of live, anesthetized Harlan–Sprague-Dawley rats. Images were collected in less than 30 s and revealed excellent delineation of all major vessels [see figures 3 and 4 of (*1*)]. Although the report is encouraging, it raises questions for future studies. First, can smaller ^{3}He bubbles be generated while significant polarization is maintained? Also, concerns about embolism lead to safety questions about microbubbles of such a poorly soluble gas in the body. Finally, the researchers injected large amounts of the fluid into the rat vasculature (about 25% of the animal's body weight), making the current approach impractical in the clinical setting. The authors believe that the bubble concentration and the degree of ^{3}He polarization can be increased, so that in the future less material can be used.

Despite these significant limitations, the approach offers hope that very high-resolution angiographic images with little background signal may be safely made in the future. In addition to revealing blood vessels, ^{3}He microbubbles could also be used to study myocardial function without the risks of nuclear medicine approaches. Also, the ^{3}He microbubbles are short-lived so they would not generate a significant signal from a second-pass effect as the dye recirculates.

—RICHARD PETERS AND ROBERT SIKORSKI

Reference
1. M. S. Chawla *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **95**, 10832 (1998).

TECHSIGHTING
ELECTROSTIMULATION
Mailbox: www.sciencemag.org/cgi/dmail?53882b

Muscular Electronics

A number of techniques are under investigation for the effective transfer of DNA into tissue. In the field of gene therapy, one interesting technique that does not involve the use of viral vectors is injection of plasmid DNA directly into skeletal muscles. This approach is theoretically attractive for several reasons. It is simple; the intramuscular injection is like a vaccination. It does not involve viruses, it seems to have no adverse effects, and many delivery sites are available in the muscular system. Last, the transferred plasmid produces a protein that is secreted by the muscle systemically. This approach has been used successfully in animal models for the production of growth factors and cytokines such as erythropoietin and interleukin-5 (IL-5). Typically, the muscle is treated several days before the injection with a toxic compound (cardiotoxin or bupivacaine), because after an injection of plasmid, regenerating muscle tends to produce more protein than healthy muscle. The secreted proteins are produced for several months after one injection; however, the overall low level of expression of the transferred gene has precluded the use of this approach in human gene therapy.

A group of researchers in Japan have just published a modification of the intramuscular approach, attempting to improve the expression level by applying an electrical current around the intramuscular injection site (*1*). In this way, the technique of electroporation was applied to living cells with the expectation that it would increase uptake of plasmid DNA and result in greater expression of the transferred gene. During electroporation, cells are stimulated in vitro with electric discharges as the cells are bathed in a solution of plasmid DNA.

The authors used a plasmid that expresses IL-5 and injected it in the tibialis anterior muscles of 2-month-old mice. A pair of electrode needles with a 5-mm gap were inserted into the muscle to encompass the DNA injection sites. Three 50-V electric pulses of 50 ms each were delivered at a rate of one pulse per second, followed by three pulses of the opposite polarity. When the scientists measured IL-5 levels in the blood of the mice 5 days later, they found that mice injected with control plasmid lacking IL-5 had IL-5 levels undetectable by enzyme-linked immunosorbent assay (ELISA). Mice injected with IL-5 plasmid had serum concentrations of 0.1 ng/ml, whereas mice that received electric pulses showed concentrations >20 ng/ml.

Additional studies revealed that neither the orientation of the electrode needles (longitudinal versus transverse to the myofibers) nor pretreatment with muscular toxin affected the efficiency of electroporation. Instead, 3 weeks after the injection, concentrations of IL-5 were lower in mice that had been pretreated with bupivacaine, suggesting that the muscle necrosis induced by the toxin may reduce the long-term expression of transferred genes. A dose-response curve (voltage delivered versus expression of IL-5) indicated that the optimal voltage was 100 V. Higher voltage decreased expression of IL-5, possibly because of damage to the muscle cells. In addition, the 100-fold rise in IL-5 production resulted from increases in both the number of plasmids per myofiber and the number of muscle cells taking up the plasmid.

Although these results were encouraging, the procedure did not increase the overall duration of IL-5 production: time-course experiments revealed peak levels of IL-5 at days 5 to 7 and then a decay to 0.9 ng/ml 10 weeks later (*2*). Additional work will have to focus on optimization of other parameters such as pulse length, volume of injection, and solution type, to further improve the efficiency of gene transfer. It will be interesting to see this procedure used in larger animals in follow-up experiments.

—RICHARD PETERS AND ROBERT SIKORSKI

References
1. H. Aihara and J. Miyazaki, *Nature Biotechnol.* **16**, 867 (1998).
2. J. Miyazaki, personal communication.

TECHSIGHTING
DNA ANALYSIS
Mailbox: www.sciencemag.org/cgi/dmail?53882c

Pocket Laboratory

The use of simple laboratory kits for everything from running the polymerase chain reaction to plasmid cloning has revolutionized the analysis of DNA. Now, a group from the University of Michigan, led by David Burke, has developed an apparatus that might put an entire molecular biology laboratory into one device small enough to fit into a pocket (*1*).

How does it work? The tiny device (47 mm by 5 mm by 1 mm) was built by photolithography and microfabrication from a variety of materials, including silicon, platinum, and glass. The investigators designed a microchannel architecture in which reagents could be processed completely without human intervention. One key to the operation is a clever means of creating and moving the droplets that carry reagents, buffers, and DNA samples. The liquid flow pauses at various places because of hydrophobic patches in the channel. A small puff of air in perpendicular channels serves to separate nanoliter-sized droplets, which are then fed into the reaction chamber. From there, the tiny drops are passed to a matrix of polyacrylamide with embedded electrodes. Electrophoresis is performed in situ, and fluorescence detectors are used to measure the flow of DNA fragments.

Although hurdles exist, such as preventing evaporation and fine-tuning the electronics and fabrication techniques, the proof of concept for a pocket molecular biology laboratory has been demonstrated convincingly.

—ROBERT SIKORSKI AND RICHARD PETERS

Reference
1. M. A. Burns *et al.*, *Science* **282**, 484 (1998).