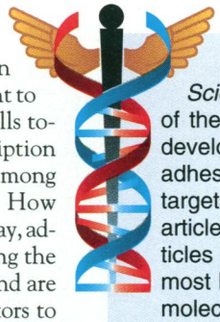
Biotech Gets a Grip on Cell Adhesion

From inflammation to cancer, the common cold to blood clotting, the potential of treatment based on "sticky" molecules is thrilling industry



ICON ILLUSTRATION: SAM WARD

In this issue on biologically based therapies, *Science*'s News Department takes a look at some of the biotech industry's most active areas of drug development research. One of those areas is cellular adhesion molecules, which are being exploited as targets for drugs for a wide variety of diseases; the article on this page explores those efforts. Two articles deal with the problems posed by the fact that most biotech drugs so far are proteins, large fragile molecules that must be injected. Research on replacing proteins with small molecules is discussed on page 910, followed by a discussion on page 912 of new ways to deliver proteins. The survey is rounded off by a look at the corporate side of things: an article on companies springing up to do gene therapy (page 914) and a grim look at the finances of the industry (page 908). Perspectives and articles follow this special news report.

Slightly more than a decade ago, the cell adhesion molecules were still thought to be mere glue that holds cells together in tissues, a description that stirred little interest among researchers in industry. How quickly things change. Today, adhesion molecules are among the hottest topics in biology and are seen as dynamic contributors to just about everything that happens in the body. In embryonic development, they are needed for tissue and organ formation. In the adult, they come into play wherever cells are moving and interacting—as in wound healing, inflammation, and cancer metastasis, which are all processes that biotech companies would love to get a handle on. As a result, cell adhesion fever has taken over in biotech.

Provided that the U.S. Food and Drug Administration (FDA) gives its approval, Telios Pharmaceuticals of San Diego, California, will soon bring the first adhesion molecule-based product—a gel for promoting wound-healing—to market. And this is just the beginning of what may be a long line of new products coming through the R&D pipeline. Other companies are working on potential therapies for cancer, multiple sclerosis, viral infections, rheumatoid arthritis, osteoporosis, atherosclerosis, and dozens of other medical problems. "It's hard to find anyone, including the large drug companies, that doesn't have an effort in this area," says Dale Cumming, one of more than two dozen scientists working on cell adhesion at Genetics Institute in Cambridge, Massachusetts.

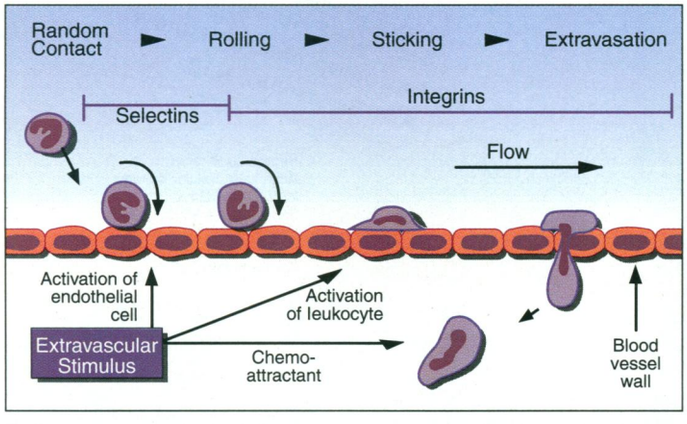
Indeed, adhesion molecule research is hot enough to buck the downturn in investment that has characterized the biotech field of late (see story on p. 908). Take a look, for example, at Glycotech, a new enterprise that in February opened its doors in Rockville, Maryland; the company's charter is to develop anti-inflammatory agents that work by preventing cells of the immune system from binding to certain adhesion molecules. And a few weeks ago, in Boston, Massachusetts, Leukon, a small firm devoted to finding novel therapies for autoimmune diseases such as rheumatoid arthritis, was born.

