then drawn into the cell's interior and the cholesterol freed in a compartmentlike cell organelle called a lysosome.

Brown and Goldstein found that cholesterol ultimately gets incorporated into the cell membrane or repackaged for storage or breakdown. They and others also showed that when a cell is glutted with cholesterol, feedback mechanisms kick into action: The cell stops making LDL receptors and shuts down its own cholesterol-synthesizing processes.

Even though Brown and Goldstein worked out much of the beginning and end of cholesterol's journey, "how free cholesterol moves out of the lysosome and into other parts of the cell [remained] a black box," explains George Rothblat, a cell biologist at the MCP-Hahnemann School of Medicine in Philadelphia. "This is where the defect is in Niemann-Pick disease." Somehow, the defective protein hampers the cholesterol's movement through the cells and the feedback mechanisms that control its level. "It's a protein that's screaming for attention," says Liscum.

Already, an examination of the NPC1 protein has revealed a stretch of amino acids where it resembles other proteins known to interact with cholesterol to regulate its level. Another section of the NPC1 protein looks as if it may somehow interact with the lysosome. In between are 16 regions with sequences that indicate, says Carstea, that "this [protein] is locked in a membrane" somewhere inside the cell.

Making sense of those clues will take more study, but in the meantime, the gene may lead to other trafficking molecules. "There could be a whole family of these proteins," says Goldstein. For example, a very small subset of people with Niemann-Pick type C disease have a different genetic defect, located on another chromosome. It affects a protein that seems to function as a trafficking molecule, much like the NPC1 protein.

The identification of the *NPC1* gene may also be a step toward developing a treatment. "We're hopeful that having the gene will let us get at a therapy faster," says Liscum, who hopes to use cell cultures and mice to screen for drugs that might slow cholesterol buildup in Niemann-Pick children. She cautions, however, that "it's going to be a long time coming in having an impact on kids."

While Cindy Parseghian realizes that the new gene find may not help her daughters, she thinks it may help other family members, by leading to a way to test for the mutant gene in those who do not have symptoms, including her one healthy son. In the meantime, she and her husband make the best of the time they have with their daughters. "We can look at our children as if they were dying, or we can look at them as though they are living," she points out. "They look at life as if they are living, and that's what we try to do."

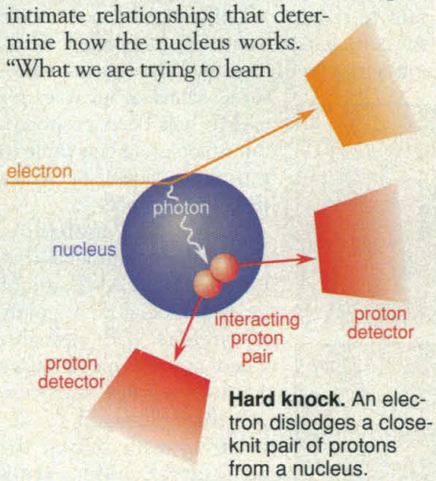
–Elizabeth Pennisi

NUCLEAR PHYSICS

Accelerator Probes the Private Lives of Nucleons

Census data can tell us much about how the average person lives, but they inevitably miss the interplay between individuals that ultimately makes our societies tick. Nuclear physicists face similar difficulties in their attempts to fathom the crowds of particles that populate the atomic nucleus. Nuclei contain as many as a few hundred protons and neutrons, collectively known as nucleons, as well as a sea of other particles that bind the nucleons together. The interaction of so many particles is immensely complicated, so physicists have had to content themselves with models that average out nucleon behavior.

Now, however, a team at NIKHEF, the Netherlands's National Institute for Nuclear and High-Energy Physics in Amsterdam, has been able to coax pairs of protons out of the nucleus just at the moment when their interactions are at their most intense, revealing the intimate relationships that determine how the nucleus works. "What we are trying to learn is how the fact that the two nucleons are bound in the nucleus influences their mutual interaction," says NIKHEF's Gerco Onderwater.



Hard knock. An electron dislodges a close-knit pair of protons from a nucleus.

Other nuclear physicists are thrilled to have this intimate glimpse of nucleons, which the Dutch team offers in the 30 June issue of *Physical Review Letters*. "The NIKHEF experiment demonstrates that it will be possible to learn about how protons interact with each other *inside* the nucleus," says Wim Dickhoff of Washington University in St. Louis. Adds Carlotta Giusti of the University of Pavia in Italy: "This is a fundamental milestone in the understanding of nuclear structure."

