temperature was about 1.4 K less in the ^{18}O sample than in the ^{16}O sample—and concluded that this was evidence that the material's superconductivity is caused by a phonon-mediated mechanism. At Bell Labs, Bertram Batlogg, Robert Cava, and coworkers found a much smaller isotope effect and concluded from this and other measurements and calculations that a completely different mechanism is at work.

The researchers were uncertain about why their measurements of the isotope effect differed. The two labs use different techniques to make their Ba-K-Bi-O compounds, and the compositions of the two materials probably varied somewhat. Even if the compositions differed, however, the two materials should have been similar in structure and should have shown similar isotope effects.

Since the results depend strongly on how much of the ^{16}O was replaced by ^{18}O, an overestimate of the replacement percentage would lead to an underestimate of the isotope effect. And Hinks pointed out that since the critical temperature depends on the potassium content, any replacement technique that accidentally removed some of the potassium could skew the results. The Argonne team had this problem originally, caused by an impure ^{18}O supply.

To sort out which team is right on the magnitude of the isotope effect, the experiment will probably have to be repeated by other groups.

The Argonne and Bell Labs groups also differ on their interpretation of the data. The Argonne researchers see the isotope effect as strong evidence for a phonon mechanism. "If the isotope effect is there, it generally means the phonons are doing something," Hinks said.

The Bell Labs workers disagree. "Nobody argues that the phonons are not involved," said Cava, but measurements of the electronic density of states in the Ba-K-Bi-O material lead them to the conclusion that the mechanism is not conventional electron-phonon interactions. The Bell Labs group believes instead that the pairing is caused by electronic excitations, which as a secondary effect create lattice distortions, or phonons. Thus the phonons would just be "along for the ride" and would not be a primary cause of the electron pairing.

"The mechanism is not going to be resolved by one experiment," Cava said. There remains a struggle ahead as the superconductivity community argues whether the Ba-K-Bi-O materials are superconducting because of the well-known, comfortable electron-phonon mechanism or whether it is something different, challenging, and exciting. ■ ROBERT POOL

Joint Soviet–U.S. Attack on Heart Muscle Dogma

Under certain experimental conditions scientists can prod heart muscle cells to divide; whether the research will lead to new therapies remains to be seen

"WE SHOULD BE AWAKENED from a state of dogmatic slumber," says Alexander Mauro, paraphrasing Immanuel Kant but referring to the prevailing state of mind about heart muscle. A long-held conviction is that adult cardiac muscle cells have lost the ability to divide. Because of this, the reasoning goes, people with massively damaged hearts need artificial ones or transplants in order to live.

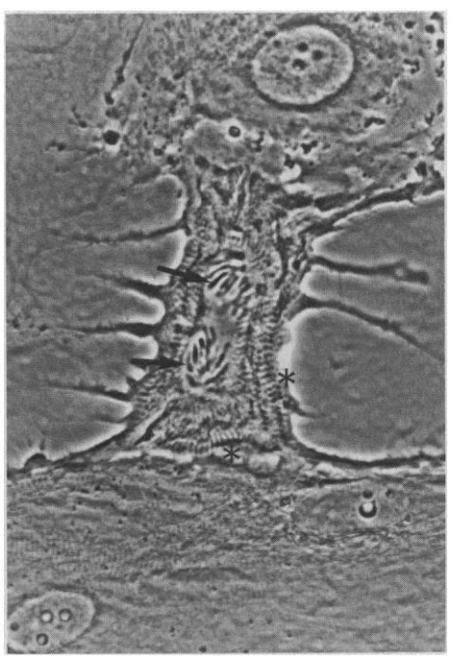
Mauro, of Rockefeller University in New York, and about 25 other researchers met recently to challenge this dogma.* The group, inspired by Pavel Rumyantsev, a Soviet scientist from the Academy of Sciences in Leningrad, is exploring new techniques to trigger mitosis-reluctant cardiac muscle cells into dividing again after they mature. As yet, the field of research is in an early stage. It is not widely recognized and not heavily funded, but its proponents envision future therapies for heart disease patients if some of the new techniques can be applied to human heart tissue.

"The interest in this field is just dawning," Eugene Braunwald of Harvard Medical School told *Science*. "Of all the medical subspecialities, cardiology has really been the slowest to take advantage of recent progress in cell biology and molecular biology." Cardiology, he says, has been dominated by the view that the heart is an electrically driven pump, so much of the research has focused on biophysics, hydraulics, and engineering. Today, that view is changing, but it is a slow process.

