the battle

against aging

EVERETT COLLECTION

THERE MAY BE A WAY TO PREVENT OURSELVES FROM RUSTING FROM THE INSIDE OUT

a radical proposal

BY KATHRYN BROWN

WIZARD OF O_2: Water killed the wicked witch in Oz, but oxygen may kill us, oxidizing our cells the way it rusted Dorothy's pal the Tin Man.

You can drop cigarettes. Avoid alcohol. But there's one toxin you just can't dodge: oxygen. With every gulp of air, oxygen gives you life. Some of it, however, gets converted inside your cells into a radical molecule that can wreak havoc, degrading those same cells and others. A growing number of scientists say this damage is what causes aging. They also think they may one day be able to fend off oxygen's ill effects and help us live a lot longer.

Scientists have long known that oxygen is capricious. As molecules go, it gets around, reacting with all kinds of things. Mostly, that's good. Oxygen combines with fats and carbohydrates, in a part of cells known as the mitochondrion, to churn out the energy that gets you through the day. But the conversion isn't perfect. A small amount of oxygen is regenerated in a nasty form called a free radical, or oxidant—the very critter that causes metal to rust. The oxidants careen about, binding to and disrupting the membranes, proteins, DNA and other cell structures that make your body work. Over time, this damage adds up, and the result just might be an older, frailer you.

According to one estimate, oxidants bombard the DNA inside every one of our cells roughly 10,000 times a day. Thankfully, most of the assailants are intercepted by a small army of antioxidant chemicals. Proteins also patch up the damage caused by the radicals that do get through. "The house is always getting dirty, and we're always trying to clean it up," remarks John Carney, chief technical officer at Centaur Pharmaceuticals in Sunnyvale, Calif., which is developing drugs to fight various diseases of aging. But eventually, the theory goes, our tired cells get less efficient at repelling free radicals and mopping up oxidative messes, and the damage accumulates. We begin to rust from the inside out.

If oxidants do send us crumbling into old age, then ramping up our biochemical defenses should extend life. That's what scientists are finding, at least in the flies, rats, worms and other animals they have under scrutiny in the laboratory. Whether the techniques they are pursuing will ever lengthen life in humans remains an open question. But some researchers think they're getting close to an answer. "The key is to really understand how oxidative damage works, and we're learning that," says biochemist Bruce N. Ames of the University of California at Berkeley. "I'm convinced life expectancy will get longer a lot faster than anybody thinks."

The Original Pollutant

Oxygen's checkered past goes way back—about two billion years. Around that time, scientists believe, cyanobacteria began releasing more and more oxygen into the earth's atmosphere, until many organisms were forced to either accommodate the gas or risk being degraded by its corrosive nature. Over time, some particularly oxygen-adept bacteria evolved into mitochondria, the tiny powerhouses in all human cells that use oxygen to help turn food into energy.

The "free radical theory of aging" was first laid out

about 45 years ago by Denham Harman of the University of Nebraska. The idea won credibility in 1969, when scientists identified a key antioxidant, superoxide dismutase (SOD), an enzyme that breaks down the harmful superoxide, a leader among the various free radicals that can form inside the human body. Soon researchers began to realize that mitochondria created oxidants in high amounts. And by now dozens of experiments have linked oxidative damage and aging.

Until recently, however, that link had been a matter of indirect correlation. In the lab, for instance, some young human cells do far better than older cells at resisting or repairing oxidative damage, whether the cells are being doused with hydrogen peroxide or stuck inside a chamber filled with pure oxygen. Also, lab flies, worms and mice carrying genetic mutations that proffer long life tend to withstand oxidative assaults better than their peers. "All these studies suggest oxidative damage may be an important part of aging, but they lack the kind of direct experiments to nail that link down," notes John Tower, a molecular biologist at the University of Southern California. "The question is, if we actually alter oxidative stress, will it extend life?"

