done by removing one cell from an embryo created in vitro and using gene amplification techniques to produce enough DNA for analysis. But that method fails in about one-third of the cases, and better results might be obtained by using a cloned two- or three-cell embryo copy for analysis.

But opinions divide sharply about whether this application would be ethical in view of the fact that one clone would be destroyed by the analysis. "You'd essentially have the situation of one identical twin being sacrificed for the sake of the other," says Somerville. The president of the American Association of Bioethics, Arthur Caplan of the University of Minnesota in Minneapolis, also finds the idea disturbing, saying that the idea of "creating embryos solely for the purpose of genetic diagnosis is morally suspect." In contrast, another bioethicist, John Robertson of the University of Texas, Austin, who's a member of the American Fertility Society's ethics committee, says he thinks the idea of using two- or three-cell clones for diagnosis is not much different from taking a single embryonic cell.

For the present, there seem to be no regulations or guidelines that could help researchers navigate these ethically treacherous waters. The United States has not had a bioethics commission since 1989 when the congressionally appointed Biomedical Ethics Advisory Committee expired in political disarray without ever issuing a report. At Congress's request, however, the Office of Technology Assessment has prepared a report reviewing past efforts, with an eye to setting up a new ethics commission. (The report, "Biomedical Ethics in U.S. Public Policy," was released on 14 October.) If Congress does establish such a commission, human embryo cloning would presumably come under its purview.

But for now, the fertility society comes the closest to having guidelines, although they deal with "pre-embryo research," rather than with human embryo cloning and its applications. They stipulate that researchers can experiment with pre-embryos not intended for implantation if they are exploring an issue of clinical importance and obtain clearance from their institutional review boards. Hall obtained such clearance, and his experiments did appear to comply with the fertility society's current guidelines, says Edward Wallach of Johns Hopkins University School of Medicine, who chairs the society's ethics committee. Wallach also says that his group will likely develop more specific guidelines for embryonic cloning in an ethics report expected in January. In view of the Brave New techniques reported at the Montreal meeting, January will be none too soon.

–Rebecca Kolberg

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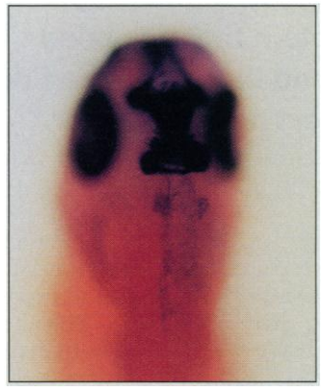
DEVELOPMENTAL BIOLOGY

New Protein Appears to Be Long-Sought Neural Inducer

Some of the most critical events in our lives happen before we are even born. Take for example gastrulation, when the ball-shaped embryo buckles, and cells stream inward to form three layers which respond to a myriad of chemical signals that tell them which mature tissues to become. Most of our tissues and organs—including the brain and nervous system—have their origins in this key event. In the case of the nervous system, part of the top cell layer, called the ectoderm, follows orders to become a plate of neural tissue that later folds inward to form the spinal cord and brain. This "neural induction" was discovered more than half a century ago, and although biologists have repeatedly searched for the molecular signal that triggers it, they have always come up empty-handed.



What noggin does. The dark areas show regions of a tadpole head where noggin turns on a gene indicating neural tissue is being formed.

Now, on page 713 of this issue, developmental biologist Richard Harland and his co-workers at the University of California, Berkeley, report finding that an embryonic protein they discovered last year, called "noggin," acts as a neural-inducing signal in frog embryos. The noggin gene has been found in rodents as well, suggesting it may perform similar functions in mammals. "It's an exciting result," says Salk Institute developmental biologist Chris Kintner. "It brings a phenomenon [neural induction] that has been known for many years to a better molecular understanding." Adds developmental biologist Jonathan Cooke of the National Institute for Medical Research in London: "Lots of people are interested in noggin....It is the only protein that has been found that is a bona fide direct neural inducer."

