THE HEALING POWER WITHIN

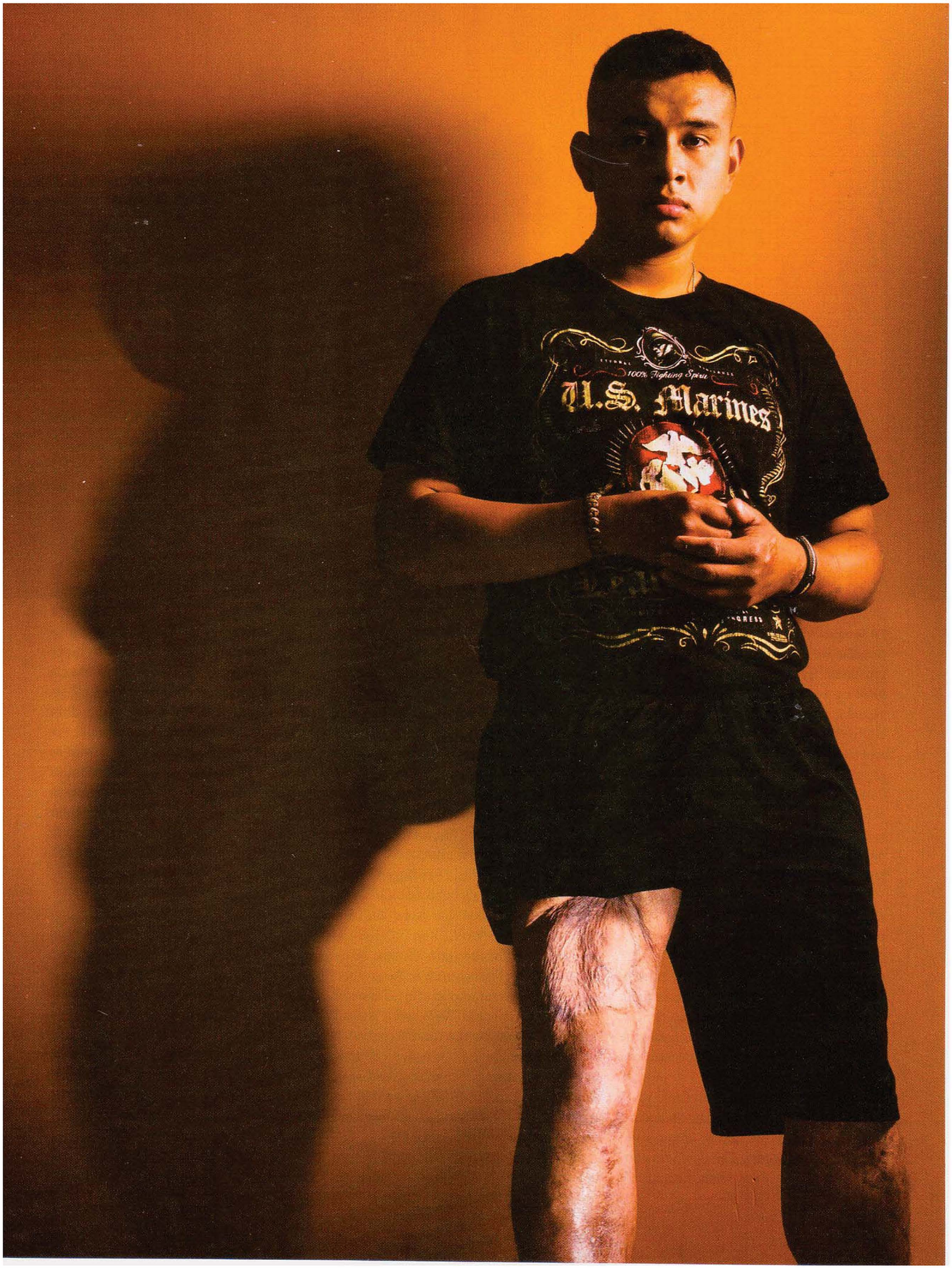
A remarkable substance enables the body to regenerate lost muscle tissue. Next up: Pioneer Stephen Badylak is working on treatments that would allow patients to regrow entire limbs.