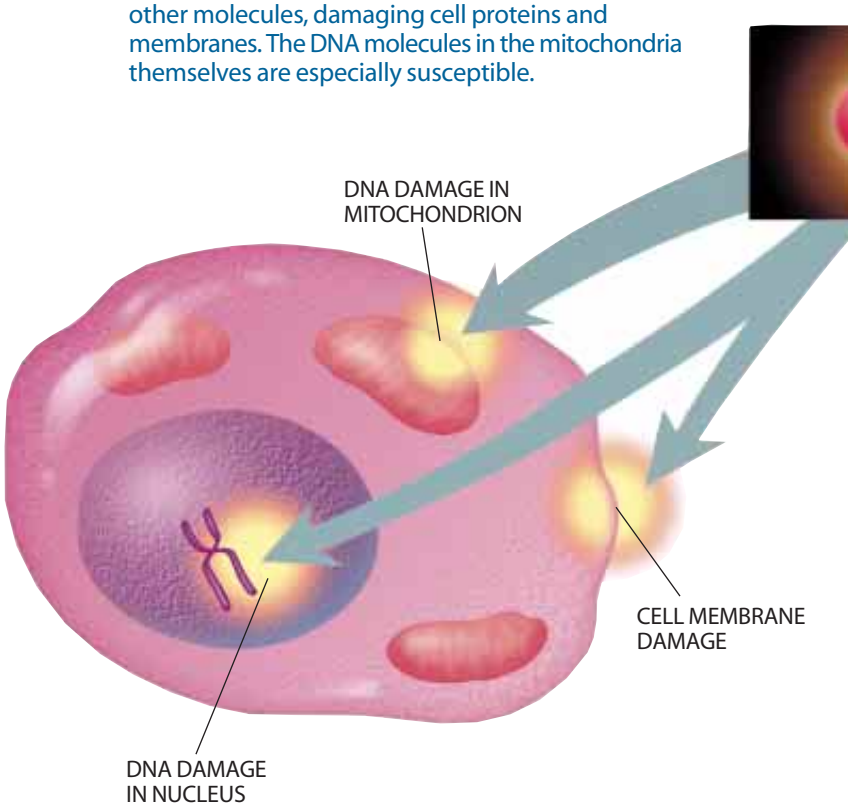
To find out, Tower and his U.S.C. colleague Jingtao Sun recently reared fruit flies with an engineered protein that could—when exposed to heat—turn up the activity of SOD and another antioxidant, catalase. The flies started life in the lab normally, along with a control group of flies. Then, on the fifth day, the experimental flies got pulses of heat, ratcheting up their antioxidant defenses. The results were striking. Most of the everyday flies keeled over long before six weeks—but those with supercharged SOD, in particular, survived an average of 48 percent longer. "That's pretty convincing evidence that overexpression of SOD extends life," Tower says.

That's not the only evidence. Five years ago William Orr and Rajindar Sohal of Southern Methodist University in Dallas equipped their own flies with extra copies of genes for SOD and catalase. Those flies lingered up to a third longer than their normal maximum life span—and seemed to age more slowly along

cellular damage and defense

In cell metabolism, the mitochondria convert glucose and oxygen into energy. Oxygen radicals also form as a normal by-product.

The highly reactive oxygen radical can bind with other molecules, damaging cell proteins and membranes. The DNA molecules in the mitochondria themselves are especially susceptible.