As the examples of Glycotech and Leukon suggest, a very wide range of adhesion molecules are attracting notice from biotech firms. Among those garnering the most attention are the integrins, a large family of proteins found on almost all of the body's cells. During the 1980s, it became clear that the integrins are a multifaceted group of cellular receptors. On the one hand, they help attach cells to one another, particularly in inflammatory responses. On the other, they join cells to proteins in the "extracellular matrix," a dense network of proteins and other molecules that surrounds and anchors cells. As cells interact with the matrix, they receive cues for migration, growth, and differentiation.

Diverse as these interactions are, in the early 1980s researchers including Telios cofounder Erkki Ruoslahti, who's also head of La Jolla Cancer Research Foundation, and company president Michael Pierschbacher noticed an important common element among the β-3 subfamily of integrins. These integrins always recognize and bind to a relatively small site, consisting of just three amino acids—arginine, glycine, and aspartate—on their target proteins. That simple peptide, known as RGD (for the standard letter codes for the three amino acids), has since become the basis of efforts to develop several new treatments.

In some cases, RGD-containing peptides are used to attract desirable cells. Take Telios' wound-healing gel, "Telioderm," which is laced with a synthetic peptide bearing the RGD recognition site. The idea is that the product, when spread on hard-to-heal skin lesions, provides "footholds" for the integrins on wound-healing cells, allowing them to migrate into a damaged area. In a recent clinical trial, says Pierschbacher, Telioderm—now awaiting final FDA approval—significantly speeded up the healing of chronic dermal ulcers and gave a four-fold increase in the number of ulcers healed.

Encouraging interactions between cells and integrins is one application for integrin-based technology. More often, however, adhesion-molecule based drugs are aimed at blocking the interaction between an integrin and its target as a way of preventing damaging cell interactions. In collaboration with Eli Lilly, for instance, Telios is tackling osteoporosis, the damaging bone loss suffered primarily by postmenopausal women. "Cells that absorb bone bind to it through integrins," explains Pierschbacher. In this case, the goal



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Rolling to a stop. In the inflammatory response, adhesion molecules on the endothelium known as selectins first slow white blood cells and cause them to roll along the surface. Other adhesion molecules on the cells, integrins, then latch on and stop the cells completely, allowing them to migrate out of the blood vessel (extravasation) and to the target tissue.

would be to use RGD-based compounds to block bones' integrins, preventing the attachment of bone-destroying cells.

It's in the blood

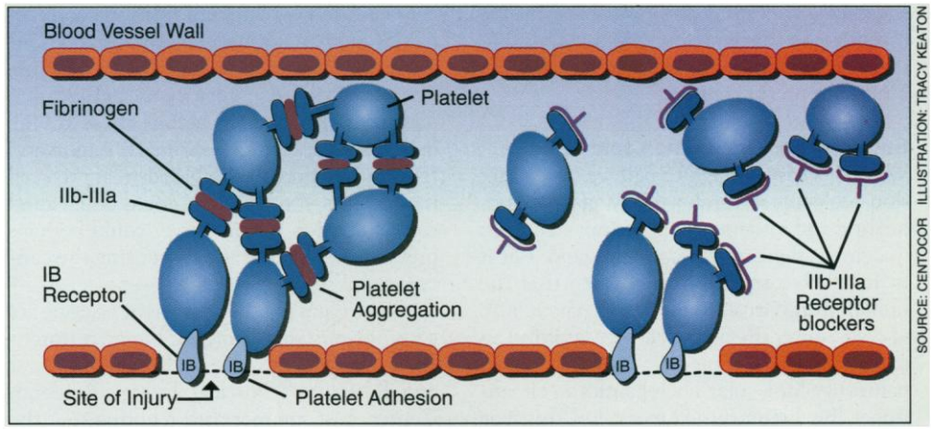
A similar strategy may lead to new drugs that prevent the life-threatening clots that cause many heart attacks and strokes. When a blood vessel wall is injured, blood cells called platelets rush in and adhere to the damaged site to prevent bleeding. That's good. The bad news, however, is that platelets carry an integrin receptor known as GP IIb-IIIa that binds to RGD sites on the blood protein fibrinogen. And since one fibrinogen can attach to receptors on two or more platelets, it may crosslink many cells, forming a potentially dangerous, vessel-clogging clump.

