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Commentary

It's the Matrix!

ECM, Proteases, and Cancer

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The involvement of the extracellular matrix (ECM) with cancer is a long-standing topic of investigation. Investigators have long recognized the dynamic nature between cell growth and cellular interaction with the ECM.[1] In this issue of *The American Journal of Pathology*, DeClerck et al report the discussions of a workshop organized by the Path B Study Section of the National Institutes of Health in October, 2002.[2] The discussions centered around four basic areas: epithelial-mesenchymal transformation (EMT), galactin(s), redefining ECM-degrading proteases, and balance between proteases and protease inhibitors in angiogenesis.

The discussions at this meeting are both exciting and frustrating. Exciting as they present new ideas and directions in several areas but frustrating in that data are not shown making analysis difficult. In some areas topics are approached from a narrow viewpoint. Understanding how the ECM participates in the abnormal growth is fundamental to understanding cancer. The purpose of this commentary is to add to the discussion on the role of the ECM and, perhaps expand the concepts that were discussed at this meeting.

The involvement of the ECM in both normal and abnormal growth is a complex topic involving cell-cell and cell-ECM interactions but equally important are the physiological parameters including the dynamics of cell function (migration, adhesion), the role of the microvascular system in the movement of material in the extracellular environment, and the mechanical properties of growth.[1,3] It is known that there are important biochemical cascades of signaling proteins inside the cells that govern cellular reaction. However, biochemical cascades occur in the ECM which are also critical to the regulation of cell growth. After all, a fundamental property of living systems is to respond to external stimuli. The ECM is like a biological reservoir that contains many components that provide a variety of functions to the enveloped cells. It is known that some of the large molecular weight glycoproteins and proteoglycans can signal biological function as well as be structural molecules. Clearly, these molecules contain signaling sequences within their structure. Growth factor repeats, arginine, glycine, aspergine (RGD) sequences, and other signaling sequences may be "released" by proteolytic action; yet how the cascade of different proteases and their inhibitors in the ECM might regulate the release of this information is not clear. Extracellular proteases have been shown to be essential to cell transformation, migration, and differentiation. Yet the underlying mechanism(s) remain elusive. Is it the cleavage and release of important sequences from these ECM molecules, the shedding of receptors during change in cellular phenotype, or the release of mechanical tension that provides the essential signals to cells?[4,5] It may be all, some, or none of the above.

Data generated by microarrays indicate that many of the ECM components and their receptors undergo change during normal and abnormal growth. These data indicate that the quantitative shifts in expression of proteins within these cascades seem to spell the difference between normal and abnormal. However, it is difficult to use these data under current experimental paradigms. The discussion on ECM, Proteases, and Cancer, like an array, point to the direction of new experimentation, but we are just not sure which way.

The discussion in the section on EMT emphasizes that the loss of cell-cell and cell-ECM contacts must be modified in order for transformation to occur. New cell contacts must be established with the ECM that results in altered migration and transformation. Clearly this is a complex reaction which requires multiple steps. EMT is particularly difficult to study in *in vivo* cancer models, as the process is believed to be slow as compared to *in vitro* models.[5] A discussion of culture models by Mercurio's group is interesting, as different culture models including organoids have been used to investigate this transforma-

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tion.[6] In these studies, cytokines and growth factors were involved in the alteration of expression of the integrin $\alpha 6\beta 4$. Beside TGFβ, the VEGF pathway was upregulated. The speculation that a specific integrin may have the ability to regulate protein translation and influence tumor survival is potentially important; however, it is difficult to assess as there are likely many other signals feeding into the pathways. These studies do stress the importance of looking at the quantitative differences in signaling of many factors and their receptors.

The relationship of EMT in tumors and development was well documented in a recent review.[5] This review pointed out that the fundamental processes of EMT that are essential to embryological development are likely similar to tumor EMT. Expression of transcription factors, growth factors, cell-cell molecules such as cadherins, and signaling pathways occur in both normal and abnormal developmental systems. What appear to be significant are the quantitative differences between normal and abnormal levels of these factors. Analyses by microarrays have shown that many of these factors and ECM proteins are potential candidates in the regulation of EMT.[8–10] The analysis of EMT in tumors as an alteration of development seems to be a productive avenue of investigation.

In the second discussion, galectin's role in tumor progression was emphasized in various tumor models. These studies, based on transfections and altered adhesion, indicate the importance of carbohydrate moieties, especially in the ECM. While these reported studies are interesting, they also leave the question of *in vivo* functions unresolved. However, the potential importance of this family appears critical. Galectins are involved in a wide a variety of processes including cell adhesion, migration, differentiation, apoptosis, and clearance of interstitial fluids.[11,12] The importance of galactins is part of a larger role for large glycoproteins and proteoglycans in the ECM. These macromolecules play numerous roles including binding growth factors, latent proteases, and the regulation of interstitial pressure. The latter appears to play a significant role in chemotherapeutic approaches[13,14] as well as metastasis.

In the final chapters, the role of ECM-degrading proteases and their specific inhibitors are discussed in some detail. Early reports have shown that there are analogies between cancer growth and wound healing.[15] In the ECM, there appears to be a proteolytic cascade which is involved in clearance of molecules from the ECM, interstitial fluid dynamics, and turnover of individual ECM components as well as remodeling of tissues. It is clear that these proteases are critical to our understanding of tumor growth and represent sites of potential regulatory intervention. The discussion of how matrix metalloproteasess (MMP) play a role in neovascularization and tumor invasion is especially good. The dynamic interaction between the protease and specific (and perhaps) non-specific inhibitors points out that these components may be multifunctional. Yet just how the various types of inhibitors and their ECM substrates may be regulated is not clear.

Finally, one of the important aspects of proteases and consequently their inhibitors may not only lay in their action on ECM component(s) but what the degraded components signal.[5] As a protease attacks a particular substrate, the products of the degradation may also signal other cellular responses or interactions with cell surface receptors. As remodeling takes place, the removal of these products from the ECM may also be important. This concept comes back to the physiology of the tumor (or normal tissue). The remodeling process in tumors is critical for creation of space for cellular growth, signaling for angiogenesis, and fluid dynamics of microcirculation.

These reports from this meeting will certainly promote discussion on the topic of the role of the ECM in cancer. The concept of the ECM as an integral component of the growth of tumors is critical to our understanding of cancer and will be part of the therapeutic approaches to the regulation of abnormal growth. After all, IT'S THE MATRIX!

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