Orphan Interferon Finds a New Home

Uncertainties about the role of interferon-β_2 are being resolved as researchers find that it has numerous activities in the body's defenses

In the 7 years since the discovery of interferon-β_2, there has been some confusion about what its role in the body might be. Although some investigators found that it blocks viral reproduction, which is one of the hallmarks of members of the interferon family, others could not detect such antiviral activity. The agent thus remained in limbo where its function was concerned.

In the past year, however, researchers have learned that several biological activities can be attributed to none other than interferon-β_2. The material stimulates several different cell types, including both the T and B cells of the immune system, liver cells, and apparently even nerve cells. Many of these findings were described at a recent workshop on cytokines*, which is the general name given to the agents used for transmitting regulatory signals among cells.

The results mean that interferon-β_2 can take its place with other interferons and interleukins as a major contributor to the body's defenses against infection and injury, even if it does not block virus reproduction directly. "Together with interleukin-1 and tumor necrosis factor, interferon-β_2 is emerging as a major mediator of communication between cells inside and outside the immune system," says Jan Vilček of New York University Medical Center in New York City.

The absence of antiviral effects in most researchers' experiments, coupled with the new discoveries about what interferon-β_2 does do, has also led to a movement to rename it "interleukin-6." Interleukins are agents used by leukocytes (white blood cells, including monocytes and the T and B cells) to communicate with one another, although the effects of the various interleukins often extend to additional cell types—as those of interferon-β_2/interleukin-6 certainly do.

The first of the factors to be identified with interferon-β_2 was a typical interleukin, namely B-cell stimulating factor–2 (BSF-2), which is produced by T cells and acts on B cells, bringing about their final maturation and antibody production. Toward the end of 1986, Toshio Hirano and Tadamitsu Kishimoto of Osaka University and their colleagues cloned and sequenced the gene for the B-cell factor and deduced the amino acid sequence of the protein that it encodes.

The Osaka group found a limited resemblance between the BSF-2 protein sequence and that of one of the several proteins needed for the growth and maturation of bone marrow cells, but did not find any similarities to other known protein sequences. However, Alfons Billiau of the Catholic University of Leuven, Belgium, noted that the BSF-2 sequence is identical to that of the "26kD" protein. Jean Content of the Institut Pasteur du Brabant in Brussels, Walter Fiers of the State University in Ghent, Belgium, and their colleagues had cloned the 26kD gene, but the sequence of the protein had apparently not yet entered the data banks at the time the Osaka workers were looking for BSF-2 relatives.

> ***"Interferon-β_2 is emerging as a major mediator of communication between cells . . . "***

In any case, 26kD protein is just another name for interferon-β_2. The Fiers group called the protein that because it has a molecular mass of 26 kilodaltons and they were unable to detect antiviral activity for it. They therefore did not want to use the interferon designation.

The interferon-β_2 gene has also been cloned and sequenced by Michel Revel of the Weizmann Institute of Science in Rehovot, Israel, and his colleagues. Early on, Revel had found that the protein has the antiviral activity characteristic of an interferon and still supports that view.

Meanwhile, because of all this confusion surrounding interferon-β_2's effects, Billiau, Jozef Van Damme, also of Leuven, and their colleagues had set out to clarify the issue by cloning the gene themselves. The early studies of the agent had been performed with material that had been purified from the producing cells and might have been contaminated with small quantities of other biologically active proteins.

A possible contamination of this type may be avoided by cloning the desired gene and using it to make the protein in question. During the course of this work, the Billiau group noted that cells that are stimulated to make interferon-β_2 release a factor that enhances the growth of hybridomas, the hybrid cells used to make monoclonal antibodies, and plasmacytomas, which are B-cell tumors. The cells that are fused to make hybridomas are themselves of B cell origin.

The cloning and sequencing of the gene for the hybridoma growth factor established that it, too, is identical to the interferon-β_2 gene. L. A. Aarden and his colleagues at the University of Amsterdam also cloned the hybridoma growth factor gene and showed that it encodes interferon-β_2.

Although the agent was originally thought to be related to the classic interferon-β—and thus given the β_2 designation—the cloning of the interferon-β_2 gene made it clear that there is little sequence resemblance between the two cytokines.

In addition, the cloning work established that interferon-β_2 has the ability to stimulate the growth of B cells, as well as inducing their maturation and antibody production. The effects would aid in fighting off infections by enhancing antibody production. Billiau says, however, that he and his colleagues do not detect significant direct antiviral activity with the interferon-β_2 produced by their cloned gene.

In contrast, Revel still finds antiviral activity with the material produced by his cloned gene, a result that puts him in the minority among the researchers studying interferon-β_2. Revel concedes that the agent is much less potent as an antiviral agent than more typical interferons, but he nonetheless considers virus inhibition to be a significant aspect of its functioning.

The reason for the discrepancy between Revel's results and those of other investigators is unclear. It is apparently not the result of the investigators having cloned different genes. Lester May, who works with Pravinkumar Sehgal at Rockefeller University reported that there is only one interferon-β_2 gene in the human genome.

He also pointed out, however, that there are multiple forms of the protein itself. This means that the protein undergoes modifications, such as the addition of sugars or phosphate groups, that could yield products

*The "International Workshop on Monokines and Other Non-Lymphocytic Cytokines," which was held on 6 to 10 December on Hilton Head Island, South Carolina.

with varying effects. "There are many different forms that do appear to be present. Maybe some of the controversy exists because the proteins are slightly different," May suggests.

The latest factor to be found identical to interferon-β_2 is, according to Jack Gauldie of McMaster University in Hamilton, Ontario, and his colleagues, hepatocyte-stimulating factor, which acts on liver cells to increase the synthesis of a group of proteins that are released into the blood during inflammatory responses. This "acute-phase protein response," as it is called, is one of the oldest and most conserved of the body's responses to injury by infection or trauma.