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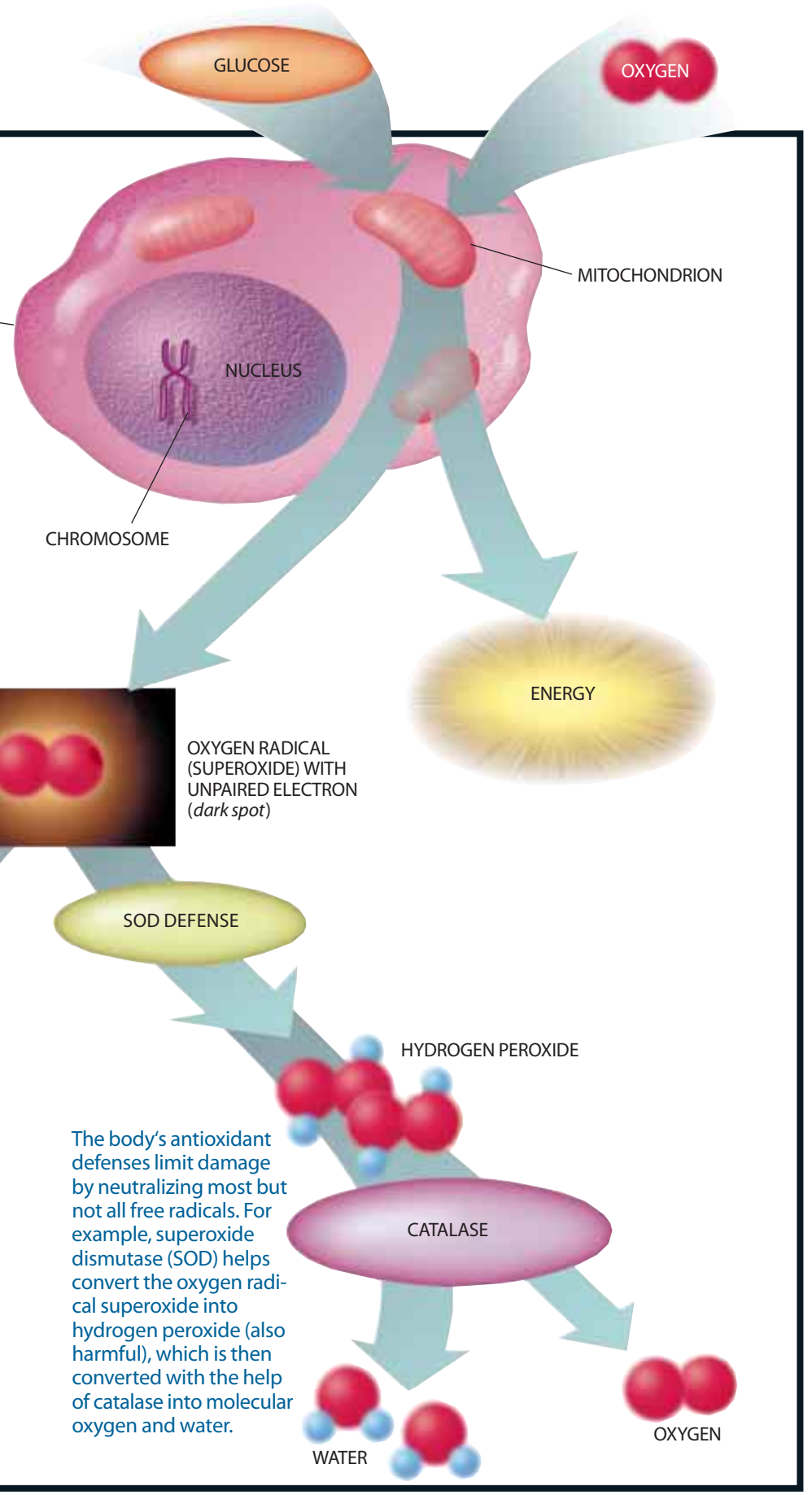


The body's antioxidant defenses limit damage by neutralizing most but not all free radicals. For example, superoxide dismutase (SOD) helps convert the oxygen radical superoxide into hydrogen peroxide (also harmful), which is then converted with the help of catalase into molecular oxygen and water.

the way, exhibiting higher energy, faster movements and less oxidative damage. Eventually, Sohal says, similar studies will be done with mammals and then, if deemed safe and efficient, with humans.

Intercepting the Interloper

In the meantime, scientists hope to pinpoint exactly where oxidants do their dirtiest work—and ways to intervene. The idea, says molecular biologist John Phillips of the University of Guelph in Ontario, is to tailor therapies to the most important injured cells, rather than trying to fight oxidative damage throughout the body. Phillips has one candidate cell in mind: the motor neuron, which directs muscles from the brain and spinal cord. People with a paralyzing disease called familial amyotrophic lateral sclerosis die early, with heavily damaged motor neurons as well as mutations in SOD. Maybe motor neurons are a critical target of oxidants, kick-starting or dominating the process of aging.

To test that idea, Phillips and his coworkers bred fruit flies with a jolt of one of the human superoxide dismutase compounds, SOD1, to be expressed only in the flies' motor neurons. Sure enough, the bugs lived 40 percent longer than normal. And those extra days were lively ones. "We didn't just delay dying, so that we had geriatric flies living longer," Phillips says. "The extended time of life was youth." In contrast, boosting SOD1 levels in unrelated muscle cells seems to have had no effect on the flies' life span, he adds. Still, questions remain. "We don't really know why these animals are living longer," Phillips concedes. To pin down SOD's relevance, the team is now spiking different types of neurons with the antioxidant to see how the various cells react.

Another target for protection is the mitochondria inside all cells. Because these tiny powerhouses are the very source of harmful oxidants, they're the first cell structures to be clobbered by the chemicals. In a 1998 study Sohal and his co-worker Liang-Jun Yan exposed flies to high doses of pure oxygen and then went looking for signs of oxidants at work in the flies' mitochondrial membranes. Rather than far-flung havoc, they

found that oxidants targeted several vulnerable proteins, attaching to their strings of DNA, forcing them out of work and upsetting the entire cell's ability to act normally. "Free radical damage during aging is not random, causing decline all around our cells," Sohal says. "We're talking about damage that's very selective, and that may mean aging comes from specific biochemical losses."

