Cystic fibrosis puzzle coming together

People with cystic fibrosis begin life with lungs that appear normal. Yet a genetic flaw leaves them singularly vulnerable to the legions of microbes in every breath they draw. Their lungs soon become infected and clogged with mucus, where germs thrive.

A new report now dovetails with one published last year to explain why this downward spiral occurs. Ultimately, the studies may lead to new treatments for this fatal illness.

The latest study indicates that infection with a bacterium known as *Pseudomonas aeruginosa* trips a previously unknown genetic switch in the epithelial cells that line the lungs, causing these cells to churn out oceans of abnormal mucin, the primary component of mucus.

The earlier report showed that the genetic defect that gives rise to cystic fibrosis destroys a natural antibiotic in the lungs.

"We're beginning to see how the pieces in the puzzle are related," says Carol B. Basbaum of the University of California, San Francisco. She and her colleagues report their findings in the Feb. 4 PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES.

Normal lungs are well equipped to ward off infection. Mucin lubricates lung tissue and traps germs and foreign particles. Hairlike projections known as cilia then gently sweep this mucus toward the throat, where it can be expelled from the respiratory tract. The lungs also produce the natural antibiotic defensin, which eradicates invading microbes.

Cystic fibrosis cripples these defensive mechanisms.

In people with the disease, mucin is abnormal. It is too thick to flush microbes away. Instead, says Basbaum, "you get stagnant pools of mucus, which is a wonderful breeding ground for bacteria. They feed on mucus, and it protects them from the immune system. The bacteria keep sending signals to the cells to make more and more.

"It's a positive spiral that wasn't recognized before."

Basbaum and her colleagues found that *P. aeruginosa* touches off a chain of enzyme reactions that switches on at least one, probably two, and possibly more of the genes that control the manufacture of mucin.

The San Francisco team's research follows an earlier report describing another mechanism by which lungs protect themselves from infection (SN: 4/4/96, p. 279). The genetic defect underlying cystic fibrosis produces an abnormality in the openings, called chloride gateways, in epithelial cells. This defect prevents salt from entering the cells, thus leaving the lungs bathed in brine. The brine disables defensin, which guards against a range of lung infections.

The destructive processes described in the two studies combine to devastate the lungs.

The toxins produced by *P. aeruginosa* poison epithelial cells. White blood cells dispatched to combat the infection cannot penetrate the thick bed of mucus harboring the bacteria, so they begin to attack lung tissue. Eventually, this dual onslaught turns the lung's air sacs into bloated cysts. Only a lung transplant can avert death.

Basbaum and her colleagues began their research using tissue from excised, diseased lungs and from the normal lungs that would be transplanted to replace them. To refine their observations, the researchers needed a more abundant source of cells, so they shifted to laboratory-grown lung cancer cells.

The researchers found that infecting lung cells with *P. aeruginosa* produced a 10-fold increase in the activity of *MUC2,* a mucin-producing gene. Another mucin-producing gene, *MUC5,* also switches on, but the group's studies of this gene remain preliminary. Nevertheless, Basbaum says, "we think this is a global phenomenon, in which the host responds to bacterial infection by [boosting] mucin production."

Scott Randell of the University of North Carolina at Chapel Hill observes that the study may lead to new ways of treating the illness. "Cystic fibrosis is always called the mucus hyperproduction disease. Clearing mucus is a big part of physiotherapy. Yet there are no therapies directed specifically at mucin hypersecretion itself."

Basbaum's group cites one avenue as worthy of exploration—tyrosine kinase inhibitors, which interrupt *P. aeruginosa's* signaling cascade and slow mucin production. The challenge, Basbaum says, is to find an inhibitor that works without disrupting necessary cell functions. —*S. Sternberg*

Understanding the language of reproduction

Chemical conversations between developing mammalian egg cells and the cells that surround them are conducted in a language scientists don't understand and through channels they couldn't identify—until now. By deleting a single gene from mice, researchers have identified one such channel and may at last be on the way to understanding the dialogue that helps an immature egg cell develop.

The gene the researchers deleted encodes connexin 37, a protein found in ovaries. Connexin 37 seems to link the developing eggs, or oocytes, and the surrounding cells, called granulosa cells. Female mice lacking the gene are infertile, report David L. Paul of Harvard Medical School in Boston and his colleagues in the Feb. 6 NATURE.

The finding confirms previous suspicions that connexin 37 forms a channel through which ions, metabolites, and signaling molecules can pass.

Oocytes and granulosa cells together make up the follicles of a normal ovary. As a follicle develops, the number of granulosa cells increases. The oocyte grows and undergoes the biochemical changes that lead to maturity. It also begins meiosis, the process that gives it a single, rather than double, set of chromosomes. When the mature egg cell is expelled from the ovary, granulosa cells change into the steroid-secreting cells needed to support a developing embryo.

"When connexin 37 is absent, the whole process falls apart," says Paul. Neither the granulosa cells nor the oocyte grows properly, and the oocyte fails to start meiosis. The granulosa cells still change into steroid-secreting cells, but the oocyte disappears.

The steps of the failed developmental process offer clues to the messages that the connexin 37 channels convey.

"We believe the granulosa cells may send at least two types of messages to the oocyte," Paul says. A positive signal tells the egg to grow and prepare for meiosis, he argues, but there's also a negative signal that delays completion of meiosis until ovulation has occurred.

A message also travels the other way, from the oocyte to the granulosa cells, Paul says. This message says, "keep on being a regular granulosa cell; don't differentiate into a steroid secretor."

To understand the process better, scientists need to identify the chemicals that make up the signals being passed through the channels, Paul says.

John Eppig, a reproductive biologist at Jackson Laboratories in Bar Harbor, Maine, calls the study "exciting" but cautions that scientists don't think connexin 37 carries all the communications between the oocyte and granulosa cells.

"This is an area where we are really just beginning to get a feel for how important and how complex the communication is between cells," Eppig says.

A possible next step to pinpointing the actions of the signals, Eppig says, is to remove the oocyte from mice lacking connexin 37. Normal oocytes removed from a follicle mature spontaneously, as if they had already received the positive developmental signal. If an oocyte removed from a connexin 37-deficient mouse failed to mature, it would suggest that the protein is necessary for that signal to be effective. —*P. Smaglik*