By Adam Piore

Photography by Scott Lewis

THE STRANGE SENSATION IN HIS RIGHT THIGH MUSCLE BEGAN AS A FAINT PULSE. SLOWLY, SURELY, IT WAS BECOMing more pronounced. Some people would have thought it impossible. But Corporal Isaias Hernandez could feel his quadriceps getting stronger. The muscle was growing back. § When he first arrived in the trauma unit of San Antonio's Brooke Army Medical Center in December 2004, Hernandez's leg looked to him like something from KFC. "You know, like when you take a bite out of the drumstick down to the bone?" Hernandez recalls. The 19-year-old marine, deployed in Iraq, had been trying to outfit his convoy truck with a makeshift entertainment system for a long road trip when the bomb exploded. The 12-inch TV he was clutching to his chest shielded his vital organs; his buddy carrying the DVDs wasn't so lucky. § The doctors kept telling Hernandez

Corporal Isaias Hernandez shows off his healing thigh muscle.


100% Fighting Spirit
U.S. Marines

he would be better off with an amputation. He would have more mobility with a prosthetic, less pain. When he refused, they took a piece of muscle from his back and sewed it into the hole in his thigh. He did all he could to make it work. He grunted and sweated his way through the agony of physical therapy with the same red-faced determination that got him through boot camp. He even sneaked out to the stairwell, something they said his body couldn't handle, and dragged himself up the steps until his leg seized up and he collapsed.

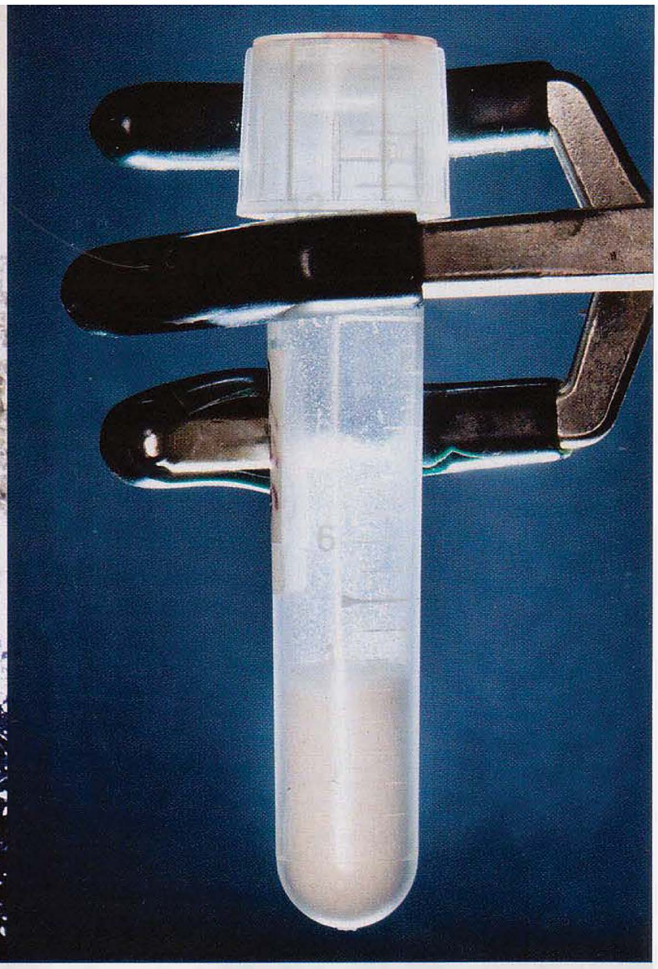
Generally people never recovered from wounds like his. Flying debris had ripped off nearly 70 percent of Hernandez's right thigh muscle, and he had lost half his leg strength. Remove enough of any muscle and you might as well lose the whole limb, the chances of regeneration are so remote. The body kicks into survival mode, pastes the wound over with scar tissue, and leaves you to limp along for life.

