Heparinoids: Integrated Control of Hemostasis and Metastasis

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Malignant transformation is characterized by two major cellular manifestations: a) impaired regulation of cell growth, survival and differentiation; and b) aberrant interactions of the cells with their microenvironment, including proteins and glycosaminoglycans of the extracellular matrix. Heparinoids are complex polysaccharide glycosaminoglycans composed of repeating disaccharide units of iduronic acid and glucosamine. Their derivatives, produced and processed by various cell types, are utilized in clinics as anti-coagulants. This review summarizes the involvement of heparinoids in the regulation of molecular events associated with malignant transformation, and the effects of their clinical use on the outcome of malignant diseases. This combined overview may point to possible future directions in the development of novel cancer therapeutics based on the application of heparinoids.

The effects of heparinoids on the clinical outcome of cancer

Heparin is often used in oncology to control the hypercoagulopathy that commonly accompanies malignancy [1]. It is a potent treatment for venous thromboembolism that complicates the course of disease [2]. It is also effective for the prevention of thrombosis after cancer surgery and thrombosis due to venous access catheters used to administer chemotherapy and supportive treatment [3,4]. Heparin has been used successfully to treat the thrombotic complications of bone marrow transplantation and for symptomatic disseminated intravascular coagulation associated with cancer [5–7]. Less common uses of heparin in cancer management indicate certain effects that are unrelated to its anticoagulant properties. For example, the use of heparin for treating symptoms of microvascular damage after radiation therapy, or to stimulate recovery of platelets after bone marrow transplantation [8,9].

Heparin may also affect *in vivo* tumor growth [10,11]. Several studies in experimental animals have suggested that heparin suppresses tumor growth. Goerner in 1930 [12] first reported the inhibitory effects of heparin on tumor growth in experimental animals. Subcutaneously implanted fragments of the Flexner-Jobling carcinoma grew and eventually led to the animals' deaths. By contrast, tumor fragments incubated with heparin before implantation failed to grow, and all animals remained alive at 3 months. Many other studies in animal models revealed effects of heparin on tumor dissemination [11,13].

The first report of unfractionated heparin administration to humans for purposes of treating malignancy itself was that of Albert-Weil and Nehorais in 1954 [14]. They reported on two cancer patients treated for 3 and 4 months respectively with daily 50 mg intravenous bolus injections of UFH, and for 1 month with daily intramuscular injections of leech extract. The first patient had complete tumor regression at the end of the course of treatment but died eventually of uremia. The second patient had a reduction of the tumor mass and improved overall well-being and was alive at the time of the report. These authors cited evidence for heparin inhibition of cell proliferation and postulated that both heparin and leech extract worked by inhibiting coagulation and proteolytic enzymes [14].

The first prospective study on the effects of heparin on cancer outcome was conducted by Lebeau et al. [15]. They performed a multicenter, randomized trial of adjusted-dose subcutaneous UFH administered daily for 5 weeks compared to no heparin in patients with small cell lung carcinoma receiving combination chemotherapy. The heparin-treated patients, especially those with limited disease, experienced significantly improved complete response rates and median survival [15]. Another prospective study [16] reported an adjuvant treatment in 304 patients with resectable colon cancer. A control group was treated with surgery alone. The treatment group was treated postoperatively with intraportal infusion of 5-FU plus 5,000 U of UFH daily for 7 days. The risk of developing subsequent liver metastasis was significantly reduced in the 5-FU/heparin group as compared to the untreated controls ($P < 0.01$). The relative contribution of 5-FU and UFH could not be determined [16]. A retrospective study [17] reported on 1,358 consecutive medical patients admitted to hospital for reasons other than VTE. Eligible patients were randomized to receive either low dose subcutaneous UFH thromboprophylaxis (5,000 U twice daily) or no heparin. Overall hospital mortality was reduced in the heparin-treated group ($P < 0.05$). In the subset of patients with malignancy overall mortality was 18.6% for heparin-treated patients versus 32.1% in the control patients (no statistical analysis provided). There is a potential bias because of the possibility that heparin protected against VTE death, but it does not rule out other effects of heparin [17]. Other retrospective analyses in patients who underwent tumor resection compared UFH thromboprophylaxis versus no treatment. Survival, overall risk of recurrence and time to

UFH = unfractionated heparin
VTE = venous thromboembolism

progression were better in the heparin-treated group, but it did not reach statistical significance [18,19].

