well. And that's why the new findings came as such a shock.

Colonna and Jacqueline Samaridis report that they have cloned a set of four related genes that appear to encode the inhibitory receptors of human NK cells, and these proteins are not lectins. They belong instead to the immunoglobulin superfamily, which also includes antibody proteins and the receptors by which T cells recognize their targets.

Colonna and Samaridis began their search for the inhibitory receptor genes by looking for either immunoglobulin superfamily or lectin genes that are active specifically in NK cells, and then used a variety of methods to confirm that they had the receptor genes. The Long and Moretta group took a different tack, using monoclonal antibodies directed against the receptors as a tool to get their clones. But they came to the same conclusion, identifying five related genes that encode immunoglobulin superfamily members. The teams have not yet compared their genes to see if any of them are the same.

Such new findings are usually a cause for celebration, but Berkeley's Raulet says that

Natural killer cells "seem more complicated than we had thought."

—David Raulet

when he first heard Nicolai Wagtmann, from the Long lab, present that team's results at a meeting in Sicily last May, he was "surprised and dismayed" to find that the new human proteins aren't related to Ly-49, as immunologists had expected. The finding seemed to be a biological paradox—how could a pair of species as closely related as humans and mice, whose immune systems are otherwise so very similar, have such totally different genes performing the same function?

Raulet says his dismay at the time came from the gut feeling that one of the findings must be wrong, although he now believes the new findings are solid. The Colonna and Long groups haven't proven directly that the proteins made by the genes they have cloned bind to class I proteins, as the Ly-49 proteins do, but they have shown that the proteins are recognized by antibodies known to block class I inhibition. That, says Raulet, makes "a reasonable case that these in fact are inhibitory molecules."

That leaves the issue of whether humans and mice use unrelated inhibitory receptors, an idea most immunologists reject. "I'm still convinced that we will find in man the Ly-49 genes, and probably in mouse we will find this other set of genes," says Lanier, "and they will be serving redundant functions." That raises "fascinating questions," says Raulet. "If you have two independent inhibitory receptors, how do you coordinate specificity, and why isn't one enough?"

It's not only the NK cells that are raising fascinating questions. In their paper, Lanier's group reports that human T cells carry on their surfaces the same family of inhibitory receptors that have been identified on human NK cells. This follows up on a report last year in the *European Journal of Immunology* by a team led by Lorenzo Moretta of the University of Turin, Italy, that a small subset of T cells have such an inhibitory receptor. But the Lanier group took their analysis further, showing that most T cells that have been activated by an encounter with foreign proteins seem to have one or more forms of the NK receptors, and that the receptors respond to class I molecules by restraining the T cells from attacking cells they would otherwise kill.

"I was very surprised that T cells have these NK receptors," says Harvard's Strominger. Indeed, at first glance, the receptors seem to be working at cross purposes to the T cells' main job, which is to kill cells that present foreign peptides. "The T cell is recognizing the class I–peptide complex on the antigen-presenting cell, but at the same time, the class I molecule is binding to an inhibitory receptor on the T cell and down-regulating it," Strominger says.

Such fine-tuning might mean that a foreign peptide–MHC protein combination that is a perfect fit for a T cell receptor will send an activating signal that is more powerful than the signal going through the inhibitory receptor, causing the T cell to kill its target. But any peptide–MHC protein complexes that don't fit snugly in the T cell receptor might send weaker activating signals that would be overwhelmed by the inhibitory signal. In such a model, the inhibitory receptor "would serve as a failsafe to make sure your activated T cells will only kill things which really provide a strong signal," Lanier says, adding that autoimmune conditions could result from failure of the inhibitory receptors to "tame down or regulate the activated T cell."

That kind of counterbalancing is likely to come into play on the NK cells, which also must be triggered to go into killing mode. But little is known about how NK cells are activated. It would be much better to study the inhibitory signal in T cells, where activation is well understood, says Raulet, "because you could say where along this [activating] pathway does the inhibitory signal intervene." As researchers tackle that question, they will likely reveal more about how the muzzle actually tames the bite of the NK cells.