For Hernandez, it had been three years and there was no mistaking it: He had hit a plateau. Lately the talk of amputation had cropped up again. The pain was constant, and he was losing hope. Then his life took another radical turn. He saw a science documentary on the Discovery Channel (no relation to this magazine) that told the story of a war veteran in Cincinnati named Lee Spievack whose fingertip had been severed by the propeller of a model airplane. Spievack's brother, a surgeon in Boston, had sent him a vial of magic powder-the narrator called it "pixie dust"-and told him to sprinkle it onto the wound. Lee was to cover his hand with a plastic bag and reapply the powder every other day until his supply ran out. After four months, Lee's fingertip had regenerated itself, nail, bone, and all.

Hernandez recalled that one of his own doctors-Steven Wolf, then chief clinical researcher for the United States Army Institute of Surgical Research in Texas-had once mentioned some kind of experimental treatment that could "fertilize" a wound and help it heal. At the time, Hernandez had dismissed the therapy as too extreme. The muscle transplant sounded safer, easier. Now he changed his mind. He wanted his leg back, even if it meant signing himself up as a guinea pig for the U.S. Army.

So Hernandez tracked down Wolf, and in February 2008 the two got started. First, Wolf put Hernandez through another grueling course of physical therapy to make sure he had indeed pushed any new muscle growth to the limit. Then he cut open Hernandez's thigh and inserted a paper-thin slice of the same material used to make the pixie dust: part of a pig's bladder known as the extracellular matrix, or ECM, a fibrous substance that occupies the spaces between cells. Once thought to be a simple cellular shock absorber, ECM is now understood to contain powerful proteins that can reawaken the body's latent ability to regenerate tissue.

A few months after the surgery healed, Wolf assigned the young soldier another course of punishing physical therapy. Soon something remarkable began to happen. Muscle that most scientists would describe as gone forever began to grow back. Hernandez's muscle strength increased by 30 percent from what it was before the surgery, and then by 40 percent. It hit So percent after six months. Today it is at 103 percent-as strong as his other leg. Hernandez can do things that were impossible before, like ease gently into a chair instead of dropping into it, or kneel down, ride a bike, and climb stairs without collapsing, all without pain.



LIFT Biological scaffolds made of extracellular matrix, or ECM; the cylinder at far left mimics the shape of the trachea. er T"ii A mound of ECM powder, or "pixie dust," which has been shown to regenerate lost fingertips. ABOVC Pixie dust in a test tube in Badylak's lab.

The challenge now is replicating Hernandez's success in other patients. The U.S. Department of Defense, which received a congressional windfall of $80 million to research regenerative medicine in 2008, is funding a team of scientists based at the University of Pittsburgh's McGowan Institute for Regenerative Medicine to oversee an So-patient study ofECM at five institutions. The scientists will attempt to use the material to regenerate the muscle of patients who have lost at least 40 percent of a particular muscle group, an amount so devastating to limb function that it often leads doctors to perform an amputation.

