, with results similar to those observed with heparin. Given that a prothrombotic state has
been described in IBD, and microvascular intestinal occlusion seems to play a role in the pathogenesis of IBD, it is reasonable that part of the beneficial effects of heparin in IBD may result from its anticoagulant properties. However, beyond its well-known anticoagulant activity, heparin also exhibits a broad spectrum of immune-modulating and anti-inflammatory properties by inhibiting the recruitment of neutrophils and by reducing proinflammatory cytokines. In conclusion, heparin or heparin derivatives may represent a safe therapeutic option for severe, steroid-resistant ulcerative colitis and other inflammatory disorders, although randomized, controlled trials are needed to confirm these data.

HEPARIN VERSUS LMWH
In contrast to UFH, LMWHs have a lower affinity to bind to plasma proteins, ECs, and macrophages. This difference in binding profile explains the pharmacokinetic differences observed between LMWHs and UFH. The binding of UFH to plasma proteins reduces its anticoagulant activity, which combined with the variations in plasma concentrations of heparin-binding proteins, is reflected in its unpredictable anticoagulant response.

LMWHs exhibit improved subcutaneous bioavailability, lower protein binding; longer half-life; variable number of antithrombin III binding sites; variable glycansaminoglycan contents; variable anti-serine protease activities (anti-factor [F]Xa, anti-FIIa, anti-FXa/anti-FIIa\(^ {Q12}\) ratio, and anti-other coagulation factors); variable potency in releasing TPFPI\(^ {Q13}\); and variable levels of vascular EC binding kinetics.\(^ {66-69}\) For these reasons, during the last decade, LMWHs have increasingly replaced UFH in the prevention and treatment of VTE disorders. Randomized clinical trials have demonstrated that individual LMWHs used at optimized dosages are as least as effective as and probably safer than UFH. The convenient once- or twice-daily subcutaneous (SC) dosing regimen without the need for monitoring has encouraged the wide use of LMWHs. It is well established that different LMWHs vary in their physical and chemical properties due to the differences in their methods of manufacturing. These differences translate into differences in their pharmacodynamic and pharmacokinetic characteristic.\(^ {67}\) The World Health Organization and U.S. Food and Drug Administration regard LMWHs as individual drugs that cannot be used interchangeably.\(^ {67}\)

Bioavailability of LMWHs after intravenous (IV) or SC administration is greater than for UFH and was determined to be between ~87 and 98%. UFH, in contrast, has a bioavailability of 15 to 25% after SC administration. LMWHs have biological \(t_{90}\)\(^ {Q14}\) (based on anti-FXa clearance) nearly double that of UFH. The \(t_{90}\) of LMWHs enoxaparin, dalteparin, tinzaparin, and others has been documented to be between ~100 and 360 minutes, depending on whether the administration of LMWH was IV or SC. The anti-FXa activity persists longer than antithrombin activity, which reflects the faster clearance of longer heparin chains.\(^ {70}\)

LMWH, in doses based on patient weight, needs no monitoring, possibly because of the better bioavailability, longer plasma \(t_{90}\), and more predictable anticoagulant response of LMWHs compared with UFH, when administered SC. Though LMWHs are more expensive than UFH, a pilot study in pediatric patients found SC LMWH administration to reduce the number of necessary laboratory assays, nursing hours, and phlebotomy time.\(^ {71}\)

LMWHs are expected to continue to erode UFH use, through development programs for new indications and increased clinician comfort with use of the drugs. In addition, as both patients and health care providers recognize the relative simplicity of administration with an SC injection, together with real cost savings and quality-of-life benefits by reducing hospital stays, the trend toward outpatient use will continue.

HEPARIN AS AN ANTI-INFLAMMATORY MOLECULE: POTENTIAL MECHANISMS
Heparin is used in the treatment and prevention of thrombotic and thromboembolic conditions such as DVT, PE, and crescendo angina.\(^ {72-74}\) Heparin activates antithrombin III to prevent conversion of fibrinogen to fibrin; it accelerates inhibition of FXIIa, FXIa, FXa, and FXa. Heparin also possesses non-anticoagulant properties, including modulation of various proteases, anticomplement activity, and anti-inflammatory actions (Table 1). Inhaled heparin has been shown to reduce the early phase of asthmatic reaction and suppress allergen-induced increase in bronchial hyper-reactivity. Heparin also inhibits the acute cutaneous reaction due to allergens. The ubiquitous distribution of heparin in tissue spaces may serve to limit inflammatory responses in various tissues where leukocytes accumulate following an inflammatory challenge. It is interesting that heparin is found in high concentrations in the gastrointestinal tract and the lung,\(^ {72,73}\) the two organs exposed to the external environment.