–Marcia Barinaga

AIDS THERAPY

New Hope Against Blindness

"I know I'm going to die, but I don't want to go blind." Retina specialist William Freeman of the University of California, San Diego (UCSD), says he "uniformly" hears that distressing plea from his AIDS patients. Yet, despite the best efforts of Freeman and his medical colleagues, many of those patients will lose some vision; a few will lose it all. The reason is retinitis, an inflammation of the retina caused by cytomegalovirus, or CMV. This damaging condition, which may affect up to 40% of AIDS patients in the final stages of their disease, is not easy to treat because the available drugs are toxic, expensive, and work best only when given intravenously every day.

Now, however, Freeman and his peers may finally be able to offer their patients a ray of hope: Three new forms of "local" therapy—treatments that involve delivering drugs directly to the eye rather than intravenously—have shown great promise in early human trials. "Local treatment is coming into its own," says ophthalmologist James O'Donnell of the University of California, San Francisco (UCSF), who heads the school's Studies of the Ocular Complications of AIDS (SOCA), a network of 11 clinics sponsored by the National Eye Institute. "It might be that a combination of systemic [IV therapy] and local treatment will be the way of the future."

CMV retinitis usually affects AIDS patients in the terminal stages of the disease, when damaged immune systems no longer suppress this member of the herpesvirus family. Although CMV, which normally is harmless, can cause everything from pneumonia to gastrointestinal bleeding, some of its worst damage is done in the retina. The best proven treatments are the anti-viral drugs ganciclovir and foscarnet, administered through an indwelling catheter. But not only is the catheter site vulnerable to sepsis; ganciclovir and foscarnet damage blood cells and kidneys respectively. So far the only alternative to the IV therapy is a less potent oral form of ganciclovir that was approved by the Food and Drug Administration (FDA) just this past December.

The local treatment closest to market is a tiny ocular implant being developed by Chiron Vision in Irvine, California, a division of Chiron Corp. The implant, which releases ganciclovir for 8 months, requires a

1-hour surgery under local anesthesia. Two controlled trials have yielded impressive results. An interim analysis of 148 patients in one study, first reported at an AIDS meeting in January, revealed that implant recipients had a median time to progression of CMV retinitis of 186 days. In contrast, patients given IV ganciclovir progressed in 72 days. The second study, involving 26 patients, was reported in the December 1994 *Archives of Ophthalmology*; it yielded similar results. "That's the most effective time-to-progression seen yet," says UCSF AIDS researcher Mark Jacobson, who recently reviewed CMV treatments in *AIDS Research and Human Retroviruses*.

Jacobson cautions that in addition to the risk of surgery, several patients in these studies suffered retinal detachments, and it is possible that the implant speeded up this event, which causes blindness. Gary N. Holland, an ophthalmologist who heads the SOCA site at the University of California, Los Angeles, also warns that the implants can cause a transient decrease in vision—a serious drawback for people with only a short time to live. "It's wrong to focus on one factor like time-to-progression without considering the other factors," says Holland.

In the direct-injection arena, UCSD's Freeman has made headway using an experimental drug, HPMPC. As Freeman and colleagues reported this month in the *American Journal of Ophthalmology* and *Ophthalmology*, their small-scale, uncontrolled trials of a single injection of HPMPC into eyes that showed evidence of CMV damage resulted in a delay of disease comparable to daily IV infusions of the marketed drugs. Even more promising, a second injection given when recurrence of the disease was detected led to a second delay in progression. "It's clever and it has potential clinical utility," says UCSF's Jacobson. Daniel F. Martin, an ophthalmologist at Emory University School of Medicine who co-led a recently completed trial of Chiron's implant, calls the results "very encouraging."

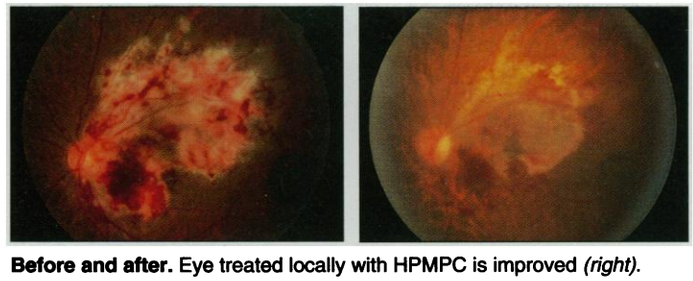
Although injections into the eye pose their own risks, Freeman is enthusiastic about his results. "A patient can go in to his eye doctor and have an essentially painless procedure that takes about 5 minutes, and then that retinitis is under control for 6 to 8 weeks," says Freeman, who has now injected HPMPC into 65 patients. The treatment's fate now rests with the company that owns the license to HPMPC (see box).