Each nucleon is made up of three quarks bound by the strong nuclear force. This force also holds nucleons together as they exchange force-carrying particles called mesons. However, calculating the details of how the hundreds of nucleons and mesons interact is impossibly complex, so physicists fall back on statistics. They define a single, averaged force field, or mean field, which describes how a nucleon interacts with its averaged neighbors. Mean-field models "are able to reconstruct and explain a lot of details with enormous accuracy in nuclear structure," says Onderwater. "Until now, these mean-field approximations worked fine."

But experiments are beginning to reveal cracks in these models. According to Onderwater, experiments using electrons to knock out single protons only seem to see about 65% of the proton population in the nucleus. Moreover, the momentum of many of the knocked-out protons is unacceptably large according to mean-field models, which predict that such boisterous nucleons should spontaneously fly out of the nucleus. "A proton in the nucleus spends only a fraction—albeit a sizable one—of its time in this average field," says Dickhoff. "In fact, because it interacts strongly and collides with other nucleons, it spends about 35% of its time doing 'other' things."

Theorists are now attempting to patch up the mean-field model by incorporating short-range correlations (SRCs) between nucleons, describing how nucleons influence only their nearest neighbors and not the nucleus as a whole. The main short-range effect is a switch from attractive to repulsive forces when nucleons are very close together. The repulsion ultimately keeps nucleons apart and prevents nuclei from collapsing to a fraction of their normal size.

In this picture, nucleons consist of a repulsive quark core surrounded by a cloud of attracting mesons. "The high energies and momenta arise from violent collisions in which the quark cores touch each other," says Derek Branford of the University of Edinburgh in the United Kingdom. The NIKHEF experiment, he says, sets out to test this image. "It is designed to catch two protons at the time they are so close that the quark cores are touching, and knock them out of the nucleus."

To do this, the NIKHEF team modified its Amsterdam Pulse Stretcher electron accelerator, transforming its beam of brief electron pulses into a more or less continuous beam, to increase the chances of catching close interactions. The beam, which has an energy of 584 mega–electron volts, is trained on a water target surrounded by three detectors. One captures electrons that bounce

from the target, while two newly built proton detectors track protons knocked from oxygen nuclei in the target. A scattered electron and two protons emitted all at once signal a rare double knockout event; the group spotted about 5000 of these in all.

From the momenta and energies of the scattered electron and emitted protons, the researchers can determine the protons' original relationship inside the nucleus. Particular patterns of momentum mark proton pairs that were originally close neighbors, on the point of being flung apart by the repulsion between their quark cores.

Focusing on those events that involved closely interacting protons, Onderwater and his colleagues were able to reconstruct the energy states of the original nuclei, which correspond to the energy of the closely interacting proton pairs. "This is the first experiment of this kind that is really able to make a distinction between the different states," he says.

By comparing their results with recent calculations that attempt to accommodate SRCs, Onderwater's team concludes that about 70% of the proton pairs they detected were originally interacting very closely. The finding supports a picture of the nucleus in which many nucleons have a higher momentum than a mean-field approach alone would predict. That would explain why earlier single-proton knockout experiments, looking at only low momenta, could not see many of the protons.

The technique should eventually reveal even more details about SRCs, says Onderwater. "It's aimed at providing more information on how the mean-field model should be extended in order to incorporate this strong repulsive core in [nucleons]," he says. Soon, the private lives of nucleons may be a mystery no more.

–Andrew Watson

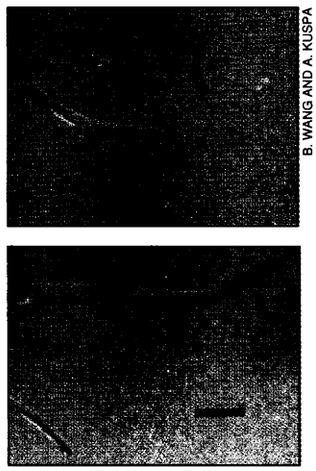
Andrew Watson is a science writer in Norwich, U.K.