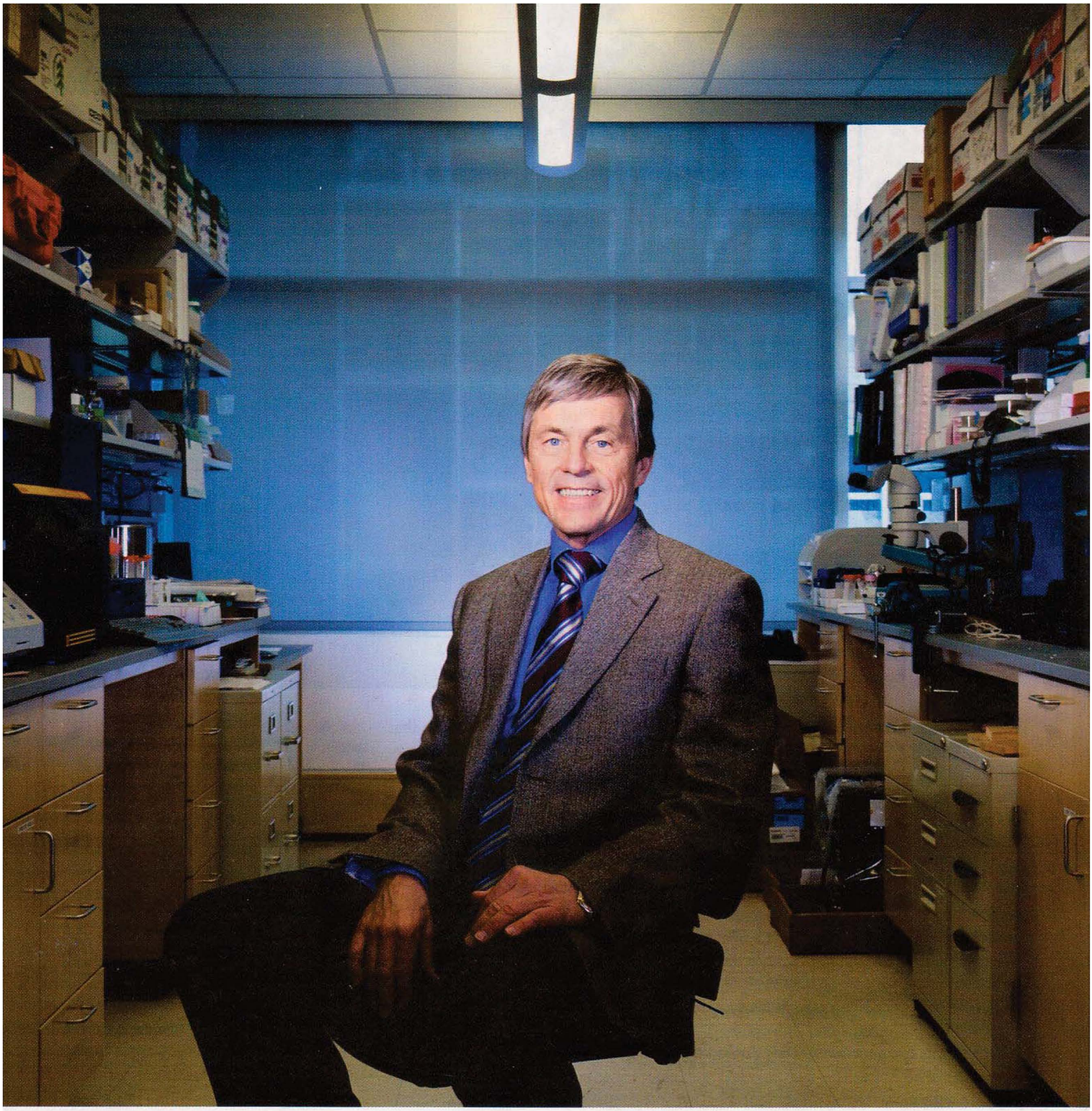
If the trials are successful, they could fundamentally change the way we treat patients with catastrophic limb injuries. Indeed, the treatment might someday allow patients to regrow missing or mangled body parts. With an estimated 17 million people in the United States alone missing limbs, promoters of regenerative medicine eagerly await the d$_{a\,y}$ when therapies like ECM work well enough to put the prosthetics industry out of business.

TO MANY MEDICAL PRACTITIONERS, THE IDEA of using pig parts to regenerate human tissue sounds outlandish-so outlandish that the doctor who discovered the technique in the mid-198os was reluctant to talk to clinicians about it for years. "They didn't believe my results;· says Stephen Badylak, a trim, extroverted researcher who is deputy director of the McGowan Institute and head of the So-patient muscle study. "Most people didn't believe it'.'

Badylak seemed to be saying that he could replace human tissue with tissue from another species without triggering a virulent immune response-something that medical scientists considered impossible. Even harder to swallow was the claim that the material could transform, in a matter of months, into whatever type of body tissue had been damaged-muscle, skin, or blood vessel.