Several companies are developing drugs to prevent aggregation without reducing the platelets' crucial antibleeding activity. COR Therapeutics of South San Francisco, Merck, and Telios are focusing on synthetic RGD-containing peptides they hope will do the trick. In contrast, Centocor Inc. of Malvern, Pennsylvania, is moving forward with a monoclonal antibody fragment that binds to and blocks the platelets' fibrinogen-binding receptor. In March, the company reported that the drug reduced clot-related complications by 35% in patients undergoing high-risk angioplasty to open clogged coronary arteries.

These clot-busting drugs are attracting attention because the market for them is substantial—an estimated $550 million a year in the United States alone. But that figure pales beside the estimated market for anti-inflammatory drugs: a cool $10 billion.

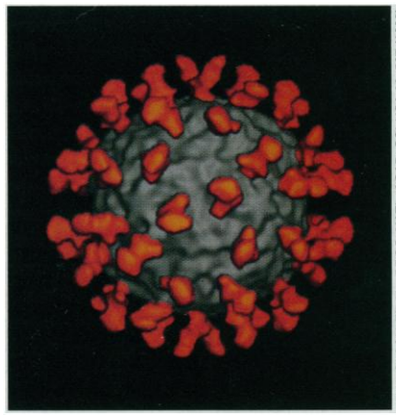
While the inflammatory response is beneficial because it helps rid the body of infections, disorders arise when it is misdirected to normal tissue or persists longer than is needed. Take ischemia-reperfusion injuries, perhaps the biggest acute inflammation problem that biotech firms are interested in. When blood flow is cut off from tissue, as happens in frostbite, heart attacks, stroke, and many other instances, the tissue becomes ischemic or oxygen deprived, and mistakenly releases signals capable of activating inflammatory blood cells. When blood flow is restored to an organ—reperfusion—there can be a wave of damage to otherwise healthy tissue. Often this secondary destruction is more harmful than whatever had caused the ischemia.

In addition to reperfusion injury, anti-inflammatory medications are also needed for diseases ranging from acute conditions like adult respiratory distress syndrome and septic shock to chronic illnesses such as rheumatoid arthritis, multiple sclerosis, asthma, and other autoimmune disorders. Given stakes like those, it's not surprising that the biggest thrust in adhesion research has been in the anti-inflammatory area, where dozens of companies are jumping in. "The anti-inflammatory market is enormous and largely unsatisfied. There's plenty of room for many drugs," comments Genetic Institute's director of research Patrick Gage.



Clot-buster. Small molecules or antibodies prevent dangerous platelet aggregation by binding to the IIb-IIIa receptor, without inhibiting the adhesion needed to stop bleeding.

One reason there's room for so many drugs is that adhesion molecules play a role at almost every step in the exquisitely detailed chain of events that constitutes inflammation. The inflammatory response gets under way when damaged tissue calls for help by releasing chemical signals that activate the inner lining (endothelium) of nearby blood vessels. Here, selectins, another important family of cell adhesion molecules, rise to prominence—literally—as the endothelium begins displaying first P-selectin and then a bit later E-selectin. These adhesion molecules draw in the white blood cells called neutrophils, slowing them down and causing them to roll along the endothelium.



Cold caps. The virus-binding portion of adhesion molecule ICAM-1 *(orange)* attaches to a rhinovirus.