DEVELOPMENTAL BIOLOGY

One Molecule Orchestrates Amoebae

For a mere soil-dwelling amoeba, *Dictyostelium* manages some impressive feats of organization. When food is scarce, individual cells band together with their comrades to form a mound that crawls to better pastures and disperses by building a stalk and a spore body. Researchers have struggled for years to understand the signals that coordinate this multifarious behavior, hoping to gain insights into the development of more complex organisms. Now, with a paper in this issue, the picture has been dramatically simplified.

On page 251, Bin Wang and Adam Kuspa of Baylor College of Medicine in Houston show that a single molecule—an enzyme called PKA—can single-handedly drive all steps of development: clumping together; forming the cell cluster or slug; and differentiating into stalk or spore cells. The finding narrows the role of another molecule, cyclic AMP (cAMP), which activates PKA and can also act as a signal in its own right. Researchers had suspected that cAMP might work via many other pathways, says longtime *Dictyostelium* researcher William Loomis of the University of California, San Diego (UCSD). Now, "the paper shows that essentially all of the internal cAMP responses are mediated by PKA."



B. WANG AND A. KUSPA

Migratory slugs. *Dictyostelium* mutants lacking cAMP *(top)* behaved like normal organisms. (Bar equals 0.25 millimeter.)

Besides telling *Dictyostelium* researchers that they needn't look for alternative cAMP signaling pathways to drive development, the finding also has an important message for researchers studying higher organisms. There, PKA and cAMP are key actors in everything from embryogenesis in fruit flies to memory in mice, and the new finding suggests that PKA may have the leading role. Says Loomis, "The universality of this is very important."

For decades, cAMP has been the center of attention in the study of *Dictyostelium* development. The molecule, synthesized by an enzyme called adenylyl cyclase, initially seemed to have many faces. Inside individual cells, it binds to PKA and sparks it into action to trigger cell differentiation. Outside the cells, it acts as a molecular magnet, drawing single cells into well-integrated structures. Many other roles for cAMP had been proposed, but no one had been able to pin them down or gauge their importance.

Kuspa and Wang had run across mutant amoebas that hinted that cAMP might be less crucial than many researchers had thought. These mutants had inactive adenylyl cyclase enzymes, yet they managed to develop normally. Given the many open questions about cAMP's roles, Kuspa says he and Wang, a graduate student in his lab, decided to take a closer look at such mutants. "We had been staring at the data for 2 years, and we just had to test this the best way we knew how."

They began with a well-characterized adenylyl cyclase mutant—one that essentially made no traces of the enzyme—provided by Peter Devreotes at Johns Hopkins University in Baltimore. Next, the Baylor researchers genetically engineered the mutant so that its cells produced high levels of a mutant PKA that was permanently turned "on," even without cAMP to activate it. The Baylor team then watched how these microorganisms developed over the course of 24 to 30 hours.

To their surprise, the mutants looked and behaved virtually the same as normal organisms. Moreover, when the researchers tested the activity of specific genes that are turned on and off at particular times in *Dictyostelium* development, they observed a normal pattern. "The cell-type specific genes were turned on at the same time as wild-type," says Kuspa. "That was probably the most shocking thing."

Not only was the cells' internal signaling taking place normally, but the collective behavior of the cells also appeared normal. They bunched together and formed stalks and spores—events that researchers had thought could only be orchestrated by pulses of cAMP. Only when the density of cells was low did the lack of cAMP seem to prevent the mutants from building normal mounds.

"The intriguing thing is that the organism can bypass a number of events that normally require cAMP in the wild," says developmental geneticist Richard Firtel of UCSD. Kuspa believes the mutants can get around the need for the chemical signal by bumping into each other, triggering alternative signaling pathways that require only PKA.

At first sight, the results appear at odds with Firtel's own findings. He and his colleagues had created similar mutants and found that they failed to develop normally. But Firtel thinks the discrepancy may stem from "too much of a good thing." While Kuspa's mutants produced twice the normal levels of PKA, Firtel's organism overproduced the enzyme in much greater amounts. "It just means that you need a specific amount of PKA ... to produce the observed phenotype," he says.

If PKA in just the right amount can serve as a master coordinator of development, "the subtleties go way beyond Wang and Kuspa," says Loomis. "The findings could be used as a guideline for studies of all vertebrate and invertebrate development—in addition to being fascinating for *Dictyostelium* in and of itself."

–Trisha Gura

Trisha Gura is a science writer in Cleveland.