When Badylak first published his findings, in 1989, the field of regenerative medicine was nonexistent. Badylak's debut paper on ECM went to press right around the time scientists first coined the term "tissue engineering" to describe what was then considered a small but burgeoning field-the far-out-there efforts to coax cells into tissue to restore, maintain, or improve tissue function or whole organs. Today, the most widely publicized efforts in the field concentrate on growing tissue outside the body in specially designed, easily controllable "bioreactors." Badylak's ECM techniques, however, stimulate the body's own army of stem cells to do the healing, no external equipment needed.

Badylak is still testing the clinical limits of ECM. Last February he and collaborators announced that they had regenerated one of the body's most scar-prone tissues, the inner lining of the esophagus, in five cancer patients. Esophagus tissue is so sensitive that even minor surgical manipulations often result in a thick buildup of strictures that make it impossible to swallow. As a result, most surgeons wait as long as possible to operate on an esophageal tumor, then remove the entire organ using a procedure that has an extremely high complication rate. Badylak was able both to suppress all scarring in his patients and to prompt the



Stephen Badylak, regenerative-medicine pioneer, in his lab in Pittsburgh. "We are changing the body's default mechanism of healing," he says.

fragile lining of the esophagus to regenerate completely. He is now awaiting FDA clearance to begin a large-scale clinical trial.

Ultimately, Badylak believes that ECM will lead to therapies that regrow amputated human arms and legs, much as salamanders and starfish regenerate limbs, although he realizes that this may not happen in his lifetime. Regrowing an entire finger is a far greater challenge than regrowing a single tissue like muscle. Badylak's strategy at the moment is to construct a dome that would cover the end of an amputated body part and re-create the conditions that exist in a human embryo, which possesses the ability to grow any tissue type. "We know that in a test tube we can get ECM to form muscle, tissue, fat, and bone:· he says. "If we can create optimal conditions, we can truly program the formation of the functional tissue

THE DISCOVERY THAT LED TO THIS RADICAL approach in wound healing happened quite by accident. It all ·started with what Badylak's associates called a "harebrained" idea and a mutt named Rocky.

In 1987 Badylak was a new hire at Purdue University, working with a well-established biomedical engineer named Leslie Geddes. Badylak, a young Indiana native, brought an unusual background to his post. After college he had attended veterinary school at Purdue and practiced animal medicine until he realized that most pet owners could not afford the tests necessary to diagnose the conditions that fascinated him. Frustrated and worried that he would grow bored, he went back to Purdue to earn a Ph.D. in animal pathology. After weighing teaching offers, he decided to go to medical school. Badylak used his old connections to help pay his way, setting up a lab in his home to diagnose ferret lymphoma and dog breast cancer for former veterinary classmates who mailed him samples.

At Purdue, Badylak became fascinated by an experimental technique called cardiomyoplasty, in which a flap of a patient's back muscle is removed and wrapped around the patient's ailing heart. A pacemaker shocks the muscle into contractions and helps the heart squeeze blood through the body. When Badylak decided to investigate the technique on his own, it was only natural that he would gravitate back toward animal patients, this time as test subjects.

He quickly discovered a downside to cardiomyoplasty. It used synthetic tubing to replace the aortic artery, and this often triggered aggressive inflammation and blood clots. Badylak became convinced that if he could find a blood vessel substitute within a patient's own body, he could stop the inflammation. So one afternoon he sedated an affable dog named Rocky, removed part of the animal's aorta, and replaced it with a piece of its small intestine, the part of the body that most resembled the tubular structure of Rocky's blood vessels. Badylak did not expect Rocky to survive the night, but he figured that if the animal had not bled out by morning, it would prove the intestine was sturdy enough to pass blood and hence worthy of further study.