Many companies have turned their eyes towards treating inflammation by blocking the selectins. "You should be able to develop [selectin] antagonists that inhibit rolling. And the assumption is that if you inhibit rolling, you inhibit inflammation," says Lawrence Lasky of Genentech in South San Francisco. Indeed, monoclonal antibodies that bind to the E- and P-selectins and prevent their attaching to neutrophils have successfully suppressed inflammation in animal models, says Donald Anderson, who directs the cell adhesion effort at Upjohn Co. And many companies are also intensely interested in developing small molecules or carbohydrate-based compounds, both of which could be taken orally, as an alternative to antibody treatment.

The selectins are only one target, however. As the neutrophils roll along the inner lining of the vessels, they encounter activating signals that cause them to turn up the binding power of the integrins they display on their surfaces. The neutrophil integrins then latch on tightly to still another class of endothelial adhesion molecules, which resemble the immunoglobulins and have names such as ICAM-1 and VCAM, that bring the neutrophils to a screeching halt. The white blood cells undergo dramatic shape changes, allowing them to squeeze through the blood vessel wall into tissue and migrate to the site of injury where they attack the bacteria or whatever foreign invader initiated the damage in the first place. Depending on the severity and type of tissue damage, other kinds of white blood cells, such as lymphocytes, may follow the neutrophils in.

The effort to find anti-inflammatory agents that work by blocking integrins is no less intense than that aimed at the selectins. A number of companies, for instance, are tackling multiple sclerosis by blocking integrins found only on lymphocytes—and early animal results are promising. Targeting a different white blood cell, Repligen is poised to begin clinical trials of an antibody fragment to a neutrophil integrin called Mac-1. Company researchers chose that target, says Repligen senior vice president for research and development Walter Herlihy, because Mac-1, in addition to being needed for neutrophil movement into tissues, is the receptor that triggers the cells to release their destructive products upon reaching targeted tissue. Scios Nova Pharmaceuticals of San Di-

ego may have come across another promising candidate in the leumedins, compounds that prevent neutrophils from displaying the Mac-1 integrin on their surface.

Sticking it to the common cold

While it's easy enough to believe that adhesion-molecule research will pay off in wound-healing and inflammation, it's not so easy to imagine it fighting the common cold. But it is. In 1989, researchers discovered that the human rhinoviruses, which cause about 50% of colds, enter the body's cells by latching on to the adhesion molecule ICAM-1. Companies like Molecular Therapeutics and Boehringer Ingelheim rushed to exploit this new knowledge and have shown, in test tubes, that soluble ICAM-1, acting as a molecular decoy, can block infection of cells. And while many of those companies are still pursuing soluble ICAM, some doubts have cropped up. "It was a fascinating discovery, but it hasn't panned out because soluble ICAM just isn't very sticky," contends Upjohn's Anderson, explaining that the concentrations of the molecule needed to show an effect were too high for medical use.

> **"It's hard to find anyone, including the large drug companies, that doesn't have an effort...."**
>
> **—Dale Cumming**

Those suffering from the sniffles need not give up hope, because research in the area is continuing. In the June issue of *The Journal of Virology*, leading cell adhesion researcher Timothy Springer of Harvard Medical School, who will chair Leukon's science advisory board, and colleagues report on a promising alternative to soluble ICAM. They have created a type of molecule called an "immunoadhesion" by using genetic engineering techniques to fuse antibody fragments to the rhinovirus-binding portions of ICAM-1.

These constructs have a dramatically improved ability to bind to rhinovirus, says Springer. One immunoadhesion, for instance, was 200 times more effective in the test tube than ICAM-1 at blocking infection by cold viruses. While a cold is more nuisance than dangerous infection, such work could lay the foundation for blocking other, lethal viruses that might also invade cells via adhesion molecules.

Why cancer cells roam

Even further afield, and perhaps more speculative, the growing understanding of cell adhesion may open up another front in the war on cancer. The link between cell adhesion and cancer lies in metastasis, in which cancerous cells separate from a primary tumor and disperse throughout the body to sprout new tumors—with deadly results. The adhesion molecules displayed by metastatic cancer cells are very different from the complement found on normal cells, or even on nonmetastatic cancer cells, notes Richard Hynes, director of MIT's Center for Cancer Research, and the changes may permit these roving cells to escape from a primary tumor and then move through the bloodstream to seed new tumors. If researchers could understand these adhesion changes they could perhaps prevent them—thereby preventing the cancerous cells from dispersing.