Proof of this notion would be good news, Ames says. "The key thing is to understand how aging really works. If it's the decay of mitochondrial DNA, well, we can do things to beef up these old mitochondria."

Ames, Tory Hagen of Oregon State University and their colleagues have done just that. In preliminary work, they found that the liver cells of older rats do not fend off free radicals as well as the liver cells of younger rats do. So last year, over a two-week period, they fed a group of older rats food laced with lipoic acid, a chemical that the mitochondria can convert into a potent antioxidant. After this high-powered diet, the older rats' liver cells deflected oxidant intruders with greater resilience. What's more, the senior rats scrambled around with new spirit and a sleeker look. "I don't want to say we've gone so far as turning old rats back into young rats," Ames says, "but that sure looks like what's going on in the mitochondria." The team has just begun a study to see whether the antioxidant-endowed rats actually outlive their lab mates.

Supermarket Solutions

If antioxidants work for flies and rats, what about us? Can you down a daily supplement that will extend your years? Don't count on it. "Everybody is talking about popping antioxidant vitamins," Phillips groans. "The evidence is strong that taking moderate amounts of vitamin C and E is not harmful, but the evidence that it's actually useful for delaying aging is very thin." For one thing, researchers say, your body can absorb only so much of these vitamins; the rest goes the way of other wastes. Also, in the industrial world, most of us get enough of the basic antioxidants in our daily diets. In contrast, lab animals that live unusually long with extra antioxidants may be deficient in those chemicals to begin with.

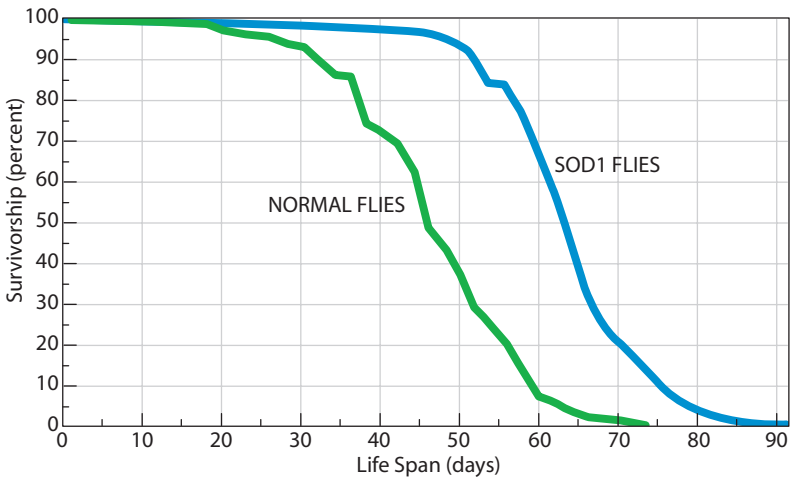
Even if antioxidant supplements do boost your defenses against free radicals, it's tricky to know which ones—or how much—to take. As with any ingredient, too much can be a bad thing. In 1996, for instance, two large studies made news when researchers discovered that beta-carotene supplements—thought to help ward off some types of cancer—actually increased rates of lung cancer among smokers who were taking the pills. Some antioxidants hawked in health food stores will never do any good; walk right past those bottles of SOD, catalase and glutathione peroxidase, because these compounds must be created inside the body. When swallowed, they are simply broken down in digestion and rendered useless, researchers state.

Still, there are some antioxidants that hold promise, Ames says, such as lipoic acid, which directly protects the mitochondria. Perhaps, he adds, some of the more obscure antioxidants dry up in the body as we age, leaving us more vulnerable to oxidative damage. If that's the case, downing extra amounts of these conditional nutrients might slow aging's cellular effects. "We just don't know yet," Ames says.

Indeed, there are a lot of unknowns. What proportion of aging changes in cells are the result of oxidative damage? Is there a way to reduce the rate of oxidants the body churns out, rather than simply boosting antioxidants? And what do all these long-lived lab mutants really explain about oxidative stress in people? Sohal worries that some of the most touted studies are misleading. For instance, biologists have won lots of attention by reporting that in worms, single mutations in a gene called *daf-2* can double life span, partly by resisting oxidative stress. But this is a "bogus kind of life extension," charges Sohal, because the worms' metabolism (energy level) plummets during their extra time on earth. "It's just like going to sleep for three years and calling those three extra years of life," he says. The extra time is akin to hibernation, Sohal adds, so any therapy based on it would rob people of the energy they normally have.