It's not only developmental biologists who are excited. Noggin has also been found in the brains of adult rats, raising some neuroscientists' hopes that the molecule may be a new type of chemical signal, with multiple roles in nervous system development and function. If that is the case, noggin might someday prove useful as a treatment for nerves damaged by disease or trauma, a possibility that has at least one biotech company interested. But in spite of the manifest enthusiasm, researchers caution that much more work is needed before they accept definitively that noggin is a player in either neural development or the adult brain.

Still, Harland's results already represent the solution to a mystery that began with experiments—some dating to the early decades of this century—in which researchers transplanted pieces of tissue from one developing embryo into new locations in another and watched to see what happened. These transplants revealed that the yolky bottom half of the embryo, known as the vegetal hemisphere, produces a chemical signal that tells a band of cells at the embryo's equator to become mesoderm, the tissue that streams inward at gastrulation, and later forms muscle, blood, and bones.

The transplantation work also showed that the mesoderm releases a welter of chemical signals. Even before gastrulation begins, one patch of mesoderm—called the "Spemann organizer" after its discoverer, German embryologist Hans Spemann—begins sending signals that tell part of the mesoderm to make dorsal, or back-of-the-body, structures such as the vertebral column, as well as signals that tell the adjacent part of the ectoderm to form the neural plate.

Researchers lost no time in starting to look for the molecules that determine mesoderm formation and the subsequent events, but for many years their efforts were unsuccessful. The hunt for the neural inducing factor proved particularly confusing—not because there were no results, but because the work produced an embarrassment of riches. Indeed, a long list of compounds, most of them biologically irrelevant, could trigger ectoderm to become neural tissue in the newt and salamander embryos commonly studied at that time. Even though those experiments were done in the 1930s and '40s, they had a lasting—and chilling—effect on the field, says developmental biologist Jonathan Slack of the Imperial Cancer Research Fund at the University of Oxford. "Even when I came in, in the mid-1970s, people said neural induction is completely nonspecific, you can't do anything with it."

It wasn't until the late 1980s that researchers began finding a way out of this

morass, when they began to discover molecules that figure in mesoderm induction in a different amphibian: the African clawed frog *Xenopus laevis*. But even in *Xenopus*, the sought-after neural-inducing molecule was nowhere to be found. It was that absence that drew the attention of both Harland and postdoc Bill Smith, who joined the lab in 1991. "There had been a lot of attention on mesoderm induction," says Harland, "and so when Bill came he decided to make a concentrated effort to find a neural inducer."

Smith began by searching through a collection of messenger RNAs (mRNAs) from gastrulation-stage frog embryos, to see whether any of their protein products would cause the formation of neural tissue in embryos that had been experimentally treated to prevent them from making any dorsal parts on their own. "We thought if we were lucky," says Harland, "we would get little patches of disorganized neural tissue." Instead, they found that a protein made from one pool of mRNAs would "rescue" the treated embryos, enabling them to form apparently normal dorsal structures. Smith went on to clone the gene for the protein that was responsible for the rescue, which the researchers named noggin because high doses produce embryos with exceptionally large heads.

Noggin "is the only protein that has been found that is a bona fide direct neural inducer."
—Jonathan Cooke

The fact that noggin caused dorsal tissue to form in frog embryos didn't necessarily mean it was the long-sought neural inducer. Other dorsalizing factors had already been found, and some of them caused formation of neural tissue indirectly, by triggering the production of dorsal mesoderm, including the Spemann organizer; signals from the organizer then caused neural induction. To prove that noggin was different from these factors, the Harland team had to show it would induce neural tissue formation in pure ectoderm that had been removed from an embryo.

At first noggin seemed to fail this test, but graduate student Anne Knecht kept the team's hopes alive when she showed that injecting noggin mRNA into early embryos causes neural tissue to form in pure ectoderm that was removed from the developing embryo before it could receive mesodermal neural-induction signals. Subsequently the group found the reason for noggin's original strike-out: There wasn't enough noggin in the preparation used. A more concentrated preparation made by graduate student Teresa Lamb, as well as pure noggin protein provided by collaborators at Regeneron Pharmaceuticals in Tarrytown, New York, both passed the direct neural-inducing test.

