Adhesion molecules and cancer

Significant functions of the multicellular organisms such as tissue formation and development, orientation and structural maintenance, the migration of leukocytes, blood coagulation, would healing, and inflammation, but also the control process of tumor invasion, metastasis and differentiation, are mediated by adhesive interac-

tions occurring either between cell (cell-cell) or between the cell and components of the extracellular matrix (cell-matrix).[1,2] The mediating molecules of these interactions are named adhesion molecules,[3] and they mediate either in cell-cell (cell-cell adhesion molecules, CAMs) or cell-matrix interactions (substrate adhesion molecules, SAMs) and some may act in both types of interaction. The past decade has been characterized by increased knowledge about the genetic, biochemical and functional properties of these molecules. Nowadays more than a hundred adhesion molecules have been identified. The importance of these molecules in health and disease is remarkable, because they determine whether a cell persists, if it is going to detached, if it migrates and finally when it stops. The concept of the adhesion forces holding the cell together, and of the decrease of those forces allowing a wide spread of the cells is not a new notion. It was initially introduced by Coman in 1944,[4] but Paget in 1889 had already addressed the question, "what is it that decides what organs shall suffer in a case of disseminated cancer?"[5] today, a century later, the issue remains unresolved. However a wealth of studies has added new facts regarding the adhesion molecules and the role they play in normal circumstances, in tumorigenesis and in metastasis. The molecular basis of the neoplastic process is better perceived as time passes. *In vitro* and *in vivo* studies helping in the understanding of the way adhesion molecules function in carcinogenesis and the biological behaviour of tumors in general, contributing to prompt diagnosis and potentially in the field of treatment. Complex and coordinated reductions and increases in adhesion have been proposed to be necessary in order of tumor invasion and metastasis to take place. This hypothesis has fueled the interest of cancer research teams in evaluating the expression of various adhesion molecules in a wide range of human malignancies in the hope of pinpointing some of the cell adhesion alterations underlying tumor behaviour. Today the use of monoclonal antibodies directed selectively against certain adhesion molecules provides the possibility of studying the majority of human cancer cases. Both basic and applied research are attracting the increasing concern of many scientists.

At present five families of adhesion molecules are known to exist, comprising the integrins, cadherines, the immunoglobulin supergene family, selectins and CD44.[6] These molecules are biochemically and genetically unique although in some instances they have related roles. Qualitative and quantitative changes in the expression and function of adhesion molecules have now been indentified in the majority of human tumors using *in vitro* experimental model systems and *in vivo* immunolocalization studies. The hallmarks of malignancy can now be explained, at least partly, by alterations in the adhesive properties of the neoplastic cells. Additionally, interest is focused on the role of adhesion molecules as potential biomarkers in malignancy for use in screening, in early and differential diagnosis, and in the assessment of prognosis as well as for the monitor of disease progression.

Neoplastic transformation results from the loss of normal controls over the growth and differentiation of cells; once transformed the cells require a reduction in adhesiveness to detach themselves. To achieve migration, the affinity between cells and endothelium of lymphatic channels needs to change. Further changes in expression of adhesion molecules, by both the target and the invading cell, are necessary for a cell to attach to a particular target organ. A prerequisite for cells to form a metastasis is an increase or re-expression of intercellular adhesion molecules, coupled with a capacity to grow independently.[7]

Several laboratories have reported experiments in which dissemination of intravenously injected tumor cells in tissues has been inhibited by the simultaneous injection of a peptide containing the protein sequence recognised by many integrin molecules as their ligand. These observations have caused special interest on the part of clinicians that integrin protein homologs which encompass the RGD sequence (arginine, glycine, aspartic acid) may be useful in cancer treatment. The loss of adhesion resulting from the peptide injection may deny the cells anchorage and traction for growth and migration.

Adhesion molecules also transduce signals in and out of cells. This phenomenon is often refered to as "inside-out" signalling. Thus, adhesion molecules are thought to involve conformational alterations affecting morphological differentiation, gene expression and cell motility.[8] The control of cellular morphology through linkage of the adhesion molecule tail to the cytoskeleton has provoked much interest. As an inverse relationship exists between growth of the tumor and morphological differentiation in many human cancers, induction of a differential phenotype by the promotion of the activity of cell adhesion molecules has been promoted as a mode of treatment. The invasive and metastatic potential of neoplastic cells falls when they are subjected to treatment by blocking with monoclonal antibodies or by genetic manipulation with cDNAs or antisense RNAs. Such find-

ings provide another perspective on the metastatic cascade. The process is, however, much more complex and other modifiers of biological responses clearly have some degree of interplay with adhesion molecules.[9]