The use of low molecular weight heparin is expanding rapidly. The relative ease of administration, reduced need for monitoring, and equivalent or superior safety and efficacy profiles compared with UFH account for this expansion [20]. The primary objective of most studies reported to date was to compare LMWH and UFH for the treatment of VTE. In fact, a number of meta-analyses of clinical trials comparing LMWH with UFH for venous thromboembolism have shown interesting effects in the subgroup of patients with cancer complicated by thrombosis. Data from these meta-analyses suggest that a survival benefit exists for cancer patients who receive LMWH compared to UFH [21–23]. Of even greater interest is the fact that this reduction in mortality may not be attributable to a reduction in fatal VTE, but rather to an inherent anti-tumor activity [23,24].

Prandoni and co-workers [25] reported subgroup analyses of cancer patients treated with LMWH versus UFH for deep vein thrombosis. Their findings showed that VTE patients with cancer treated with UFH had a mortality rate of 44%, whereas in the LMWH-treated patients the rate was 7% ($P = 0.021$).

One prospective trial in randomized cancer patients represents progress in this line of research. Von Tempelhoff et al. [26] conducted a study to determine specific survival rates among women with untreated breast and pelvic cancers undergoing surgical resection. Patients were given thromboembolic prophylactic doses of either LMWH (certoparin 3,000 U/day) or UFH (5,000 U, three times per day) for 7 days postoperatively. Follow-up at 650 days showed that among patients who had received the LMWH the mortality rate was 5.7% compared with 15.6% in those receiving UFH ($P = 0.005$). This difference was no longer apparent after about 3 years ($P = 0.136$) [26].

Regulation of cancer cell adhesion, signaling and motility by heparinoids

Metastasis is a multistep process, initiated by scattering of cancer cells from their tissue of origin and migration of some of them through blood vessels and lymphatics to form metastases at distant organs [27]. Biochemical signaling induced by several growth factors, specifically defined as scatter factors (e.g., hepatocye growth factor), results in reorganization of the actin cytoskeleton, and the dynamic turnover of cell-ECM adhesion sites, leading to cancer cell locomotion [27,28]. The motility-inducing signal is transduced into the cytoplasm via the combined activity of several growth/scatter factor receptors and transmembrane heparinoids, predominantly heparan sulfate proteoglycans (e.g., syndecans), and may be inhibited by soluble heparinoids [10,29] [Figure 1-I]. The motility-associated biochemical signaling also regulates cell adhesion, which is a crucial factor in tumor invasion and metastasis. Cell adhesion is mediated by complex multimolecular assemblies, collectively called adhesion complexes, which establish and regulate the contact between cells and the ECM. Adhesion complexes have a wide range of functions in cells – from mechanical arrest along blood vessel wall and extravasation, to transmission of forces during cell migration, or the transduction of transmembrane signals. Transmembrane HSPG are emerging as molecules that regulate cell adhesion and control cell shape, adhesion, proliferation and differentiation [30]. Cell surface HSPG may bind ECM components (e.g., fibronectin, laminin) directly, or in cooperative interactions with integrins, thereby linking cancer cells to the endothelium and the subendothelial basement membrane [Figure 1-II]. The cytoplasmic tail of membrane HSPG associates with PIP_2 and PKC, localizes them to forming adhesion sites and regulates PKC activity [30]. Laminin may stimulate cell motility and scattering in an integrin-independent mechanism, which can be blocked by heparin [31]. While cell-surface HSPG modulate cell-ECM adhesive interactions, the assembly and maintenance of the subendothelial basement membrane depend on the cross-linking of its components (e.g., laminin, collagens) by extracellular HSPG. Therefore, the heparan sulfate-degrading enzyme heparanase disintegrates the subendothelial basement membrane, facilitating the trans-endothelial migration and metastatic spread of malignant cells [32]. A causative link between heparanase expression and metastasis was demonstrated in several experimental systems [32]. In addition, heparanase expression was correlated with poor patient survival in several cancers. Eldor and Vlodavsky [33] established that soluble heparinoids inhibit the *in vitro* heparanase enzymatic activity and significantly reduce metastasis in experimental models [Figure 1-III]. Thus, soluble heparinoids may