The question everyone wants to answer is whether it is possible to repair damaged tissue in the human heart after a heart attack. As yet no answer exists. But basic research on myocytes, the contracting muscle cells that make up about 25% of all the cells in the heart, is in a new phase. Current studies address two fundamental issues: why do heart muscle cells stop dividing shortly after birth and can they be stimulated to divide again? In practical terms most of the research falls into three categories—experiments on intact animals, studies of myocytes in tissue culture, and experiments with transgenic mice that have been induced to grow unusual heart tumors.

Until about 25 years ago, no one had evidence that mature heart myocytes retain the capacity to divide. Then in the early 1960s, Rumyantsev showed that they do. He cut off the blood supply to the left ventricle in young rats, which triggered DNA synthesis and nuclear division in muscle cells of the left atrium. Much of the cardiology community remained unaware of the work until 1977 when Rumyantsev wrote a review on the subject. The work not only stimulated the field, it also established one major experimental approach, namely research on intact animals.

Rumyantsev's experiments raised problems that still exist today. For example, atrial myocytes divide more in young rats, possibly because their regenerative potential is greater than that of older animals. Also, as Victor Ferrans of the National Heart, Lung, and Blood Institute (NHLBI) in Bethesda, Maryland, points out, "You would like to



Newt ventricular myocyte *dividing in a 12-day culture. Both chromosomes (arrows) and myofibrils (asterisks) are visible (×1250).*

*The "Workshop on Myocardial Regeneration" was held on 20 and 21 June at Rockefeller University.

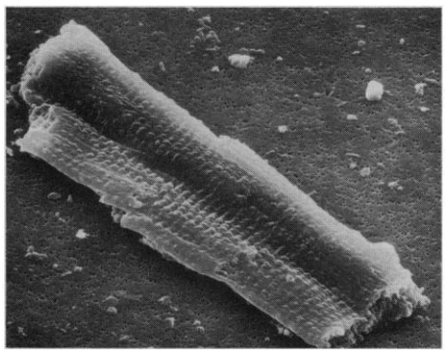
see cells responding in the area of injury, but this is not what you get. Instead you get the response primarily in the atrium." This paradox—mitosis in atrial myocytes after ventricular myocytes are damaged—is still largely unexplained. Finally, although atrial myocytes may undergo cytokinesis—complete division of the cytoplasm to form two new cells—after an infarct in the ventricle, most do not. Instead, they synthesize DNA and form more binucleated cells than normal. No one knows why, but Andrei Borisov and colleagues, also at the Soviet Academy of Sciences, are exploring the issue.

Over the past 10 years, several other lines of evidence have emerged indicating that the ability of cardiac myocytes to divide is repressed rather than lost. For example, as mammals age, an increasing number of their cardiac myocytes have multiple nuclei. In the heart of an adult rat, for instance, about 80% of ventricular myocytes have two nuclei, whereas 35 to 50% of atrial cells are binucleate. In humans, large myocytes may contain four times the normal amount of genetic material. Additionally, disease, exercise, or experimental manipulation can increase myocyte size and the number of chromosome copies and nuclei more than normal. In hypertrophic cardiomyopathy, for instance, the muscle partition that separates the two ventricles grows disproportionately. This may be due to a combination of cell enlargement and cell division—an issue that is not yet settled, says Ferrans.

To John Oberpriller of the University of North Dakota in Grand Forks, all of this clearly indicates that mature cardiac muscle cells retain a capacity to synthesize DNA. But DNA synthesis and nuclear division are not enough if the ultimate goal is to replace damaged heart muscle. Cell replacement requires cytokinesis and that is where much of the research hits its biggest snag.

The strongest in vivo evidence that mature cardiac muscle cells can undergo cytokinesis is in the amphibian. "Our work centers around the newt or salamander," says Oberpriller. "Both in the ventricle and in the atria—15 to 20 days after wounding—the muscle cells undergo DNA replication and mitosis." Amphibians are ideal for these experiments because the blood clots quickly and the blood pressure is so low that it does not blow out the heart wall.

Earlier this year, Oberpriller, Jean Oberpriller, also of North Dakota, A. Arefyeva and Victor Mitashov of the USSR Academy of Sciences, and Bruce Carlson of the University of Michigan in Ann Arbor reported that 45 days after wounding nearly 50% of myocytes in the damaged area of the newt ventricle are labeled with tritiated thymidine, a marker of DNA synthesis. Oberpriller and his co-workers think that this evidence, although indirect, strongly indicates that true cytokinesis has occurred, because about 85% of the labeled cells have a single nucleus and the normal diploid number of chromosomes.