A few miles up the road from UCSD, researchers at Carlsbad's ISIS Pharmaceuticals have also been making progress with direct injections of an anti-CMV drug. ISIS and its Japanese partner, Eisai Co., are jointly developing an "anti-sense" compound that, because it is complementary to a portion of CMV's messenger RNA, can bind to the RNA and thus prevent the virus from replicating. ISIS has tested the drug in 22 patients who no longer responded to IV treatment with foscarnet or ganciclovir. Although ISIS President Daniel Kisner says eye exams with sensitive scopes have shown that the drug has "profound" anti-CMV effects, the studies were not designed to calculate progression rates. The biweekly injections appear safe, and, in December, the company launched larger efficacy studies.

Later this year, Chiron Vision plans to ask the FDA to allow the marketing of the company's ganciclovir implant. "I think an implant along with oral ganciclovir will be the treatment of choice in the future," predicts Judy Gordon, vice president of scientific affairs at Chiron Vision. A trial of this combination is now under way.

Few researchers expect local treatments to replace systemic approaches entirely. "If patients have CMV in the retina, they probably have it in other places," says UCSF's O'Donnell. And unlike systemic therapy, if local therapy is applied to one eye, it leaves the other vulnerable. But local treatments may offer AIDS patients some relief from the fears William Freeman hears too often in his medical practice.

–Jon Cohen



PHOTOS BY WILLIAM FREEMAN/UCSD

Before and after. Eye treated locally with HPMPC is improved *(right)*.

AIDS Drug: Experiencing Local Delays

What if a method for treating a crippling AIDS-related illness looked promising, but industry showed no interest in developing it? Until recently, that was the fate of intravitreal HPMPC, a promising approach for treating retinitis caused by cytomegalovirus, or CMV, a condition that frequently causes late-stage AIDS patients to lose vision (see main text).

Three years ago, William Freeman, an ophthalmologist at the University of California, San Diego, told Gilead Sciences in Foster City, California, that he wanted to inject HPMPC directly into the eyes of AIDS patients with CMV retinitis. Gilead, which owns the rights to HPMPC, was developing the drug for intravenous use, and Freeman says the company refused to provide it for his intravitreal injections. He was nevertheless able to test the approach by getting HPMPC from European chemists who manufactured it.

Three researchers interviewed by *Science*, all of whom insisted on anonymity, contend that Gilead showed no interest in local treatment with HPMPC because of worries that it wouldn't make much profit from the small, infrequent doses used in the technique. Intravenous (IV) infusions of HPMPC, in contrast, require larger doses every day. Indeed, Gilead's reluctance to work with Freeman at one point led AIDS activists to "zap" the company with a barrage of phone calls and faxes. "Any effort to move forward on intravitreal treatment has been completely stonewalled by the company," charged activist Kevin Robert Frost of Treatment Action Group.

Gilead's VP for clinical affairs, oncologist Howard Jaffe, says this criticism is groundless. Gilead did not supply Freeman with the drug, says Jaffe, because at the time it was a small start-up company that could only pursue a few options for HPMPC. Scientifically, Jaffe says, it made better sense to develop HPMPC for intravenous use first because CMV affects the entire body. "We've always taken the approach that the disease is a systemic disease, and we initially decided to develop the drug systemically," says Jaffe. Now, he says, buoyed by Freeman's impressive results, Gilead plans to pursue intravitreal treatment, too.

Because Gilead's HPMPC and the version Freeman has tested are not identical, the company plans to repeat his early work. Frost remains disgruntled about Gilead's earlier reluctance to supply Freeman with the drug. "Unfortunately, I think this has set us back by 2 years," says Frost. Gilead, for its part, hopes to win approval from the Food and Drug Administration later this year to market IV HPMPC. "I personally believe we've done the correct thing by doing this in a staged manner," says Jaffe.

–J. C.