"We need to show [successful] gene transfer into marrow cells in vitro," Miller says. The ability to transfer genes into large animals, which may represent important differences compared with mice, needs testing. The persistence of the genetically engineered virus in the host animal without adverse effects also must be demonstrated, he adds. "I think we will need a committee to review all the technical aspects of a proposal."

That is precisely what RAC and its working group on gene therapy seem eager to be and do. □

Understanding the Genetics of Bacterial Invasiveness

Scientists are uncovering the genetic machinery that enables certain bacteria to invade cells and cause disease. The findings may pave the way to more effective vaccines and the development of new ways to combat infections. "If we can gain knowledge about particular steps in bacterial growth, such as invasion, then we may be able to design therapies that will block those steps without involving antibiotics," says Ralph R. Isberg of Stanford Medical School's department of medical microbiology.

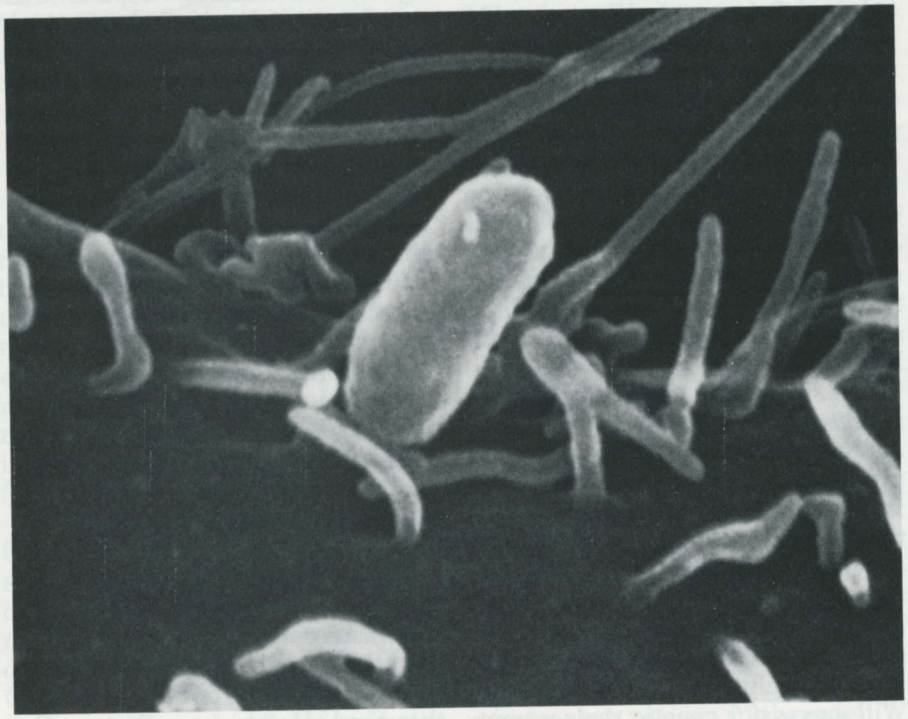
Isberg and Stanley Falkow recently uncovered the gene that allows the bacterium *Yersinia pseudotuberculosis* to invade cells. A close relative of the microbe that causes bubonic plague, this bacterium is responsible for a serious veterinary infection nicknamed "guinea pig plague." Meanwhile, in a separate development, scientists at the Walter Reed Army Institute of Research in Washington, D.C., have identified seven genetic sequences in *Shigella flexneri* that enable it to invade cells. Invasiveness is a crucial step in enabling such bacteria to cause dysentery. Similar genetic sequences have also been obtained from certain strains of pathogenic *Escherichia coli*.

In their search for the gene encoding invasiveness, the Stanford investigators cloned a wide array of genetic sequences from *Y. pseudotuberculosis* and inserted the clones into the cells of a noninvasive strain of *E. coli*. The genetically engineered recipient bacteria were added to cultures of animal cells, which were then subjected to an antibiotic that cannot penetrate mammalian cells. Because only the bacteria that were able to invade the animal cells survived this treatment, the researchers were able to isolate the bacterial strains harboring the genes for invasiveness. Further analysis of the surviving *E. coli* strains revealed that they all shared an identical sequence, which codes for a single protein.

The simple genetics behind the ability of *Y. pseudotuberculosis* to invade cells contrasts with the complex genetic arsenal that *Shigella* species commandeer when penetrating mammalian cells. Samuel B. Formal, Thomas L. Hale, and their colleagues at Walter Reed Army Institute of Research find that, to be invasive, at least seven different polypeptides—presumably the products of seven separate genes—must be produced by *S. flexneri* and also by a dysentery-causing strain of *E. coli*.

Walter Reed investigators also have shown that at least some of the genes for invasiveness apparently are carried on plasmids rather than on the bacterial chromosome. By systematically inserting mutations in different segments of the plasmid carried by *S. flexneri* and then assessing the ability of the resulting bacterial mutants to invade cultured animals cells, Philippe J. Sansonetti found two regions of the plasmid responsible for invasiveness. He notes that 37 kilobases of DNA are required for *S. flexneri* to invade cells, compared with the 3-kilobase segment of DNA that *Y. pseudotuberculosis* needs to be invasive.