Freshly isolated myocyte from an adult rat, *viewed with scanning electron microscopy (×3000). [W. C. Claycomb and M. C. Palazzo,* Dev. Biol. ***80****, 466 (1980)]*

What does regeneration of the tip of the newt heart mean in terms of the human heart? "No one in his right mind thinks that you can cut off a piece of the mammalian heart and get regeneration," says Oberpriller. "What we are studying is the potential for regeneration and that is why we focus on cell replication." He and Jean Oberpriller now have additional evidence that newt myocytes can divide because they film the cells undergoing cytokinesis in vitro. These and other new studies of cultured myocytes constitute a second major area of research.

Until recently, researchers could not grow adult heart muscle cells in tissue culture. In contrast to myocytes from neonatal animals, which survive well and divide to form a single cell layer that contracts spontaneously and synchronously, the adult cells seemed to die shortly after being put into culture. Then in 1980, William Claycomb of the Louisiana State University Medical Center in New Orleans and his co-workers successfully maintained adult myocytes in vitro. "You get a whole sheet of pulsing cells," says Claycomb.

Still, the adult cells did not divide. So in their new work, Claycomb, Joseph Delcarpio, and Nicholas Lanson, also of Louisiana State, stimulate division by inserting the large tumor antigen from the SV40 virus into the myocyte genome. Cardiac myocytes that contain this transfected gene do undergo cytokinesis, which gives the researchers an opportunity to study molecular mechanisms that govern muscle cell division and differentiation. Perhaps not surprisingly, Claycomb and his colleagues are finding that protooncogenes and anti-oncogenes, which regulate the processes in other tissue, also control them in the cardiac cells.

While these experiments may seem far removed from what happens in the normal human heart, Claycomb offers a possible link—still very much in the idea stage. He proposes that shortly after birth, as nerve endings grow into the heart and release norepinephrine as a transmitter, it may set up a series of responses in myocytes that ultimately affects oncogene expression. "Everything that we have done so far indicates that norepinephrine inhibits cell division and promotes cells differentiation," he says.

A different kind of problem arises when researchers study cardiac myocytes at a very early developmental stage. Using tissue obtained from 16- to 24-week-old human fetuses, Stave Kohtz of Mount Sinai School of Medicine and Bruce Goldman of Temple University Medical School in Philadelphia culture cells that are committed to become myocytes but that lack many characteristics of fully differentiated cells. The strength of the system is that the cells will divide many times in vitro; its weakness, at least so far, is that the cultured precursor cells do not differentiate to a point that matches their in vivo counterparts.

The cells are not all alike when they are put in vitro—some contract spontaneously and some do not. Kohtz and his collaborators select for the noncontracting population, which divides much more rapidly, by adding fibroblast growth factor and insulin-like growth factor–2 to the medium. Under these conditions, the cells replicate quickly and for many generations, but they do not differentiate. Removing the growth factors causes certain markers of differentiation to appear, including atrial natriuretic factor, a peptide hormone normally produced by the atria that helps control blood pressure, and titin, a large protein thought to help with the assembly of contractile proteins.

A third approach toward understanding what regulates heart muscle cell division is to use foreign genes to trick nondividing cardiac myocytes into dividing again. In these experiments, researchers inject a fused gene—the large tumor antigen gene from the SV40 virus coupled to the promoter for a mouse gene—into fertilized mouse eggs. "About 20% of the mouse pups that arise from the microinjection carry the transgene in their cells," says Loren Field of Cold Spring Harbor Laboratory in New York. Field was trying to stimulate division of atrial myocytes, but was surprised because only the right atrium developed a tumor (*Science*, 26 February, p. 1029).

"I did not expect to get an asymmetrical tumor," says Field. He expected tumor growth in both atria because he had linked

the SV40 gene to the gene for atrial natriuretic factor, which is normally produced by both atria. Although Field still does not know why the tumor appeared only in the right atrium, he thinks that a gene in the host mice may play a role because one mouse strain developed tumors much more quickly than another. The tumor cells divided in an uncontrolled way but contracted spontaneously and were similar in other respects to normal myocytes, he says.

Working independently, Jacques Peshon, Richard Palmiter, and Stephen Hauschka of the University of Washington in Seattle and Richard Behringer and Ralph Brinster of the University of Pennsylvania in Philadelphia and their colleagues also produced transgenic mice with tumors of the right atrium—for them, a completely unexpected result. "We were not even thinking that we were going to study cardiac cells," says Hauschka. "This was just serendipitous." The group had expected to stimulate tumor growth in the testes, but instead got tumors in the right atrium of the heart and a section of bone near the ear. Over a 2- to 3-month period, the atrial tumors of the mice became larger than the entire heart.

