Proteoglycans and glycosaminoglycans in tumor growth and migration: first experience with tumors of bladder and prostate origin*

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Summary. Proteoglycans (PGs), macromolecules that contain bound glycosaminoglycans (GAGs), are an abundant and ubiquitously distributed group of proteins with a large variety of heterogeneous structures. In recent years a whole range of functions, varying from structural/mechanical support to involvement in the regulation of cell proliferation and cell mobility, cell-cell interactions, and angiogenesis and modulation of the extracellular matrix, have been attributed to these compounds. In this communication an overview is presented dealing with the available knowledge of the (regulatory) properties of PGs and GAGs as participants in tumor growth and metastasis. Acquaintance with the biochemical, biophysical, and biological characteristics of PGs and GAGs may provide a rational basis of the therapeutic use of GAGs in the prevention of tumor growth and invasion. The scarcely available information related to the role of PGs and GAGs in uro-oncology and the potential application of GAGs in the treatment of urological tumors are discussed.

Tumor metastasis is an insidious character of malignancy and considerably affects the prognosis of cancer patients. Improvement of surgical procedures and adjuvant treatment modalities for local tumors have meant that the prognosis of the majority of patients depends largely on the release of cells by the primary tumor or the occurrence of metastasis. Thus, at the time of diagnosis, up to 30%-40% and 40% of patients with prostatic and muscle-invasive disease, respectively, have developed metastases. The metastatic cascade involves a multitude of sequential steps, and basic and clinical research intends to unravel and manipulate these steps at need. After the cellular development of transformation and metastatic potential, as the result of an accumulation of a multitude of genetic (oncogenes and tumor-suppressor genes) events and reflected by an uncontrolled proliferation and migration ability [12], three major phases are generally recognized (Fig.1): (1) during the intravasation phase, prior to their release into the circulation, cells lose their cell-cell interaction and pass through several connective-tissue barriers, including the epithelial basement membrane [41, 69], the interstitial matrix [22, 49], and the endothelial basement membrane; (2) dissemination and embolization of small tumor aggregates in the blood stream or, alternatively, a lymphatic channel; followed by (3) the extravasation phase, during which surviving cells arrest in the capillary beds of (specific) target organs, lodging to the vascular endothelial cells, penetrate the endothelium and the basal lamina, and proliferate in the parenchyma of the organ. Finally, initiation and progression of neoangiogenesis occurs [3, 13, 34-36].

The list of factors and properties intrinsic to both tumor and host that influence the development and outgrowth of metastases is rapidly extending [13]. Recently, increasing knowledge of the basic biochemical structure and several crucial, cell biological observations have contributed to a considerable interest in proteoglycans (PGs) and glycosaminoglycans (GAGs) in tumor biology [20, 29, 42, 54, 55].

The purpose of this communication is to summarize recent progress in identifying the role of PGs and GAGs in the regulation of cell growth and motility. The involvement of PGs and GAGs in the expression of uncontrolled cell proliferation and migration is a relatively new field in tumor biology and has hardly been introduced in uro-oncological research. Current understanding of the biochemical structures of these compounds is not treated in detail in this paper, since recent reviews are available that offer an excellent entry into the relevant literature [20, 26, 30, 53]. Special attention is given to the (potential) clinical benefit of introducing GAGs into the system of tumorbearing hosts for the treatment and/or prevention of metastasis.
**Proteoglycans and glycosaminoglycans**

*Biochemical structure and synthesis*

PGs are complex macromolecules, each of which contains a core protein with one or more covalently bound GAG chains. GAGs are linear and/or branched polymers of repeating disaccharides that contain one hexosamine and either a carboxylate or a sulfate ester or, usually, both [11]. These simple definitions encompass an exceptionally large range of structures involving different core proteins, different classes of GAGs [hyaluronic acid (HA), chondroitin sulfate (CS), dermatan sulfate (DS), keratan sulfate (KS), heparan sulfate (HS) and heparin], and different numbers of individual GAG chains. PG metabolism in most cells is a strictly regulated, dynamic process that contributes directly to cell and tissue function. The half-life of PGs in the extracellular matrix (ECM) can range from a few days to a few weeks, whereas cell-surface PGs generally have half-lives of only a few hours. The core protein of PGs is synthesized in the rough endoplasmatic reticulum (RER). The N-asparagine- and O-serine/threonine-linked oligosaccharides are synthesized in the RER and Golgi stacks. HA synthesis is not localized in the Golgi system; rather, it appears to occur in a compartment associated with the cell surface [11, 21]. The size and ratio of the core protein-linked GAGs may change with development, aging, or disease.

*Structural/protective functions*

PGs are abundant and ubiquitously distributed and the different types of core proteins are reflected by a variety of locations. PGs are found inside cells, on the cell surface, and in the ECM. The (protective) role of PGs (and free GAGs) at luminal surfaces of the endothelium and bladder wall have been extensively reviewed elsewhere [27, 40, 42] (Hurst, this issue). Currently the research effort on PGs, major constituents of the ECM, is highly focused on their role in the ECM, both structurally mechanically and in the control of cell proliferation and motility. The large structural diversity no doubt contributes to a whole range of biological functions. The ECM contains large PGs such as perlecan, aggrecan, and versican and the small PGs decorin and fibromodulin.

The polysaccharide chains of PGs are highly hydrophilic and are too inflexible to be capable of folding up into compact globular structures. They occupy a huge volume relative to their mass, and their high density of negative charges attracts a cloud of cations such as Na⁺ and K⁺ that are osmotically active, causing large amounts of water to be sucked into the ECM [55]. This creates a swelling pressure, or turgor, that enables the matrix to withstand compressive forces, in contrast to the fibrils, which resist stretching forces. The content of GAGs in connective tissue usually amounts to less than 10% by weight of the amount of fibrous proteins. GAGs form porous, hydrated gels and their chains fill most of the extracellular space, providing mechanical support to tissues while allowing the rapid diffusion of water-soluble molecules and the migration of cells. Unlike the other GAGs, HA is not bound to a protein core and is hence classified simply as a GAG. In some tissues, particularly during early development, HA can constitute the major structural macromolecule in the ECM, where it can promote both cell proliferation and migration [74]. The organization of the large HA molecules requires considerable folding, which probably involves specific interactions with other structural macromolecules present in the matrix. For instance, the G1 domain of aggrecan, the large CS/KS PG