AIDS VACCINES

Glimmerings of Hope From The Bottom of the Well

The number of AIDS vaccines entering clinical trials is at an all-time low, but researchers are planning to begin testing new approaches soon

Five years ago the AIDS vaccine field went into a tailspin when the two leading vaccines failed critical laboratory tests, and it still has not recovered. The number of new vaccines entering early testing in the U.S. trials network set up by the National Institutes of Health (NIH) has reached an all-time low; three trials were begun last year and none has been launched so far this year, compared with an average of six a year from 1990 through 1997. And earlier this month, researchers gave a decidedly mixed reception to results from preliminary trials of a combination vaccine—a strategy deemed more promising than the one that derailed 5 years ago. But some researchers now see glimmerings of hope.

Science has learned that Merck & Co., a pharmaceutical powerhouse that dropped out of the HIV vaccine field in the early 1990s, is aggressively reentering the arena with plans to launch tests of two different vaccines before the end of the year. And researchers are experimenting with new combination vaccines that they hope to move into early clinical trials next year. AIDS vaccine researchers are especially encouraged by the rebirth of Merck's program. "The more [players] we can get in with good ideas, the more the whole field benefits," says Donald Burke, who heads AIDS vaccine clinical trials at Johns Hopkins University. "The momentum right now is less than optimal."

The momentum virtually halted in June 1994, when NIH declined to fund large-scale efficacy trials of vaccines made by Chiron and Genentech. Both vaccines were based on genetically engineered versions of HIV's surface protein, gp120. The hope was that these viral proteins would raise antibodies that would attach to the virus in the bloodstream, before it infected cells, but NIH got cold feet when test tube experiments showed that the antibodies these vaccines produced could only stop laboratory-grown strains of HIV—not ones freshly isolated from patients (*Science*, 24 June 1994, p. 1839). (Genentech's vaccine now is in efficacy trials, funded privately by a new offshoot, VaxGen.)

When it pulled back from supporting the gp120 vaccines, NIH had high hopes that another approach would soon be ready for efficacy trials. As Jack Killen, a top AIDS official at NIH, predicted at the time, "the very realistic likelihood is within 2 to 3 years we would be ready to go with a different concept."

The "different concept" Killen and others had in mind was a combination of a gp120 vaccine and a vaccine manufactured by Pasteur Mérieux Connaught that consists of various HIV genes spliced into a live, but harmless, canarypox virus "vector." The logic behind this so-called "prime-boost" approach is that the two vaccines activate different arms of the immune system, which theoretically should work in concert to thwart HIV. The gp120 vaccine triggers antibodies, while the canarypox vaccine stimulates cell-mediated immunity, which occurs when the immune system dispatches cytotoxic T lymphocytes (CTLs) and other forces to rid the body of cells that the virus has infected.

AIDS VACCINE TRIALS UNDER WAY OR RECENTLY COMPLETED

Manufacturer	Vaccine	Location
VaxGen	gp120	U.S., Canada, Thailand, Netherlands
Chiron	gp120	Thailand
Pasteur Mérieux Connaught	gp160	U.S.
Pasteur Mérieux Connaught	canarypox with various HIV genes	U.S., France, Uganda
Pasteur Mérieux Connaught + Chiron	canarypox with various HIV genes gp120	U.S.
Pasteur Mérieux Connaught	lipopeptide	France
Pasteur Mérieux Connaught + VaxGen	canarypox with various HIV genes gp120	U.S.
Wyeth-Lederle	DNA vaccine with various HIV genes	U.S.
Pasteur Mérieux Connaught + Wyeth-Lederle	canarypox with various HIV genes DNA vaccine	U.S.
Therion	vaccinia with HIV envelope gene	U.S.
St. Jude Children's Research Hospital	23 HIV envelope proteins in 23 vaccinia vectors	Memphis, Tennessee
Univ. of Maryland + VaxGen	salmonella with gp120 gp120	U.S.
Cel-Sci	p17	Europe

SOURCES: NIAID, MANUFACTURERS

Two weeks ago, researchers at a conference on sexually transmitted diseases in Denver, Colorado, revealed results from the largest test yet done of a prime-boost vaccine. The study, sponsored by the National Institute of Allergy and Infectious Diseases (NIAID), began in May 1997 and involved 435 people, more than 80% of whom had a high risk of HIV infection because of drug use or sexual behavior. Although the study was not designed to determine whether the vaccines worked—it aimed to assess safety and the various immune responses triggered by the approach—researchers can glean hints of the vaccine's chances of success from the results.