Noggin passed another key test when the Harland group showed that the protein is made in the Spemann organizer, which is known to be the site of production of the neural inducer. All in all, those results make noggin a solid candidate for the neural inducing factor. "It has two things going for it," says Salk's Kintner. "It has the right biological activity and it is in the right place at the right time."

But though the body of evidence points in the direction of noggin, the proof isn't airtight. The most worrisome problem is that high concentrations of the protein—20 times higher than those required for inducing dorsal mesoderm—are needed to trigger formation of neural tissue from ectoderm. Harland suggests that such high concentrations may be necessary because of the artificial nature of the experiments: When ectoderm tissue is removed from the embryo, it tends to curl up into a ball, making it hard for noggin to penetrate. But some skeptics suggest that there's another, less pleasing explanation. Noggin "may not be the endogenous [neural-inducing] factor" after all, says developmental neurobiologist Jane Dodd of Columbia College of Physicians and Surgeons. This concern was echoed by several other researchers who suggested that at high enough concentration noggin might simply mimic the effects of the actual, and as yet undiscovered, inducer.

To rule out that possibility, noggin needs to meet yet another test. "In order to prove that a substance is really doing a particular job in vivo," offers Oxford's Slack, "if you take it away, the process has to fail." One way to do this would be to create "knockout" animals, in which the noggin gene is inactivated, to see what effect gene loss has on embryonic development. Harland is working with Andrew McMahon at Harvard University to knock out the noggin gene in mice and see how their nervous systems develop in its absence.

Even if noggin proves necessary for nervous system development, Harland points out, that doesn't mean the puzzle of early nervous system formation has been fully solved. The Berkeley group has found that neural tissue induced by noggin has characteristics suggesting it is destined to become the forward part of the brain, implying that other molecules are needed to trigger the formation of hind brain and spinal cord. And even for forebrain, noggin gets the differentiation process going, but isn't sufficient to make the tissue to go on to become full-fledged neurons.

Despite those caveats, noggin's neural inducing ability remains an exciting finding, and researchers would like to know how it acts. The protein's amino acid sequence suggests it is secreted into the extracellular fluid, and it presumably exerts its effects by binding to a receptor on target cells. But noggin bears no more than a minor resemblance to other known proteins, and, as a result, the Berkeley team still has few clues to noggin's function.

One intriguing suggestion about its mode of action comes from developmental biologist Doug Melton of Harvard University, and is based on a discovery made recently in his lab by then-postdoc Ali Hemmati-Brivanlou, who is now at Rockefeller University. Hemmati-Brivanlou found that blocking the action of activin, a signaling protein that acts early in mesoderm induction, caused ectodermal cells to differentiate into neural tissue. "It's as if neural [development] is a default pathway," says Melton. Noggin's role, he says, may be to block the actions of activin in ectodermal cells, allowing them to express their basic "neural nature." That might be a surprise to neurobiologists, adds Melton, because traditional thinking has often assumed that the neural state is "the highest to which a cell can aspire," rather than being merely a default state.

That's unlikely to be the last surprise noggin has in store for the neuroscience community. Another could be noggin's as yet unknown role in the adult nervous system. Noggin is "expressed in discrete patterns in the adult nervous system," says George Yancopolous of Regeneron, who has been studying noggin in collaboration with Harland. "I think it's a very good bet that it's serving a variety of roles." That finding, along with the fact that noggin doesn't resemble any known families of molecules that act as signals in the nervous system, has the Regeneron team particularly excited. "The major reason we're interested in noggin is certainly the fact that noggin could be the first member of a new family [of signaling molecules]," says Yancopolous.

One thing that sets noggin apart from many other neural signaling molecules is that while many of the other signals are also found in tissues outside the nervous system, noggin is not. "Whatever its role is," Yancopolous says, "it might be quite specific to the nervous system." If that role proves to have therapeutic applications, he adds, noggin's specificity may mean it will have few side effects on non-neural tissue. In that event, noggin might not only be the solution to the 50-year embryological mystery of the brain's beginnings, but it would also be poised to make some contributions to the study and therapy of the mature nervous system as well.

–Marcia Barinaga