Cell growth factor: Use with caution

A cell-stimulating growth factor that has shown promise in patients with shortages of white blood cells can also cause blindness and death in mice, according to new research. There are significant methodological differences between the new study and previous trials on humans, making it difficult to draw parallels between the two types of studies. But the researchers warn that excess stimulation by the factor "may lead to the development of serious disease states . . . [that] could be of importance in the context of current clinical trials."

The researchers looked at mice that had been genetically engineered to produce excess quantities of granulocyte-macrophage colony stimulating factor (GM-CSF). Normally present in the blood in small quantities, GM-CSF stimulates the proliferation and function of certain white blood cells. When given intravenously in moderate amounts to patients with AIDS, it has stimulated increased production of disease-fighting white blood cells (SN: 9/12/87, p.165).

Working at research institutes in Melbourne and Parkville, Australia, and at the Duke University Eye Center in Durham, N.C., the researchers used a retrovirus to transfer an extra GM-CSF gene into male and female mice and examined the mouse progeny for effects of increased production of the cell growth factor. They found that GM-CSF blood levels were at least 40-fold higher than in normal mice, but along with that increase came a number of problems.

Most strikingly, all of the transgenic mice had opaque eyes. Tissue samples showed that the eyes had been ravaged by an overabundance of macrophages—a variety of white blood cell that is especially activated by GM-CSF. Degeneration of the lens and retina was apparent, along with varying degrees of retinal detachment. Other parts of the mice were heavily infiltrated with macrophages — many of them larger than normal and containing two to 18 nuclei instead of one. The overgrowth of macrophages was accompanied by tissue destruction in all 81 recombinant mice, and within five months most of them died.

It's likely, the researchers say, that the increase in macrophages was a consequence of the growth-promoting properties of GM-CSF, and that the tissue destruction was a direct consequence of the accumulation of those cells. Macrophages are known to secrete chemical factors that can destroy muscle tissue while attracting a variety of reactive, inflammatory cells. Surprisingly, however, there was no evidence of GM-CSF production in the bone marrow of the transgenic mice, where, under normal circumstances, GM-CSF exerts its proliferative effects on progenitor white blood cells. Further tests led the researchers to conclude that in the gene-altered mice, the macrophages themselves were both producing and reacting to the stimulating factor.

"The macrophages in these transgenic mice may be autocrine [self] stimulated," the researchers say. "Furthermore, CSF is probably produced in the transgene very early in development, while therapeutic use would be more likely to involve short-term exposure in adults."

Those differences, say the authors and others, are significant. According to H. Franklin Bunn, a hematologist from Brigham and Women's Hospital in Boston, the new research "means uncontrolled release of GM-CSF could lead to problems. But keep in mind that you have to interpret it very carefully in how it applies to clinical application. This transgenic model is very different because the gene is being expressed in tissues where it normally wouldn't be expressed," Bunn told SCIENCE NEWS.

Nevertheless, the researchers warn, the findings "highlight the need for careful toxicological studies before these agents are used for prolonged periods in the treatment of chronic conditions."

— R. Weiss

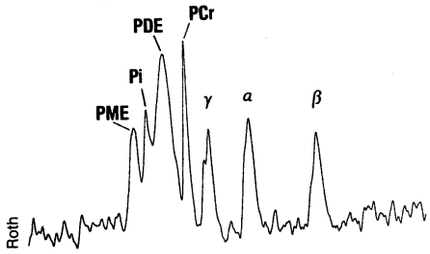
Focusing on brain-tumor phosphates

High-energy phosphates are the "energy currency" of cells, fueling everything from protein synthesis to cell movement. Getting an accurate reading of the levels of these molecules is crucial to understanding how cells create and use energy.

Using nuclear magnetic resonance (NMR) spectroscopy, Klaus Roth and his colleagues at the San Francisco Veterans Administration Medical Center have obtained the first noninvasive, quantitative measurements of energy-storing phosphates in human brains and brain tumors. Roth spoke in Chicago at a meeting of the Radiological Society of North America.

NMR spectroscopy, which is based on the behavior of nuclei in an applied magnetic field, is not the only way of quantifying levels of energy-storing molecules. In fact, there are other methods that are much more sensitive. But according to Gregory Karczmar, a physical chemist at the same center, NMR is the only practical way of studying metabolic reactions *in vivo* without taking biopsies. Moreover, while NMR spectroscopy has been used before to study phosphates in the brain, those studies could only provide ratios of compounds. In contrast, Roth's group can measure absolute levels by comparing the brain's NMR signals with those from a sample placed near the patient's head and containing a known concentration of phosphorus atoms.

"Our goal is to apply these quantitative measurements to get a quick test of the effectiveness of chemotherapy," says Roth, whose group is one of a number using NMR to look at energy-storing molecules in living tissue. At present a physician must wait two or three weeks to see whether a tumor has shrunk in response to treatment. "But if chemotherapy works, there are biochemical changes you can observe in the tissue within days," says Roth. Animal studies show that the high-energy phosphate concentrations reflect most biochemical changes inside cells. If these levels drop, a physician will know that tumor tissue has died and the chemotherapy is working.



From an NMR spectrum, Roth's group can measure the levels of these phosphate compounds: phosphocreatine (PCr), adenosine triphosphate (γ, α, β), inorganic phosphate (Pi), phosphomonoester (PME) and phosphodiester (PDE).

Roth also would like to use NMR spectra in humans to detect the early stages and extent of ischemia, in which the flow of oxygen-carrying blood to tissue is blocked. Normally, cells use oxygen to make adenosine triposphate, a high-energy phosphate, but when there isn't enough they use glucose to produce it. This process also produces lactic acid and a drop in pH, which can be measured with NMR by noting the distance between NMR peaks of inorganic phosphorus and phosphocreatine.

Karczmar thinks that basic researchers, such as those studying energy use and fatigue in muscles, will also be interested in Roth's quantitative measurements. Such measurements would enable them to get into the nitty-gritty of cell bioenergetics — which involves various reactions and their properties, such as speed and direction. While the technique itself isn't novel, he says, "Roth has gone much farther than past work by doing the very difficult calculations" necessary for interpreting NMR data.

— S. Weisburd