Cell Biology:

Enhanced: Stretching Is Good for a Cell

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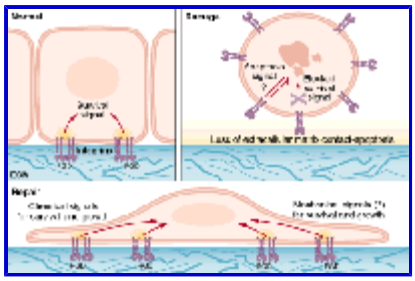
Cells in tissues are attached to one another and to the fibrillar protein meshwork known as the extracellular

matrix (ECM). Most cells require this attachment and subsequent spreading on the ECM substrate for proper growth, function, and even survival. Without it, they often die by undergoing apoptosis, or programmed cell death (1). [HN2], [HN3] This dependence of cell growth and survival on substrate attachment is known as anchorage dependence; the apoptosis resulting from lack of anchorage has been named "anoikis." [HN4] Only the cells circulating in the blood are designed to survive without attachment and spreading. (Some tumor cells acquire this ability and leave their original tissue site to form metastases.) On page 1425 of this issue, Chen *et al.* (2) report a significant advance in the understanding of how anchorage dependence works.

Cell adhesion is mediated by cell surface receptors; one class of these, the integrins, are the primary receptors for ECM. [HN5] Originally discovered as a mechanical link between the cell surface and ECM (and, in some instances, the surface of a neighboring cell), integrins also link the external ECM "cytoskeleton" to the intracellular actin cytoskeleton. This linkage takes place in cultured cells at specialized membrane structures called "focal adhesions," which are thought to function similarly in intact tissues. Focal adhesions consist of a cluster of ECM-bound integrins that serve as a membrane attachment site for actin fibrils inside the cell. Many accessory cytoskeletal proteins concentrate at focal adhesions, as do various signaling proteins (3).

The presence of these signaling molecules reflects the fact that integrins do more than simply link structural elements. The integrins also trigger a number of signaling pathways, some of which are primarily related to cell adhesion, whereas others are shared by growth factor receptors. The focal adhesion kinase (FAK) pathway is used by several integrins and appears to participate in the control of anchorage dependence. Thus, a form of FAK that does not require integrin and cell attachment for its activity can render cells independent of anchorage (4). Moreover, integrins also activate many signaling molecules already known to be associated with growth factor signaling. These include the Ras-Raf-mitogen-activated kinase pathway, protein kinase C, and phosphatidylinositol 3′-kinase (5). Some integrins activate Shc, an adapter that serves as a link in various growth factor pathways (6). Indeed, integrins cooperate with growth factors to enhance mitogenic signaling (7).



The shape is the thing.

The biochemical signals originating from ECM-bound integrins would seem to provide an adequate explanation for anchorage-dependent survival and growth of cells. However, Chen *et al.* propose something different; their results indicate that cells forced to extend themselves over a large surface survive better and proliferate faster than cells with a more rounded shape. The fact that spread (flattened) cells thrive, whereas rounded cells do not, has been shown before. Thus, small beads coated with an integrin-ligand peptide can bind and activate integrin-specific signals but, owing to their small size, do not induce spreading and fail to rescue detached endothelial cells from anoikis (8).

The new feature of the work by Chen *et al.* is that they were able to equalize the surface area to which the cells were attached, while varying the shape (degree of spreading) of the attached cells. They accomplished

this with a clever use of microfabricated surfaces. In the key experiment, these investigators created a pattern of adhesive ECM dots on an otherwise nonadhesive surface that allowed cells to attach either to a single circular dot or onto several smaller dots. By varying the size and spacing of the small dots they were able to make the cells spread wider, while keeping constant the total area of contact with the ECM. Their rationale was that if chemical signals from the integrins were the predominant factor, the cells should survive and grow equally whether attached to one large dot or several small dots with the same combined area; the same amount of integrin engagement should generate the same amount of signal. What they found instead was that the cells spreading on the multiple small dots had less tendency to undergo apoptosis and thrived compared with the more rounded cells on the single dots. In addition, when the attachment was mediated by one set of integrins, the anti-apoptotic effect of spreading was clearer than when other integrins were involved. This reflects the fact that different integrins, while causing equal attachment and spreading, differ in their signaling pathways.

One striking feature of the endothelial cells used in this study is how malleable the shape of these cells is to instructions from the ECM. By plating the cells on tiny adhesive squares, the investigator produced square cells, whereas cells presented with round spots for attachment assumed a round shape; and so on with more complex patterns. Some cells--free-floating cells, such as red blood cells, and yeast--can determine their own shape by their cytoskeleton or other intrinsic signals (9). Endothelial cells, and presumably other adherent cells, clearly differ in that their shape is determined by the properties of the surrounding ECM and by adjacent cells.