This was, Badylak would later admit, the kind of outside-the-box experiment that would probably never get past a university animal-care committee today. His third-year cardiovascular surgery resident called the operation "cruel" and "ridiculous" and refused to participate. Even Badylak's habit of referring to the dog by name was contentious, since researchers typically conform to the colder convention of identifying laboratory animals by numbers. But when Badylak arrived for work the morning after Rocky's surgery, he found the mutt wagging his tail and ready for breakfast.

Badylak could not find the transplanted intestine. That, he recalls, is when "things got really weird."

Badylak kept expecting the dog to die, yet every day he would find Rocky healthier and more energetic than the last. Days turned to weeks and Rocky continued to thrive. "I didn't want to go in surgically and look because I wanted to see how long the intestine would hold;' he says.

Hoping to make sense of his unexpected result, Badylak repeated the procedure on 14 other dogs. They, too, thrived. Six months later he finally operated on one of the dogs to understand why. That, he recalls, is when "things got really weird:' Badylak could not find the transplanted intestine.

After checking and double-checking to make sure he had the right animal, he placed a piece of tissue culled from the transplant target area under a microscope. What he saw floored him. "I was looking at something that wasn't supposed to happen;· Badylak says. "It went against everything I had been taught in medical school'.' Under the glass he could still see traces of the sutures, but the intestinal tissue was gone. The aorta had grown back in its place. "Nobody would confuse an intestine and an aorta:· Badylak says. "The microscopic picture is entirely different. I tried to get everybody I could think of to look at it. I kept asking, 'Am I seeing what I think I'm seeing?'" Intestine is composed of soft, smooth, thinly lined walls, with hairlike projections known as villi. Aorta is thick, with the meaty, striated layers of the tissue that characterizes muscle.

Badylak examined several other dogs in the weeks that followed and watched the intestinal tissue transform again and again. He began to suspect that something in the intestine was suppressing inflammation and simultaneously promoting regeneration. Thinking back, he recalled a bizarre finding on liver regeneration he had heard about in a veterinary school pathology lecture: If you eat poison and it destroys all the cells in your liver, the organ can still regenerate, but only if its structural scaffolding remains intact. Destroy the scaffolding and the body responds by producing massive scar tissue and no regeneration. Perhaps the scaffolding was the key.

The next step, then, was to strip away the layers of the intestine, including its mucosal and muscle strata, until he was finally left with a paper-thin sheet of connective tissue called the extracellular matrix-the magical ECM.

When he replaced the dog intestine with just this tissue, the transplant still worked. Badylak repeated the experiment, this time using ECM derived from cat intestine. He was sure the dog's immune system would reject the cat gut, but once again the transplant was successful. At this point Badylak realized he would be working with small intestines for a long time, and he was going to need lots of them. So for his next experiment, he used intestine obtained

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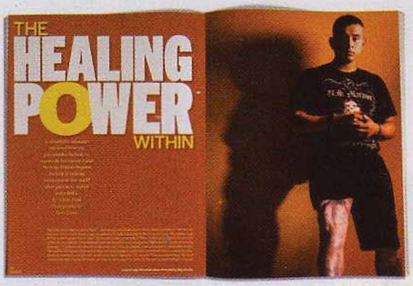

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from one of the many pig slaughterhouses dotting the Indiana countryside surrounding Purdue. There would be no shortage of material if it worked. He tried it and, sure enough, his test dog was up and waiting for breakfast the day after it received the first ofBadylak's pig intestine transplants. (Porcine entrails-not only intestines but bladders, which were found effective as well-have been a staple in the doctor's laboratory ever since.)

As for Rocky? He lived another eight years.

BADYLAK HAD SOLVED THE MYSTERIOUS "how" of Rocky's miraculous recovery. Now he faced a much larger enigma as he contemplated the "whf He relentlessly pursued answers in the lab; at the same time, he eagerly looked to expand the medical applications for ECM. If it healed, why not start using it right away? People took aspirin for 30 years before anyone understood how it worked, he reasoned.