Three of the invasiveness-conferring polypeptides are extremely basic, which suggests to the Walter Reed investigators that these polypeptides may play a role in regulating DNA expression or, alternatively, may help the microbe bind to and enter host cell membranes.



Fortified with a gene taken from a disease-causing bacterium, an E. coli *cell (jellybean shape near center) invades a human lung cell.* E. coli *cells that carry only their own genes cannot enter human cells. Fingerlike protrusions are part of the normal outer membrane of the lung cell.*

Photomicrograph courtesy of Stanley Falkow, Stanford University Medical Center. Taken with a scanning electron microscope.

The other four proteins provoke an immune system response in monkeys recovering from *Shigella* infections. "Apparently these four polypeptides, which are quantitatively minor components of the bacterial cell, allow the primate immune system to distinguish invasive bacteria from harmless endogenous intestinal flora," Formal says. The polypeptides might bind to host cell membranes, somehow prompting them to take up the bacteria. Hale and his colleagues are currently testing this hypothesis.

The *Y. pseudotuberculosis* invasion gene codes for a large protein which also is found on the bacterial surface, suggesting that it too facilitates the microbe's entry into host cells.

Both research groups say their findings could lead to development of new approaches to blocking or treating for invasive bacteria. For example, the absorption of antigens from orally administered *Shigella* vaccines has proved to be neither efficient nor preditable. Formal and his colleagues at Walter Reed now have developed a vaccine that they hope will improve absorption.

The new vaccine consists of *E. coli* cells into which the researchers inserted the *Shigella* family of genes, which promote invasiveness and encode the antigens known to evoke a protective immune response from the host. The plasmid enables *E. coli* to invade the epithelial cells of the bowel, increasing the likelihood that the vaccine antigens will reach the antibody-producing cells inside the bowel. Safe and effective when tested on monkeys, this vaccine is expected to enter clinical trials within a year.

The Walter Reed researchers also are developing a vaccine that uses a weakened typhoid strain instead of *E. coli* to transmit the *Shigella* antigens. Preliminary studies indicate that this vaccine is both safe and effective in humans, and an extensive clinical trial is expected to begin by this summer. □

Margie Patlak

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Not Simply Photosynthetic, Lake Algae Also Dine on Bacteria

Photosynthetic algae have been thought to derive their energy from sunlight and their carbon from atmospheric carbon dioxide. Now scientists in Canada report that some common species of lake algae graze on bacteria to supplement their photosynthetic diets (*Science* **231**:493, 1986). This finding changes the picture of the early stages in the aquatic food chain.

The algae obtain at least half their total carbon from ingested bacteria, rather than from photosynthesis, says David F. Bird of McGill University in Montreal, Quebec. Bird and his colleague Jacob Kalff calculate that an alga cell can consume, on average, 36 bacteria each hour. "This is the equivalent to an individual *Dinobryon* . . . ingesting almost 30% of its weight in bacteria per day," they report. Algae of two species of another genus, *Uroglena*, also ingest bacteria, but at only about one-sixth that rate.

Bird and Kalff tested four species of the alga *Dinobryon* found in Lac Cromwell in Quebec. To demonstrate the algae's appetite for bacteria, the scientists mixed the algae with bacteria-sized fluorescent latex beads. The algae ingest the beads, which are easy for the scientists to track, only a little less readily than they ingest bacteria. Electron micrographs also reveal bacteria entrapped in food vacuoles within algal cells.

Groups of relatively sluggish *Dinobryon* cells form cooperative pools to comb the water for bacteria. The single algal cells share a branching fibrous casing called a lorica, into which they insert their flagella. The flagella sweep water into the casing, and cells can then easily engulf passing bacteria.

"Our results show that [algal ingestion of bacteria] is quantitatively important in nature," Bird and Kalff say. Because large numbers of *Dinobryon* cells populate eastern North American lakes, averaging 150,000 to 650,000 cells per liter, they remove more bacteria from the water than do the combined communities of crustaceans, rotifers, and ciliates, the animals long known to eat bacteria. Bird says the algal grazing rates are similar to those measured for marine microflagellates, abundant nonphotosynthetic microorganisms recently recognized as important in consuming algae. He says that the algae and microflagellates provide a way of "returning plant nutrients captured by bacteria back into plants and animals." □

Huge Research Plans for Hughes Medical Institute

Donald Fredrickson, president of the Howard Hughes Medical Institute (HHMI) and former director of the National Institutes of Health (NIH), recently announced plans for spending at least $1 billion over the next 5 years. HHMI will be supporting basic medical research in four areas—genetics, immunology, metabolic regulation, and neuroscience.

More than 900 scientists and research staff will be employed at 22 HHMI laboratories that are either already established or are being planned at academic medical centers in 14 states. Plans call for spending over $100 million on Boston-area laboratories and hospital research facilities, including Massachusetts General Hospital, Children's Hospital, and the Brigham and Women's Hospital units of the Harvard Medical School.

The institute's charter calls for promoting "human knowledge within . . . the basic [medical] sciences . . . and the effective application thereof for the benefit of mankind." Formally chartered in 1953, the institute underwent a growth spurt at the end of 1985 when its endowment grew by $5 billion with the sale of Hughes Aircraft stock. HHMI is