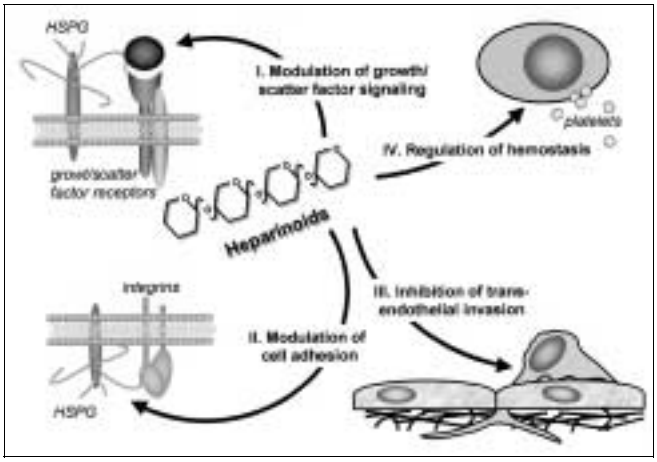

Figure 1. Heparinoids may impair molecular mechanisms associated with metastatic cell dissemination. Cancer cell motility is triggered by trans-membrane biochemical signaling, and involves cell-ECM adhesive interaction. Soluble heparinoids may compete with membrane-associated heparan sulfate proteoglycans that participate in scatter/growth factor signaling complexes **[I]**, or membrane adhesion complexes **[II]**. Components of the coagulation systems (e.g., platelets) associate with metastatic cells and enhance their invasiveness. These interactions may be perturbed by heparinoids **[IV]**. Heparinoids inhibit the activity of heparanase that promotes the trans-endothelial migration of metastatic cells **[III]**.

LMWH = low molecular weight heparin
ECM = extracellular matrix

HSPG = heparan sulfate proteoglycans

obstruct several stages of the adhesive interactions of cancer cells with the ECM, including adhesion, signaling and invasion, thereby restraining metastatic cell dissemination [Figure 1].

Hemostasis and metastasis

Components of the coagulation mechanism may be involved in cell adhesion and motility in addition to their well-defined roles in the regulation of hemostasis. Thrombin, at concentrations that precede fibrin formation, is a potent inducer of tumor cell expression of various integrins and tumor cell adhesion to the matrix and other activated cells [34]. Tissue factor, the cell-surface receptor of factor VIIa, may form a transmembrane link between the actin cytoskeleton and extracellular enzymes of the coagulation system [35]. Thus, tissue factor may promote the formation of proteolytic enzyme complexes on the surface of cancer cells, which may contribute to their motility and invasive potential [35,36]. Moreover, circulating metastatic cells may form aggregates with platelets [Figure 1-IV]. Eldor and Vlodavsky [33] showed that platelets may facilitate trans-endothelial migration of tumor cells. Heparin treatment attenuates tumor metastasis in mice by inhibiting P-selectin-mediated interactions of platelets with carcinoma cell-surface mucin ligands [37]. Platelet aggregates may protect circulating metastatic cells from immune cells and provide them with enzymes (e.g., heparanase) essential for trans-endothelial migration. Therefore, heparinoids used to regulate hemostasis in cancer patients may exert anti-cancer effects via inhibition of the molecular mechanisms of cell adhesion and motility [Figure 1].

Conclusion

Cumulative clinical evidence suggests that heparinoids may have anti-tumor effects in human malignancy, with an apparent improvement in cancer outcome in LMWH-treated patients. During recent years several molecular mechanisms were defined that may account for the beneficial effects of heparinoid treatment on the clinical outcome of malignant diseases. Heparinoids may exert several anti-cancer effects, including the modulation of biochemical signaling, cell adhesion and motility. Treating cancer patients with heparinoids may provide clinicians with an integrated therapeutic mode to control both hemostasis and metastasis [38].

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Silence – one of the hardest arguments to refute

Anonymous

Capsule

Protecting the heart

Within the mitochondria of heart cells, ion channels control the flux of different ions and change the physiologic status of the mitochondria, which in turn affect the relative health of the heart cell. Xu et al. describe the role of a calcium-activated potassium channel in the inner mitochondrial membrane of guinea-pig heart cells in protecting the cells from ischemia. A drug that opened the channel could protect the heart from infarction.

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Capsule

Eluting stents revolutionize heart care

A step up from the widely used bare metal stents for treating heart disease, the new drug-coated stents prevent reclogging of the arteries. In a multinational clinical trial, including Israel's Hadassah and Rambam Medical Centers, the new version of the traditional stent has so far shown much lower rater of reclogging or restenosis. New clinical data are expected to further confirm the previous results. The three most prominent stents in the market are those coated with sirolimus, rapamycin, and paclitaxel.

In Israel, the Cypher, a sirolimus-eluting stent produced by Johnson and Johnson, has been approved for use by the Ministry of Health but is not included in the basket of reimbursable items. Hospitals will allow interventional cardiologists to insert the stent on condition that the patient purchases the item (which costs $2,600). Johnson & Johnson have arranged that the Lavie company will supply the stent to the catheterization laboratory.

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