"It is the most important regulator of the acute-phase protein response," comments Vilček. "That puts it right in the middle of responses that occur after infection and injury." Interleukin-1 and tumor necrosis factor, both of which are major contributors to inflammatory reactions, also stimulate some aspects of the acute-phase protein response.

The effects of interferon-β_2 generally resemble those of interleukin-1 and tumor necrosis factor. Interleukin-1 is a fever inducer, for example, and so is interferon-β_2, according to Aarden. Moreover, Aarden has found that interferon-β_2 induces the proliferation of murine thymocytes in an assay that had been considered to be specific for interleukin-1.

The similarities in the activities of interferon-β_2 and interleukin-1 raised concerns for a while about the validity of results with early interleukin-1 preparations that may not have been 100% pure. "The people who had 'interleukin-1' before cloning could have had interleukin-6 instead," explains Charles Dinarello of Tufts University–New England Medical Center in Boston.

Moreover, both interleukin-1 and tumor necrosis factor stimulate interferon-β_2 production, a circumstance that increases the possibility that the interferon might have been contaminating interleukin preparations. Experiments with pure cloned interleukin-1 confirm that its does have the effects originally found for it, however. "Recombinant interleukin-1 worked in most of the assays in which it was supposed to work," Dinarello says.

The overlap of the activities of interferon-β_2, interleukin-1, and tumor necrosis factor illustrates a prominent feature of the cytokines. Cytokines often display a high degree of redundancy in their effects. The body apparently takes no chances when it comes to fighting off foreign invaders and injury. "All these activities [of interferon-β_2] should not confuse us," Revel declares. "They all play a role in the defense of the organism against infections." ■ JEAN L. MARX

Gamma Rays for Christmas

The long-awaited observation of gamma rays from Supernova 1987A has given astronomers their first direct observational evidence for the theory of explosive nucleosynthesis—the idea that iron and most of the other heavy elements in the universe were created by supernovas at the instant of detonation.

The observations, which were reported on 14 December at a nuclear spectroscopy symposium in Washington, D.C.,* were made independently by two instruments. The first was the gamma-ray spectrometer aboard the National Aeronautics and Space Administration's (NASA's) Solar Maximum Mission satellite. The data reported at the symposium ran from August, when the gamma rays were first detected, until October. More recent data is still under analysis, according to principal investigator Edward L. Chupp of the University of New Hampshire.

The second set of observations were obtained by two balloon-borne experiments flown in October and November from Alice Springs, Australia, as part of NASA's Fall Supernova Observation Campaign.

Visible in both sets of data are gamma-ray emissions at energies of 847 kiloelectron volts (KeV) and 1238 KeV—precisely the experimental signature astronomers have been waiting for. These two energies correspond to the most prominent gamma rays emitted by the decay of radioactive cobalt-56. Their observation thus confirms a key link in the supernova's chain of element creation.

According to the standard theory of supernovas, the detonation of 1987A produced copious amounts of the radioactive isotope nickel-56. Cobalt-56 was formed soon thereafter as a decay product of the nickel. Now, several hundred days later, the cobalt is slowing decaying in its turn and transforming itself into iron-56, which is stable. Indeed, this latter isotope is the most common form of iron. Astronomers are confident that virtually all of the iron in the universe—including all of the iron found on Earth—ultimately came from supernovas just like this one.

Although this theory has now been confirmed in its essentials, however, the details of the new gamma ray observations present the astronomers with a number of mysteries.

Most striking is the fact that the Solar Maximum instrument began seeing gamma rays last August, at about the same time that x-rays were first detected from the supernova by instruments aboard the Soviet Union's Mir space station and Japan's Ginga satellite. This is odd because most astronomers had assumed the x-rays were actually gamma rays that had lost energy as they tried to pass through the supernova's expanding shell of debris. Since gamma rays come from the cobalt, and since the cobalt is thought to be located in the deep interior of this shell, most astronomers had assumed that the gamma rays would not appear until several months after the x-rays. Before that could happen, they said, the shell would have to expand still more and become thin enough to let the gamma rays come out directly.

Apparently, the astronomers' assumptions were wrong. On the other hand, the x-rays themselves were detected about 100 days earlier than predicted. According to some researchers, both anomalies could be explained if there were turbulence in the supernova shell. If so, then swirling eddies of gas might cause some of the cobalt to well up from the deep interior, and thus become visible on the surface. ■

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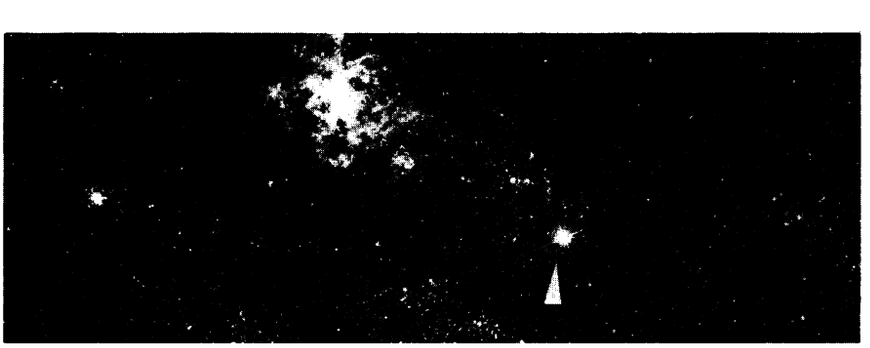
* "X-Ray Spectroscopy of Astrophysical Sources," Washington, D.C., 14 to 16 December 1987.



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