The results of Chen *et al.* indicate that after the ECM controls cell shape, cell shape in turn controls survival and growth. This relation makes sense in situations such as tissue regeneration. For example, if an epithelial or endothelial cell layer is damaged, there will be fewer cells covering the damaged area, the remaining cells will be able to spread, and this would stimulate them to proliferate until the tissue gap is filled.

A highly speculative, but tantalizing, possibility raised by these and earlier results from the same laboratory is that cell shape might directly control gene regulation. The authors have shown that tugging on an integrin outside the cells can cause deformation and movement of the nucleus (10). It will be interesting to see whether such a physical connection might alter the regulation of growth and survival genes without chemical intermediates.

References

1. E. **Ruoslahti** and J. C. Reed, *Cell* **77**, 477 (1994) [Medline]; J. E. Meredith and M. A. Schwartz, *Trends Cell Biol*. **7**, 146 (1997).
2. C. S. Chen *et al.*, *Science* **276**, 1425 (1997).
3. K. Burridge and M. Chrzanowska-Wodnicka, *Annu. Rev. Cell Dev. Biol*. **12**, 463 (1996) [Medline].
4. S. M. Frisch, K. Vuori, E. **Ruoslahti**, P.-Y. Chan-Hui, *J. Cell Biol.* **134**, 793 (1996) [Medline].
5. E. A. Clark and J. S. Brugge, *Science* **268**, 233 (1995) [Medline]; M. A. Schwartz, M. D. Schaller, M. H. Ginsberg, *Annu. Rev. Cell Dev. Biol*. **11**, 549 (1995) [Medline].
6. K. K. Wary, F. Mainiero, S. J. Isakoff, E. E. Marcantonio, F. Giancotti, *Cell* **87**, 733 (1996) [Medline].
7. K. Vuori and E. **Ruoslahti**, *Science* **266**, 1576 (1994) [Medline]; S. Miyamoto, H. Teramoto, J. S. Gutkind, K. M. Yamada, *J. Cell Biol*. **135**, 1633 (1997), T. H. Lin, Q. Chen, A. Howe, R. L. Juliano, *J. Biol. Chem.* **272**, 8849 (1997) [Medline].
8. F. Re *et al*., *J. Cell Biol*. **127**, 537 (1994) [Medline].
9. F. Verde, J. Mata, P. J. Nurse, *J. Cell Biol*. **131**, 1529 (1995) [Medline].
10. A. J. Maniotis, C. S. Chen, D. E. Ingber, *Proc. Natl. Acad. Sci. U.S.A.* **94**, 849 (1997) [Medline].

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HyperNotes

Related Resources on the World Wide Web

The Dictionary of Cell Biology (London: Academic Press, 1985) defines some of the terms used in this article.

CELLS alive!, an educational service of Quill Graphics, provides timely and visually exciting material about cells of the immune system, bacteria, and parasites. The section on apoptosis provides a description of programmed cell death, illustrations, and a well-annotated list of links to other resources. The section on the cytoskeleton includes a movie that illustrates the redistribution of the cytoskeleton during phagocytosis.

The Biology 7.01 Hypertextbook, developed at the Massachusetts Institute of Technology, includes chapters on the cytoskeleton, cytoplasmic signal transduction, and other aspects of cell biology discussed in this article.

SCOP: Structural Classification of Proteins aims to provide a detailed and comprehensive description of the structural and evolutionary relationships among all proteins whose structure is known, including all entries in Brookhaven National Laboratory's Protein Data Bank (PDB). A keyword search can be performed to find records for Integrin CD11a/CD18 (LFA-1) from Human (*Homo sapiens*) and other proteins. For each protein, structures are given.

The World Wide Web Virtual Library: Physiology and Biophysics is a comprehensive list of Web resources related to physiology and biophysics. It is a component of the World Wide Web Virtual Library: Biosciences.

The World Wide Web Virtual Library: Biomolecules is a list of Web resources related to biological molecules. It covers molecular sequence and structure databases, metabolic pathway databases, and other lists of Web resources. The World Wide Web Virtual Library: Biochemistry and Molecular Biology is a list of resources listed by provider.

Pedro's BioMolecular Research Tools is a collection of WWW links to information and services useful to molecular biologists. It provides links to molecular biology analysis tools, bibliographic databases, guides, tutorials, journals, and other information on the World Wide Web.

CSUBIOWEB, the California State University Biological Sciences Web server, provides links to other Web sites on genetics, cell biology, and molecular biology.

1. **The Web page of Erkki Ruoslahti summarizes his research and lists some of his publications.**
2. **The Apoptosis/Programmed Cell Death Home Page, maintained by the New York Area Cell Death Society, provides definitions and illustrations related to apoptosis. It includes announcements of meetings and symposia and links to related resources.**
3. **Ask the Experts, maintained by Scientific American, includes a discussion of apoptosis.**
4. **Cell Biology/Apoptosis is a brief article in The Scientist that discusses apoptosis and anoikis.**
5. **Online Mendelian Inheritance in Man (OMIM) is a catalog of human genes and genetic disorders**