The most basic challenge is understanding aging itself. Growing old is a slow, subtle process that's hard to de-

rust inhibition: an enzyme extends life



LONGER YOUTH: Fruit flies bred with a dose of SOD1, an antioxidant enzyme that breaks down free radicals, lived 40 percent longer than normal fruit flies did in a University of Guelph laboratory. Notably, the phase of life extended was youth, not old age.

LAURIE GRACE; SOURCE: JOHN PHILLIPS *University of Guelph*

fine with blood tests or cellular studies. Oxidants can muddy the picture, observes Carney of Centaur Pharmaceuticals. After all, these omnipresent molecules can strike a cell's proteins, fats or DNA, all very different beasts. "Understanding oxidative damage and the biology of aging is a massive undertaking," he points out.

In the short run, Carney says, researchers may first unravel the role of oxidants in specific diseases of aging. Centaur, for instance, is working on drugs to fight Alzheimer's and Parkinson's diseases. People who suffer from these conditions show telltale signs of oxidative damage in the brain. Eventually these studies may inch scientists closer to understanding basic brain changes during aging. Carney has reason to be optimistic. Some 10 years ago, while at the University of Kentucky, he and his colleagues were the first to report that high levels of a synthetic antioxidant, PBN, can decrease harmful oxidative proteins in the brains of old gerbils. "Aging may indeed be a treatable process," Carney maintains.

Self-Imposed Treatment

Some individuals are prescribing their own treatments. According to one idea, you can starve yourself, cutting back on calories until your metabolism drops so low that fewer free radicals are formed in the first place. A more pleasant alternative, perhaps, is munching on fruits and vegetables that are high in antioxidants. Last year neuroscientist James A. Joseph of Tufts University and his colleagues reported that middle-aged rats fed extracts of spinach, blueberries or strawberries for eight weeks showed marked declines in oxidative stress in their brain cells, as well as improved memory and coordination. The most successful rats noshed on blueberries—the equivalent of a cup a day for humans.

The research also highlights how much scientists have to learn about the processes that contribute to aging. Apparently, it's the blend of ingredients inside blueberries—not just isolated antioxidants—that benefited the racy rats. Studying the rats' brain cells, Joseph was surprised to find relatively few signs of increased antioxidants. Instead he found a host of cell changes, from better anti-inflammatory activity to more pliable membranes—all of which could act together to combat aging changes.

"If you take a supplement, you never get the benefit of a fruit or vegetable that contains hundreds of compounds," Joseph says. Right now researchers can't even identify all the compounds, much less explain how they might work together to fight free radicals. The answers could be years in coming. In the meantime, he asks, why not stroll down the produce aisle? A few berries might just offset a little oxidation—or at least make the wait for answers to aging that much sweeter.

the antioxidant diet

Your best bet for fending off cellular damage from free radicals, scientists say, is to maintain a healthy supply of antioxidant compounds by eating fruits and vegetables—not by taking a pill. Here are some foods rich in antioxidants.

Fruits: blueberries, cherries, kiwis, pink grapefruit, oranges, plums, prunes, raisins, raspberries, red grapes, strawberries

Vegetables: alfalfa sprouts, beets, broccoli flowers, Brussels sprouts, corn, eggplant, kale, onions, red bell peppers, spinach

KEVIN R. MORRIS *Corbis;* SOURCE: U.S. AGRICULTURAL RESEARCH SERVICE

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Further Information

The Free Radical Theory of Aging Matures. Kenneth B. Beckman and Bruce N. Ames in *Physiological Reviews,* Vol. 78, pages 547–581; April 1998.

Extension of Drosophila Lifespan by Overexpression of Human SOD1 in Motor Neurons. Tony L. Parkes et al. in *Nature Genetics,* Vol. 19, No. 2, pages 171–174; June 1998.

Reversals of Age-Related Declines in Neuronal Signal Transduction, Cognitive, and Motor Behavioral Deficits with Blueberry, Spinach or Strawberry Dietary Supplementation. James A. Joseph et al. in *Journal of Neuroscience,* Vol. 19, No. 18, pages 8114–8121; September 15, 1999.