Few studies have reported an effect of heparin on reactive oxygen species (ROS) generation\(^ {75}\) and cytokine secretion\(^ {76}\) by leukocytes in vitro. It has been demonstrated recently that heparin, when injected intravenously, into normal subjects at a dose of 10,000 IU inhibits ROS generation by mononuclear cells and polymorphonucleocytes.\(^ {77}\) Heparin has been also shown, in a series of experiments using N-acetyl heparin, to protect the heart from ischemia-reperfusion injury both in vivo and in vitro, independently of its antithrombin
HEPARIN DERIVATIVES AND ANGIOGENESIS

Angiogenesis is an essential feature of normal biologic processes such as growth, development, reproduction, and repair of damaged tissue.\(^8\) Endogenous promoters and inhibitors regulate the complex process of angiogenesis.\(^8\) The later stages involve proliferation and organization of ECs into tube-like structures. Vascular endothelial growth factor and fibroblast growth factor-2 (FGF2) are important promoters of this process; these promoters exert their effects by binding to cell surface receptors. Human ECs in culture can form tube-like structures with lumens and represent an in vitro model system for the study of the angiogenesis process. Pathologic angiogenesis, which may occur when a normal mechanism is defective, contributes to the growth and metastasis of tumors,\(^8\) as well as to inflammatory and certain ocular diseases.

Drugs that inhibit angiogenesis may be effective in the treatment of these human disorders.\(^8\) The mechanisms by which antiangiogenic drugs exert their effects can vary widely, acting on different points in the complex process of tumor angiogenesis. Potential points of control include blocking the action of endogenous stimulators; inhibiting the growth, migration, and tube formation of EC; and inhibiting the turnover of the capillary basement membrane.\(^3\)

The role of heparin in angiogenesis modulation and its potential anticancer effect has been described previously, but without clear delineation of its pro- versus antiangiogenic effect as well as its mechanism of actions.\(^6\) The LMWH tinzaparin demonstrated potent inhibition of angiogenesis.\(^8\)

Tinzaparin, a LMWH (average molecular weight, 6.5 kd), produced by enzymatic degradation of heparin, has proven efficacy in the treatment of DVT and PE.\(^8\) The antithrombotic activity of tinzaparin is mediated by binding to microvascular tissues and activation of antithrombin III, a potent anticoagulant.\(^9\) Tinzaparin also causes release of tissue factor pathway inhibitor (TFPI)\(^16\), an important endogenous inhibitor of tissue factor (TF)/FVIIa\(^17\). Several clinical trials have shown improved survival of cancer patients following heparin therapy.\(^9\)–\(^15\) In one double-blind, multicenter clinical trial, tinzaparin was shown to be effective in the treatment of proximal DVT in a patient population that included a large percentage of cancer patients.\(^8\) These clinical data suggest that tinzaparin may have some benefit in the treatment of cancer patients. The present study was undertaken to elucidate the mechanisms through which tinzaparin may affect tumor angiogenesis and to assess its efficacy in inhibiting the angiogenesis processes.

LMWH AND TFPI INHIBIT EC TUBE FORMATION

A pivotal stage of angiogenesis is the formation of tube-like structures from ECs,\(^8\)–\(^8\) a process that is mediated by cytokines binding to EC surface receptors. In addition, Mousa and Mohamed\(^8\) reported that the inhibitory efficacy of tinzaparin or r-TFPI on FGF2-induced EC tube formation was totally reversed by a specific monoclonal TFPI antibody directed toward TF/FVIIa but not FXa\(^18\) binding domain. Similar data were shown for this antibody in reversing the inhibitory effect of tinzaparin or r-TFPI on TF/FVIIa\(^19\)–induced EC tube formation. Furthermore, additional reports demonstrated that the antiangiogenic and antimetastatic activity of LMWH to be independent of anticoagulant activity.\(^9\)

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