Limb regeneration in mammals

Research indicates that electricity stimulates partial regrowth of amputated limbs of rats

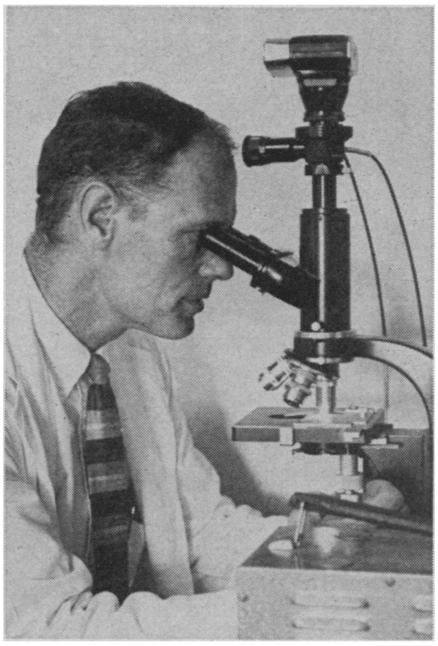
For all its sophistication, modern biology has not been able to explain why certain lower organisms can grow back certain traumatized parts—the worm its lower body, say, or the lizard its tail. Nor has it been able to explain why higher animals, particularly man and other mammals, do not have this capacity. Now Robert O. Becker, research orthopedic surgeon at the State University of New York Upstate Medical Center in Syracuse and also associate chief of staff for research at the Veterans Administration Hospital in Syracuse, believes he has a partial answer. More crucially he has shown that the art of limb restoration in mammals might be capable of development after all.

Becker has partially regenerated the amputated limbs of several dozen rats. The forelimbs were cut off between what corresponds to the shoulder and the elbow in humans. Minuscule amounts of electric current then applied to the severed sites stimulated the limbs to grow down to the elbow. Becker planned to announce his work publicly late this week at a meeting of the New York Academy of Medicine. NATURE will be publishing a technical report soon.

Becker's is the most advanced success yet in regenerating a limb in an animal as high up the evolutionary ladder as the rat. Limbs have previously been partially regrown in a newborn opossum and in adult frogs. Becker's work is also noteworthy because it was undertaken to prove a theory he has doggedly pursued for 15 years: The reason man and other mammals are not able to regenerate damaged limbs is that they have lost the ability to generate enough electricity to provide ample stimulus to the formation of a new limb bud.

If adequate current were applied to a severed limb site, Becker reasoned, it would seem that the current might lead the cells to propagate into tissue and thus into limb muscle, nerve and bone. His success with partial regeneration of rat forelegs seems to indicate that there must be something to this theory. In fact the methods used for partial reconstruction of limbs in the opossum and in frogs, such as transplanting nerve tissue onto the amputated extremity or injuring tissue at the amputated site, could likewise reinforce the theory, since they might increase electrical current flow to the site of amputation.

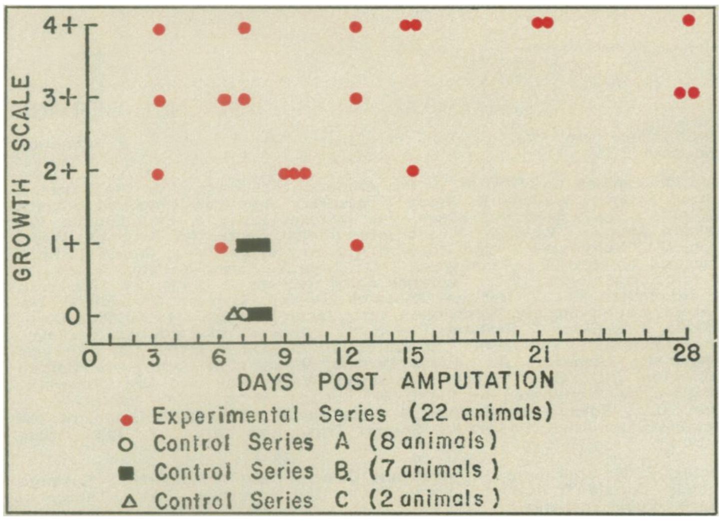
How cells and tissues actually regroup themselves at the amputated site, however, has not yet been determined. Becker says that electrical current applied to cells at an amputated site might cause them to revert to a primitive, undifferentiated—that is, unspecialized—state, somewhat like embryo cells. Then somehow the undifferentiated cells would become specialized again and start making more cells of their type—muscle, nerve or bone. Instructions