More than 90% of those who received a "prime" shot of canarypox and a "boost" of gp120 developed antibodies that, in a test tube, could stop a laboratory-grown strain of HIV, and some 30% produced CTLs against the virus. These results meet milestones that NIAID has said must be achieved before launching an efficacy trial (*Science*, 1 March 1996, p. 1227). "This trial is a necessary step toward initiating an efficacy trial," says Peggy Johnston, head of NIAID's AIDS vaccine effort. "But," she adds, "several questions remain."

One question is whether newer canarypox vaccines made by Pasteur Mérieux stimulate higher levels of CTLs. A comparative trial of these vectors should be completed this summer. Johnston says another small-scale trial of this concept is also needed to determine the best timing of the booster shot and whether a different gp120 vaccine might work better. So even if these tests pan out, an efficacy trial is at least 1 year away.

Some AIDS vaccine researchers, however, have serious reservations about the results so far. Oxford University's Andrew McMichael, a CTL expert, is underwhelmed by the fact that killer cells were elicited in only one-third of the vaccinees. "Two-thirds of the people [may have had] no CTL response, and, if CTLs are important, they wouldn't be protected," he says. Similarly, HIV antibody specialist John Moore of the Aaron Diamond AIDS Research Center in New York City has strong doubts about the value of the antibody response, which still only blocks laboratory-

grown HIV. "It's a completely meaningless antibody response," says Moore. "It just gives them a security blanket. They might as well not use [the gp120 boost] at all."

McMichael, with support from the privately funded International AIDS Vaccine Initiative (IAVI), plans next spring to start trials of a different prime-boost approach that he hopes will produce much higher levels of CTLs. He believes that priming with the poxvirus presents the immune system with too many proteins from poxvirus as well as HIV, so he intends to use a prime vaccine that contains HIV genes stitched into a stretch of DNA called a plasmid. He theorizes that this so-called DNA vaccine, which can infect cells and produce viral proteins, will focus the immune system's attention; he then hopes to boost these primed CTL responses with a modified vaccinia virus that holds HIV genes.

Although Merck is sketchy about its vaccine plans, it, too, is focusing on triggering strong CTL responses. Emilio Emini, a virologist who heads the company's vaccine program, says that, like McMichael, the company is working on a DNA vaccine. It also is developing a live, but defective, viral vector that Emini declines to discuss publicly. "One of the reasons we've kept a low profile is we don't want to raise expectations," he says. "The likelihood for failure is pretty high." Then again, he says, Merck is putting a lot of resources into the project. "It's a big program for us."

Wayne Koff, who formerly headed the AIDS vaccine program at NIAID and now is scientific director at IAVI, hopes Merck is more committed to its vaccine program than in the past. More support from the pharmaceutical industry is sorely needed, says Koff, who notes that NIH has fewer trials under way now than he has seen in 10 years. "Right now we're at a nadir. But it's clear there will be a lot more vaccines in trials in a few years." —JON COHEN

MARINE CHEMISTRY

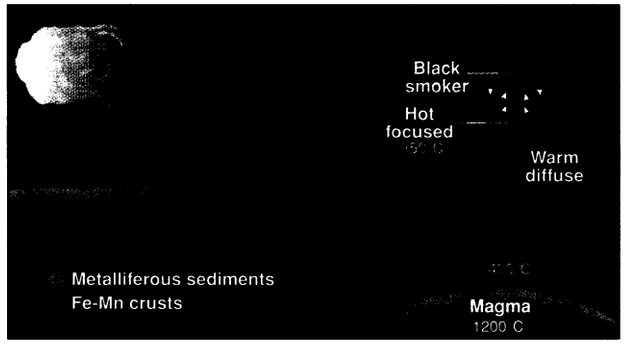
A Cooler Way to Balance The Sea's Salt Budget