Some clues to how that might be done are coming from two groups of German researchers at Heidelberg's Cancer Research Center and Karlsruhe's Nuclear Energy Research Center. Last summer they reported that the adhesion molecule CD44, found normally on lymphocytes, also studs the surface of pancreatic tumor cells that are metastatic. They hypothesize that the CD44 disguises the cancer cells as white blood cells and allows them to circulate freely in the bloodstream (*Science*, 31 July 1992, p. 614). Metastasizing cells from other cancers may hitch a ride on platelets, by binding to their P-selectin, and again evade the immune system's detection.

The research on adhesion molecules' roles in cancer, while in its early stages, does suggest some medical uses. There is the potential for new diagnostic tools, assuming researchers can accurately correlate levels of certain adhesion molecules with how invasive a tumor might be. Or, as a temporary treatment, "you could envision giving an anti-adhesion agent during the surgical removal of a primary tumor," says Cytel's Jim Paulson. That could increase the chances that the immune system would destroy any cancer cells shed into the bloodstream because of the operation.

The work on cancer may be in the early stages, but the same cannot be said of cell-adhesion work overall, which is in full swing. Indeed, researchers believe that most of the major cell-adhesion molecules have already been identified, and they now face the task of determining which ones are crucial for specific diseases and how their deleterious sticking—or failure to stick—can be manipulated. There most certainly will be a mad dash from biotech and drug companies to get products into clinical trials and through FDA approval, a process that, if history is any indication, will weed out a tremendous number of experimental treatments. For the moment, however, optimism reigns. Says MIT's Hynes: "The field is very exciting, the science is rolling quickly, and the biotech applications add extra spice."

–John Travis

INDUSTRY FINANCE

Biotech Sails Into Heavy Financial Seas

If biotech executives are beginning to have something of a sinking feeling, they have good reason. Aside from a temporary surge last fall, biotech stocks have been declining since January 1992. In the past 4 months alone they've dropped 40%—bringing the total loss to 50% overall. Investors are shying away partly because of the unease created after three highly touted sepsis drugs failed to perform up to Wall Street's expectations in clinical trials (*Science*, 26 February, p. 1243).

But the antiseptic backlash isn't the only problem for the industry. In March, President Bill Clinton, unhappy with the rising cost of prescription drugs, began floating the idea of price controls as a possible fix. Clinton's tough talk instilled fear among investors, many of whom are cashing in their biotech stocks in case the president follows through on his "hints." "You can see the kind of impact just talk of price controls has had on the biotechnology industry," says G. Kirk Raab, president and chief executive officer of Genentech Inc.

At the heart of the apprehension generated by price controls is one fear: If controls were in place, a biotech company would be unable to shower its investors with profits from a blockbuster new drug. "Price controls on new drugs have the potential of killing the industry," says the downbeat Raab. The reason is that while biotech firms have lots of promising new drugs in the pipeline, they don't have many products on the market. Therefore, they have to tap investors constantly to keep their books balanced. And if the investors constantly see returns that—due to price controls—seem minuscule compared to what they're used to, they're going to look elsewhere for their profits.

In foreign hands?

To some industry analysts, this scenario may wind up putting some of the United States' best biotech research in the hands of foreign owners, much as Japan ended up controlling the semiconductor industry in the late 1970s. "This is a watershed year," says venture capitalist Robert McNeil, general partner of Sanderling Ventures, which counts nine biotech companies in its stable. McNeil plans a business trip to Japan in June 1994. If price controls are enacted, he says he'll be there to sell off companies, rather than to forge collaborations with Japanese firms.

In past years, scientists and businessmen