Becker: Understanding tissue repair.

for resuming specialization and replication, Becker hypothesizes, might be provided by neighboring cells or by the central nervous system. Hormones too might assist in redifferentiation and replication. Becker doesn't rule out the possibility of eventual total regeneration of human limbs, but he believes it will be a long time coming. His more immediate aim is better to understand cell and tissue repair in general and to stimulate this repair. If various kinds of tissue could be induced to grow just a little instead of scarring, there could be widespread clinical applications. For example, the myocardium of damaged hearts might be regrown, possibly proving more suitable than prosthetic heart parts, artificial hearts and heart transplants. Similarly, damaged hip joints might be removed from patients, the damaged joint tissue regenerated and the joint then reimplanted. Becker plans to try this procedure in dogs.

Becker's work is with experimental animals, but a clinical application of electricity to human patients was reported last week by Carl Brighton, Z. B. Friedenberg and their team at the University of Pennsylvania. In a patient with a fracture that had failed to heal for two years they caused healing by applying electrical current to the fracture site. The cathode, or negative pole of the circuit, was implanted at the point of fracture, and the anode was taped to the skin nearby. A small cast was placed around the limb and the power source, a small battery pack, was fastened to the cast. It delivered a constant 10-microampere current to the fracture 24 hours a day for eight or nine weeks, until X-rays showed that the break was healed. The technique is



Experimental rats treated with electrical current showed considerable amounts of limb regrowth; control rats did not.

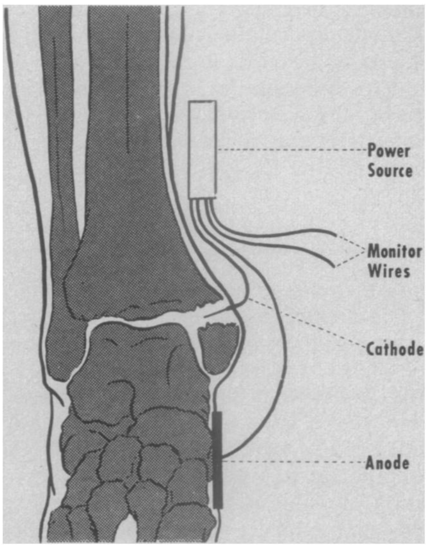
R. O. Becker

now being tried on two more patients whose fractures were stubborn about healing. The Pennsylvania orthopedic surgeons say the outlook is hopeful.

In all three cases, though, the fractures were without gaps in the bone. When current was applied to several patients with fracture spaces, healing was not stimulated. The investigators are now trying to see how they might get electric current to heal this kind of fracture. They also plan to undertake limited clinical trials, with double blind controls, to determine what electrical current does to fresh fractures.

Becker knows the Philadelphia investigators and respects their research. But he believes the clinical use of electrical stimulation in humans is premature.

"Our work [with rats] would seem to indicate that small electric currents are potent stimulators of cell activity," he told SCIENCE NEWS. "Neither our group nor any other to my knowledge has evaluated the possibility of inducing a malignant change in cells by means of electric current. That many cancer cells



Journal of Trauma

Philadelphia group's fracture therapy.

possess negative electric charges shows this possibility isn't farfetched. Until tissue culture studies and animal experiments show that electricity does not make cells cancerous, I would advise against applying electricity to human fractures."

In remarks prepared for the New York Academy symposium, he and colleague Joseph A. Spadaro reiterated this cautionary proviso, calling such applications to humans "unwarrented at this time."