Hauschka, like Field, does not know why the transgene stimulated asymmetrical tumors. But he and Cindy Gartside have capitalized on it by using the tumors to set up a permanent line of heart muscle cells. "We have been able to maintain cells from these tumors in culture for over a year," says Hauschka. "Some of them still contract." Now they can study cardiac-specific genes, gene regulation, and factors that alter myocyte growth in the cultures, he says.

The long-term goal is to develop new strategies for treating heart disease. "The whole concept is very exciting," says Isidore Rosenfeld of the Rosenfeld Heart Foundation in New York City. "The heart fails for a number of reasons—stress, bad valves, hypertension. But the most common cause is damage from myocardial infarction."

When a person has a heart attack, the muscle tissue disintegrates over a period of several days. "It lyses and is attacked by phagocytes [scavenger cells from the bloodstream]," says Ferrans of NHLBI. "Then after 3 to 4 days fibroblasts invade the muscle and make a fibrous tissue scar." The time delays in these responses may make it possible to intervene in several ways, he suggests—perhaps to prevent extensive muscle loss, retard the growth of the fibrous scar, and also to stimulate myocyte regeneration and differentiation.

Claycomb is about to submit a grant application along these lines that he describes as "kind of science and kind of science fiction." The first part is science. "If we can understand why cells stop dividing then we may be able to restimulate their division." Then comes the science fiction. If researchers learn how to trigger myocyte replication, they may be able to take a biopsy of heart cells from someone after a heart attack, stimulate them to grow in vitro, and place them back into the damaged heart where they could divide and differentiate in a controlled manner. Such a procedure, if successful, would avoid the problem of tissue rejection, which is the major complication with heart transplants.

Despite recent progress in the field, not many scientists are studying heart muscle cell replication or regeneration. "The funding is not good," says one researcher. NHLBI officials are quick to point out that the institute cannot fund a grant unless it gets a high rating during the peer review process. In 1983 the institute began soliciting grants on the development and cell biology of cardiac myocytes, and today funds 22 such grants. While none of them addresses the problem of regeneration specifically, perhaps some of the new information will change that.

■ DEBORAH M. BARNES

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Slices of Continental Crust Coming into View

The study of sections of deep crust exposed at the surface is supplementing expensive geophysical methods and deep drilling

Killarney, Ontario, Canada

THE DEEP CRUST OF EARTH is as exotic as it is unreachable. Under the crushing weight of more than 20 kilometers of rock and temperatures of hundreds of degrees Celsius, rock sweats off its last vestiges of water, transforms its minerals, and, given time, can bend, twist, and tear like soft dough. It is under such conditions that much of the growth and modification of continents occurs. Geologists would love to take a great scoop out of the crust to expose these lower reaches, just as rivers or road engineers slice through mountain sides to reveal their geologic structure.

Lacking godlike powers, earth scientists settle for what the vagaries of volcanoes, continental collisions, and erosion might bring to the surface. This has included plenty of once deep-seated rock from pebble-size on up, but the ideal—vertical cross sections through much of the crust that are still linked to similar rock at depth—have been hard to come by. At a recent workshop in Ontario on continental cross sections,* a group of about 60 geologists, geophysicists, petrologists, and geochemists traded professional tales of cross sections they have known. Thanks to more fieldwork, new techniques, and new perspectives, the selection of cross sections, especially those through a substantial part of the crust, has been considerably expanded.

A key to this improving outlook is the changing perspective of geologists. Perspective can be all important when interpreting rock strata. There is an old saying among geologists—"I wouldn't have seen it if I hadn't believed it"—that certainly applied to Grahame Oliver's experience in New Zealand. In the early 1970s Oliver, who is now at St. Andrews University in Scotland, was the first to make a geological map of the Doubtful Sound area of far southern New Zealand. As he told the meeting, the rocks revealed among the fjords, glaciers, and temperate rain forests of Fiordland (10 meters of rainfall annually) seemed familiar from his training in Europe.

Oliver presumed that, as in European examples, the so-called granulite group of rocks of Fiordland's lowest exposed strata, which had obviously suffered alteration at high temperatures and pressures, would be old, probably older than 500 million years. Those rocks above the granulites would be

*NATO international advanced course on Exposed Cross Sections of the Continental Crust, Killarney, Ontario, Canada, 17 to 27 September 1988.