So Badylak moved the focus of his experiments from the large aortic artery to large veins. The pig intestine worked there. Then he found the material worked on small arteries, too. Finally, in 1989, he conducted a more radical experiment, removing a chunk of a dog's Achilles tendon and replacing it with pig ECM. The normal response of any mammal's body to significant damage is to create scar tissue, a hasty but crude way of replacing what has been lost. Scar tissue has a clear evolutionary advantage: The body is quickly sealed off from bacterial infection, and the injured creature has a better chance of surviving. A cut to the Achilles tendon normally produces a stiff lump of scar tissue that causes the animal to limp. Badylak's dogs grew their entire tendons back. They developed no scars, and hence no limps.

In 1992 Purdue's patent lawyer mentioned Badylak's work to another client, an orthopedic device manufacturer called DePuy, based in nearby Warsaw. Like everyone else. executives at DePuy were initially skeptical. "It sounded like magic;' recalls Richard Tarr, who was then DePuy's vice president of research and development. "But I have learned in research that you never stop listening. You can always say no:·

Badylak delivered a detailed presentation on ECM to a team at DePuy and explained that he had created a three-centimeter gap in the hind-leg Achilles tendons of three dogs. Then he left and returned with three so-pound hunting dogs that bounded in and jumped up on their hind legs to greet the visitors. Tarr ran his fingers over one mutt's hard, newly regrown Achilles tendon. Three months later DePuy licensed Badylak's ECM-derived "biologic scaffolds" for all orthopedic applications. Suddenly Badylak had an industry sponsor to push for FDA approval, as well as $250,000 a year to continue his research.

It was around this time that Badylak first met Alan Spievack, a Boston-based surgeon who approached Badylak after he delivered a lecture on ECM at an orthopedic conference in Atlanta. As an undergrad at Kenyon College in Ohio in the 1950s, Spievack had performed amputations on salamanders and studied the way the creatures regenerated their limbs. He went on to a long, successful career as a surgeon. But Badylak's talk rekindled Spievack's fascination with tissue regeneration, and he persuaded the researcher to join him for a cup of coffee. Spievack visited Badylak's lab and soon after joined the growing number of researchers who had begun pursuing their own research on ECM.

Despite this flurry of independent investigations, the true mechanism ofECM's healing power was still unknown when Badylak sat down for a series of meetings in 1996 with representatives from DePuy and the FDA to discuss plans to begin initial testing of biologic scaffolds in humans. He worried that this missing piece of the puzzle would be a deal breaker. There was not much information to go on. ECM was known to be the glue that holds tissue together, a cellular-level skeleton upon which nerve, bone, and muscle can plant themselves and get to work. It is composed of some of the body's most enormous protein molecules-laminin, collagen, and fibronectin-woven together in an intricate, seemingly impregnable web to form a scaffold. Few scientists had ever suggested ECM was anything more than a dumb structural element.

To Badylak's surprise. the FDA investigators did not seem especially concerned about the mechanics of the scaffolds. DePuy had developed a patch composed of 10 layers of the material laminated together, which it intended to market for use in rotator cuff repair. And the company had come up with a strategy to win quick FDA approval. DePuy combed the industry for already approved therapies with similar properties - a n d found a soft-tissue reinforcement patch already used in hernia repairs made from bovine heart tissue. Then company scientists sought approval through a truncated process called a 510K, arguing that Badylak's pig scaffolds shared many of the earlier therapy's characteristics. Never mind that the bovine product had no regenerative properties; ifDePuy could win approval for pig bladder as a safe soft-tissue hernia repair method, doctors could legally use it off-label in other ways.

The FDA asked the routine questions used to evaluate medical devices: Does it cause cancer? Does it have an adverse inflammatory reaction? What techniques are you using to sterilize it? "They had a series of standard boxes they needed to check for medical devices;· Badylak recalls. 'i\nd when we told them the material actually breaks down and gets replaced, there was no box to check. It was one of the first things like this they had ever looked at:'

In 1999 the FDA approved the material for clinical use, and soon surgeons across the nation began using it on patients to repair rotator cuffs, abdominal hernias, and esophageal reflux damage, and even to induce the regrowth of the outer lining of the brain.