Whether application in the clinical setting is premature or not, there seems little doubt that the work of both groups will offer valuable insights into the role of electricity not just in bone growth and fracture healing but in all kinds of tissue repair. □

How protein-like compounds could evolve in space

The hydrocarbon compounds on which life depends are too complex to form directly from their constituent atoms. They have to be the result of a series of steps of chemical combination. The nature of this series of events is an important question for scientists who are attempting to find out how life originated.

In 1953 Stanley Miller showed that amino acids could be formed from a mixture of inorganic substances that represented the supposed atmosphere of the primitive earth. But it is a long chemical way from amino acids to proteins. At the Southeastern Regional Meeting of the American Chemical Society in Nashville last week, Duane L. Rohlfing and Mary A. Saunders of the University of South Carolina at Columbia reported some laboratory work on intermediate steps in protein evolution. Among the conclusions it leads to is that protein-like compounds may exist outside the earth.

The next chemical possibility beyond the existence of simple amino acids are polymers called polyamino acids. Terrestrial proteins are polymers in various combinations of 20 amino acids called proteinous. In addition there are many amino acids called nonproteinous, which do not appear in protein compounds of life as we know it, but which can form part of compounds called protein-like. Amino acids of the nonproteinous variety have been found in samples from meteorites and in the results of the primitive-earth experiments.

The problem that Rohlfing and Saunders wanted to solve was whether these nonproteinous amino acids could coexist in the early stages of the evolution process. Could amino acids of both kinds form part of the same polyamino acid and were the proportions of amino acids found in the early experiments correct for the formation of polyamino acids? Rohlfing and Saunders took a number of polyamino acids formed by heating amino-acid mixtures and attempted to incorporate into them several nonproteinous amino acids alone and in combination with proteinous amino acids.

They found that all the nonproteinous amino acids they used could be incorporated into the polymers, though some went in more easily than others. They found important chemical similarities between polymers with and without nonproteinous amino acids. Says Saunders: "Those polymers containing only nonproteinous amino acids were composed of the same type of bond as those containing only proteinous amino acids, and the molecular weights were in the same range."

These chemical similarities indicate that proteinous and nonproteinous amino acids were linked together in the polymerization stage of the evolution of proteins on the primitive earth. "We conclude that the polymerization stage probably was not selective for proteinous amino acids," says Saunders. Some later process may have been responsible for sorting the proteinous amino acids from the nonproteinous ones. The basis of the separation may have been a difference in solubility, enzyme specificity, genetic selection or something else, but none of these possibilities yet has experimental support, she says.

When they used nonproteinous amino acids in the exact proportions reported by the earlier experiments that simulated primitive atmospheres the South Carolina investigators found that they did yield polyamino acids. "That these proportions yielded polyamino acids supports the concept of an evolutionary continuum from 'primitive-earth' gases to amino acids to polyamino acids," says Saunders.

Another result showed that the exact proportions of amino acids (including nonproteinous ones found in meteorite samples will polymerize thermally to yield protein-like compounds. "Because the moon and meteorites probably have a thermal history, our results are consistent with the possibility that protein-like compounds exist extraterrestrially." □

Cannikin's 7.0 explosion causes local damage only

In the wake of the furor over the Cannikin nuclear test on the Aleutian island of Amchitka, the explosion itself came as something of an anticlimax. None of the feared disasters materialized, and the Atomic Energy Commission was calling the test a complete success.

The five-megaton blast, which registered 7.0 on the Richter scale, produced many small aftershocks in the immediate vicinity of ground zero, but failed to set off a major earthquake or tsunami as feared by some environmentalists. Several large rockslides damaged the cliffs on the Bering Sea side of the island, and a few birds, sea otters, seals and fish were killed. No radiation leakage has been detected. The 800-foot-diameter underground cavity created by the blast collapsed Monday leaving a depression on the surface.

On the other hand, preliminary results indicate that the Spartan missile warhead being tested has met all specifications, and the AEC says further tests will probably be unnecessary. Indications are that Amchitka will be abandoned as a nuclear testing site. □