Mineral-laden volcanic springs in the deep sea had seemed to explain the ocean's chemistry, but cooler springs away from the volcanoes may play a bigger role

The hot springs and billowing black smokers of the deep sea looked like a spectacular answer to a long-standing mystery when they were discovered in the late 1970s. Perched along the crest of the volcanic midocean ridges, these spouts of mineral-laden, often blistering-hot water not only hosted a menagerie of bizarre animals but promised to be the missing factor that balances the ocean's chemical books. Seawater isn't simply river water concentrated by eons of evaporation; it contains too much of some minerals and too little of others. Ridge-crest hydrothermal activity—where seawater sinks into the crust, is heated and chemically transformed by hot rock, and then gushes back into the sea—looked like it might explain these disparities. But it now appears that a cooler, gentler interplay of water and rock may play a far bigger role in setting seawater's composition.

On page 721 of this issue of *Science*, oceanographers Stephanie de Villiers of the University of Cape Town in South Africa and Bruce Nelson of the University of Washington, Seattle, present high-precision measurements tracing a plume of chemically altered seawater that includes water from warm springs kilometers away from the seething black smokers of the ridge crest. "This is a very exciting discovery," says oceanographer Michael Mottl of the University of Hawaii, Manoa. "If proved to be correct, it will solve a lot of problems. The data point to the importance of hydrothermal activity other than the spectacular black smokers that have gotten so much attention."

Until the discovery of deep-sea hot springs, oceanographers had hardly a clue about how or where seawater took on its distinctive composition. Rivers carry in so much sodium, magnesium, and potassium that the ocean should be far richer in these elements than it is. Calcium presented the opposite problem. Shell-forming plankton appeared to be taking calcium and carbonate out of seawater and incorporating it into sediments twice as fast as rivers carry the metal into the sea. But black smokers seemed to be balancing the books. Seawater was sinking into the fractured ridge crest, picking up heat, calcium, and other elements from the rock, leaving behind its own magnesium, and rising back into the sea.



Sea-floor chemical factory. Searing hot seawater reacts with crustal rock to produce dramatic black smokers at the ridge axis, but cooler waters far from the smokers may have a greater effect on seawater chemistry.

But then surveys began suggesting that superhot water might not be the only factor controlling seawater chemistry. As Keir Becker of the University of Miami in Florida and Andrew Fisher of the University of California, Santa Cruz, will soon report, something like 10 times more fluid seeps from the flanks of the ridge than from the crest. Although this water interacts with the crust at lower temperatures—20° to 200°C, compared with 350°C degrees for a black smoker—it too might deposit some minerals and pick up others, transforming the ocean's chemical composition.

Compared to black smokers, with their heat and dramatic mineral formations, these warm springs are hard to find. So de Villiers and Nelson developed a procedure for analyzing seawater using mass spectrometry that was precise enough to pick up telltale variations in the chemistry of deep waters that have flowed across a ridge. Catching a ride on a research ship that happened to be crossing the East Pacific Rise at 17.5°S, de Villiers collected seawater from the surface to near the bottom above and to the west of one of the most active midocean ridges in the world.

Armed with the high-precision technique, de Villiers and Nelson found a plume of water trailing off to the west of the ridge in which magnesium was depleted by as much as 1% and calcium was enriched, just as expected from hydrothermal alteration. To work out the proportions of plume water from black smokers and tamer warm springs, they checked helium isotope measurements made near their sites by other researchers. The lighter isotope of helium, helium-3, is a signature of black smokers, because only the hottest water manages to extract helium-3 from the newly formed rock of the ridge crest.

Helium-3 was scarce in the plume given the amount of missing magnesium, leading the researchers to estimate that "the low-

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