'11le next year, Badylak visited one of these surgeons in Los Angeles and had a revelation that would lead him to finally discover the true source ofECM's power. The surgeon, John ltamura, had implanted an ECM scaffold into the shoulder of a patient who returned eight weeks later in need of surgery for an unrelated problem. The coincidence allowed the doctor to obtain a rare human sample from the area of the shoulder operation. A biopsy showed that the scaffolding had disappeared, as expected. But there

was a surprise: Seen under the microscope, the surgery site was alive with activity. Disparate cells appeared to be swarming the area in a process that looked similar to an inflammatory response. However, these new arrivals were not blood cells, as you would expect, but something altogether different and unusual.

At first Badyl k was puzzled. He knew that the scaffolding could not be the source of the activity, because it had long since broken down. The cause, he realized, had to be the products left behind-molecules, perhaps, that had been lurking within the scaffolding waiting to be released.

Badylak combed the scientific literature for answers. He quickly discovered that components called cryptic peptides, or "crypteins:· would explain much of ECM's unique phenomena. Researchers in other fields had previously discovered that certain proteins give rise to these hidden peptides when they degrade, and that the peptides have potent antimicrobial effects and important signaling abilities. "Almost everybody considered the extracellular matrix just the structural support that allowed you to stand up and support weight and hold things together;· Badylak says. "But now we know it's almost just the opposite. It's primarily a collection of signaling proteins and information that is held within the structural molecules'.'

Badylak understood the recruitment process, but he still could not figure out what the crypteins were recruiting. He went back to the microscope and watched armies of cells converge on the site of the broken-down ECM. In their number and characteristics these new arrivals looked nothing like muscle, nerve, or blood cells. Badylak soon suspected that the recruits were stem cells, the all-purpose cells that can develop into any type of tissue.

He proved it in 2003 by first X-raying mice to kill off all the stem cells in their bone marrow, then repopulating the bone with stem cells fagged with a fluorescent marker. When he removed a piece of mouse Achilles tendon and added ECM, fluorescent stem cells flooded into the area. Months later, some of these tagged cells were still present-implying that some of them ad matured into regenerated tissue.

Badylak's published results caused a stir in the fast-growing field of regenerative medicine, and his professional reputation flourished. To the outside world, however, the researcher remained largely unknown until 2007, when an odd confluence of events involving his old friend and collaborator, Alan Spievack, and Spievack's injured brother catapulted him into the public eye. Spievack, who had coauthored several papers with Badylak, eventually went on to found a company called ACell to market his own special formula of the powder.

That is how Spievack, by then 73, was in a position to heal his younger brother, Lee. When news got out that Lee had regenerated his fingertip with a mysterious powder he called pixie dust-and graphic pictures displaying the regenerative process landed on editors' desks-a media frenzy erupted. The stories and the photos sparked the imagination of amputation victims around the world, including Corporal Isaias Hernandez.

Four years later, Badylak still gets several emails a day asking about his miraculous pixie dust. Spievack did not get to share in much of the glory; he died of cancer in May 2008.

NOW THAT BADYLAI<'s REGENERATIVE WORK has finally gone thoroughly mainstream, he is once again seeking to push the outer limits of healing-and is back to square one seeking grants for his far-out research.

Badylak, along with Tufts University biomedical researcher David Kaplan and Susan Braunhut of the University of Massachusetts Lowell, is using a device called a bio-dome, a sleeve with a liquid reservoir that envelops an amputated mouse digit and allows researchers to control the healing environment. What he is trying to do, in a sense, is make us born again. By adding growth factors, liquids such as water and amniotic fluid, and varying electric currents, he and his colleagues are replicating the conditions that exist in a human embryo-an environment that is perfectly conducive to the transformation of stem cells into the complex tissues that make up a body.

The idea of replicating an embryo on the end of a mammal limb to regrow it is considered too unconventional by most peer reviewers. The project is still without funding. But Badylak is undeterred. After all, he never let skepticism stop him before. D