In clinical practice, cell adhesion molecules may serve as selective markers for some tumors, which could be useful for either diagnosis or prognosis. The location of these molecules or their neoplastic isoforms on the surface may facilitate the use of specific monoclonal antibodies coupled with drugs or radioactive markers in tumor imaging and delivery of drugs.[10] Advances in the understanding of how the function of adhesion molecules is controlled may lead to their future incorporation into treatment strategies. The majority of integrins are SAMs, prime mediators of cell-matrix interactions, functioning as receptors for proteins including collagen laminin and fibronectin.[8] Some of the integrins have overlapping ligand specificities. Thus $\alpha 1\beta 1$ binds laminin or collagen, $\alpha 6\beta 1$ binds laminin and $\alpha 3\beta 1$ binds laminin, collagen and fibronectin. Some even show alternative specificities when expressed on different cell types. Other integrins, primarily found on leukocytes, can function as CAMs. These integrins mediate heterotypic cell-cell adhesion by binding to some members of the immunoglobulin supergene family, such as the intercellular adhesion molecule-1 (I CAM-1), I CAM-2 and the vascular cell adhesion molecule (VCAM). Other integrins, e.g. $\alpha m\beta 2$ (MAC-1), may function both as SAMs and CAMs.[6,8] Integrins are also capable of binding ligands such as thrombospondin, fibrinogen and vitronectin, and because of this they are implicated in inflammation, wound healing and development processes. In addition, it seems that integrins play a central role in cell adhesion and migration.[11] Alterations in integrin expression are seen *in vivo* in many epithelial malignancies, including carcinoma of the lung, breast, colon, prostate, stomach, pancreas, kidney and skin. Clinicopathological studies have shown an association between integrin expression and tumor staging.[12,13] The evaluation of integrin expression, together with other markers, may help to identify the malignant potential of low grade tumors. The $\beta 1$ integrin show characteristic distribution patterns in solid tumors with a cell lineage dependent distribution.

Cadherins are transmembrane glycoproteins which are prime mediators of cell-cell adhesion via calcium dependent homotypic interactions. The most important cadherins are the epithelial E-cadherin, the placental P-cadherin, the neural N-cadherin, the vascular-endothelial VE-cadherin and the retinal R-cadherin. The cadherin molecules require the presence of calcium for their configuration and binding to their ligands. Cadherins are rapidly degraded by protease action in the absence of the calcium ion. Cadherins are the most important of all adhesion molecules and when they are expressed, the inactivation of the other CAMs has little effect.[14] E-cadherin and N-cadherin participate in the formation of adherence junctions. In 1989 Eidelman showed that there was a total disappearance or down-regulation in E-cadherin expression in tumors of low grade differentiation. In 1993 a number of studies confirmed that the low expression of E-cadherin is a general phenomenon occurring in a variety of human tumors, as breast carcinoma, colorectal tumors, prostatic adenocarcinoma, bladder tumors, pancreatic tumors and squamous carcinoma of the head and neck. It is known that in cervical intraepithelial neoplasia as well as in invasive squamous cell carcinoma, there is a loss of normal surface E-cadherin expression that correlates directly with the degree of dysplasia.[15] This could be of additional help in grading of cervical intraepithelial neoplastic lesions and/or in the definition of their invasive potential.

The cadherin function is regulated via the -COOH terminal intracellular domain by α, β and γ catenins which interact with cytoskeletal elements of the cells with which they bind.[16] A recently identified protein p120 has a role similar to that of catenins and therefore regarded as a new catenin molecule.

The adhesion molecules of the immunoglobulin supergene family have in common the immunoglobulin structural homology unit, which consists of a length of 70 to 110 amino acids forming β-pleated sheets.[17] This family includes adhesion molecules which are considered to be important in cell adhesion, such as the neural cell adhesion molecule (NCAM), the VCAM, the ICAM-1, the carcinoembryonic antigen (CEA) and the product of the deleted colon cancer gene (DCC). NCAM is expressed in a wide variety of cell types, mainly of neural and mesenchymal origin. NCAM is present in a range of neural and mesenchymal and neuroendocrine tumors, including Wilm's tumor, pituitary adenoma, pheochromocytoma and small cell carcinoma of the lung. V-CAM serves as a receptor for $\alpha 4\beta 1$ integrin, i.e., VLA-4 which is found in leukocytes, but can also be expressed in malignant melanoma.

Selectins are transmembrane glycoproteins with a N-terminal C-type lectin domain. They also have a varying number of complement-like repeats, a single EGF-like repeat (epidermal growth factor), and a short cytoplasmic

domain. The lectin domain binds to fucosylated and sialylated carbohydrates on adjacent cells. Selectins mediate heterotypic interactions between blood cells and endothelial cells during lymphocyte homing and leukocyte adhesion in the inflammatory process.[18]

CD44 is a cell surface glycoprotein important in lymphocyte homing, T-cell activation, and adhesion to hyalouronate and matrix proteins; mRNA splice variants exist and as many as nine isoforms of the CD44 molecule have been described.[19]

In conclusion, the implication of adhesion molecules in many normal processes as well as in various pathological conditions and mainly in tumorigenesis, has been researched in detail during recent years. The study of these molecules helps in the understanding of the underlying mechanisms of malignant transformation and uncontrolled proliferation of cancer cells. Some of these molecules have already been used as biomarkers in the diagnosis and prognosis of many malignancies. Following observations that the integrin protein homologues with the RGD sequence may inhibit the metastatic process in mice, special interest has been aroused over the possible use of such substances in cancer treatment. Peptides conjugated with drugs may be directed selectively to certain neoplasmatic cell sites. The integrins via metalloproteinases may have therapeutic applications in the near future. The increased interest all over the world regarding the adhesion molecules and cancer will possibly generate useful data on the therapy procedure of this great problem of humanity, named cancer, which was referred to by Hippocrates long before the birth of Christ.

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