

The Serious Search for an Antiaging Pill

In government laboratories and elsewhere, scientists are seeking a drug able to prolong life and youthful vigor. Studies of caloric restriction are showing the way

By Mark A. Lane, Donald K. Ingram and George S. Roth

No treatment on the market has been proved to slow human aging—the buildup

of molecular and cellular damage that increases vulnerability to infirmity as we grow older. But one intervention, consumption of a low-calorie yet nutritionally balanced diet, works incredibly well in a broad range of animals, increasing longevity and prolonging good health. Those findings suggest that caloric restriction could delay aging in humans, too.

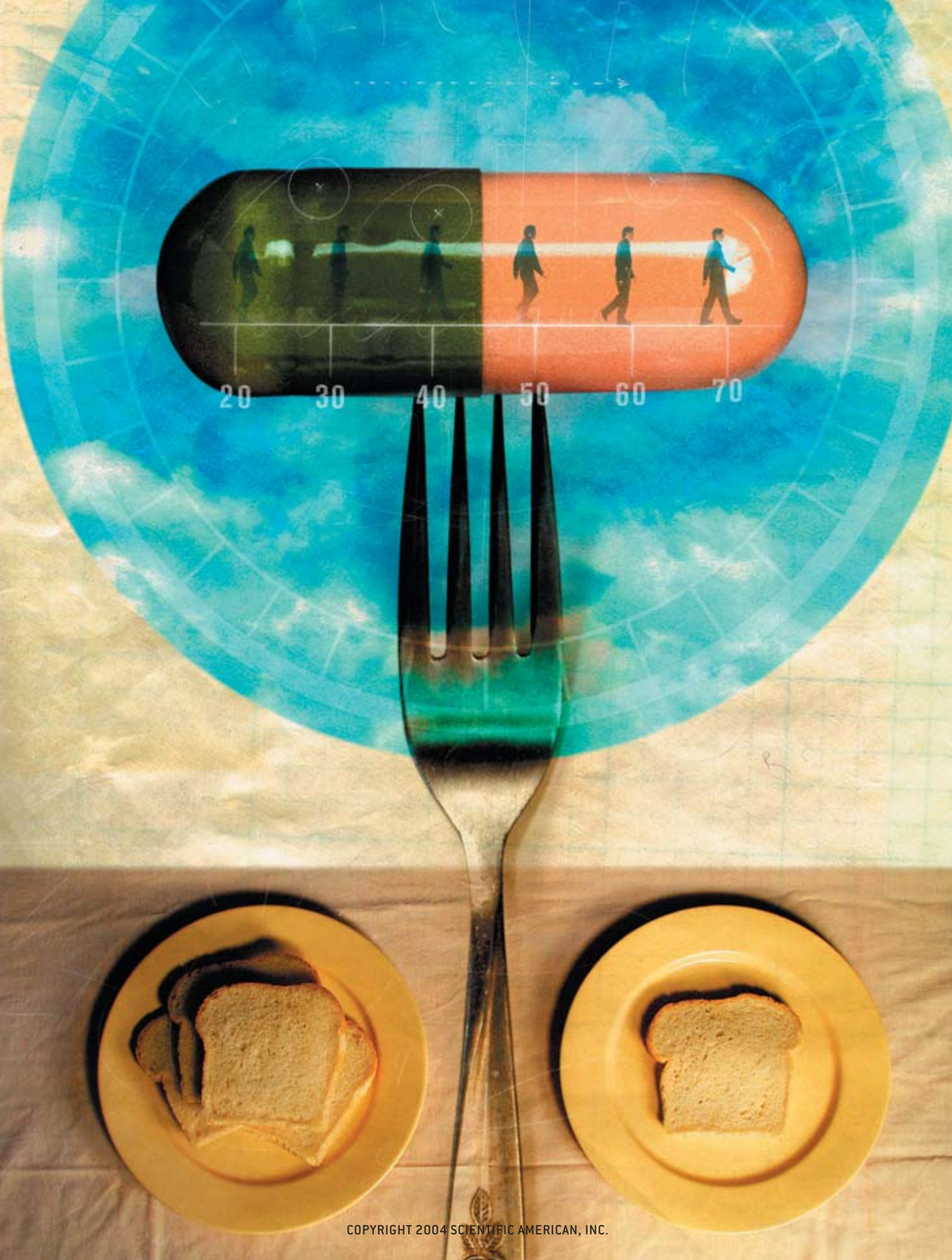
Unfortunately, for maximum benefit, people would probably have to reduce their caloric intake by roughly 30 percent, equivalent to dropping from 2,500 calories a day to 1,750. Few mortals could stick to that harsh a regimen, especially for years on end. But what if someone could create a pill that mimicked the physiological effects of eating less without actually forcing people to go hungry? Could such a caloric-restriction mimetic, as we call it, enable people to stay healthy longer, postponing age-related disorders (such as diabetes, atherosclerosis, heart disease and cancer) until very late in life?

We first posed this question in the mid-1990s, after we came upon a chemical agent that, in rodents, seemed to reproduce many of caloric restriction's benefits. Since then, we and others have been searching for a compound that would safely achieve the same feat in people. We have not succeeded yet, but our failures have been informative and have fanned hope that caloric-restriction, or CR, mimetics can indeed be developed eventually.

Our hunt for CR mimetics grew out of our desire to better understand caloric restriction's many effects on the body. Scientists first recognized the value of the practice more than 60 years ago, when they found that rats fed a low-calorie diet lived longer on average than free-feeding rats and had a reduced incidence of conditions that become increasingly common in old age. What is more, some of the treated animals survived longer than the oldest-living animals in the control group, which means that the maximum life span (the oldest attainable age), not merely the average life span, increased. Various interventions, such as infection-fighting drugs, can increase a population's average survival time, but only approaches that slow the body's rate of aging will increase the maximum life span that an animal can achieve.

The rat findings have been replicated many times and extended to creatures ranging from yeast to fruit flies, worms, fish, spiders, mice and hamsters. Until fairly recently, the studies were limited to short-lived creatures genetically distant from humans. But long-term projects under way in two species more closely related to humans—rhesus and squirrel monkeys—sug-

CALORIC-RESTRICTION MIMETIC would, if successful, enable humans to derive many of the health and life-extending benefits seen in animals on restricted diets—without requiring people to go hungry.



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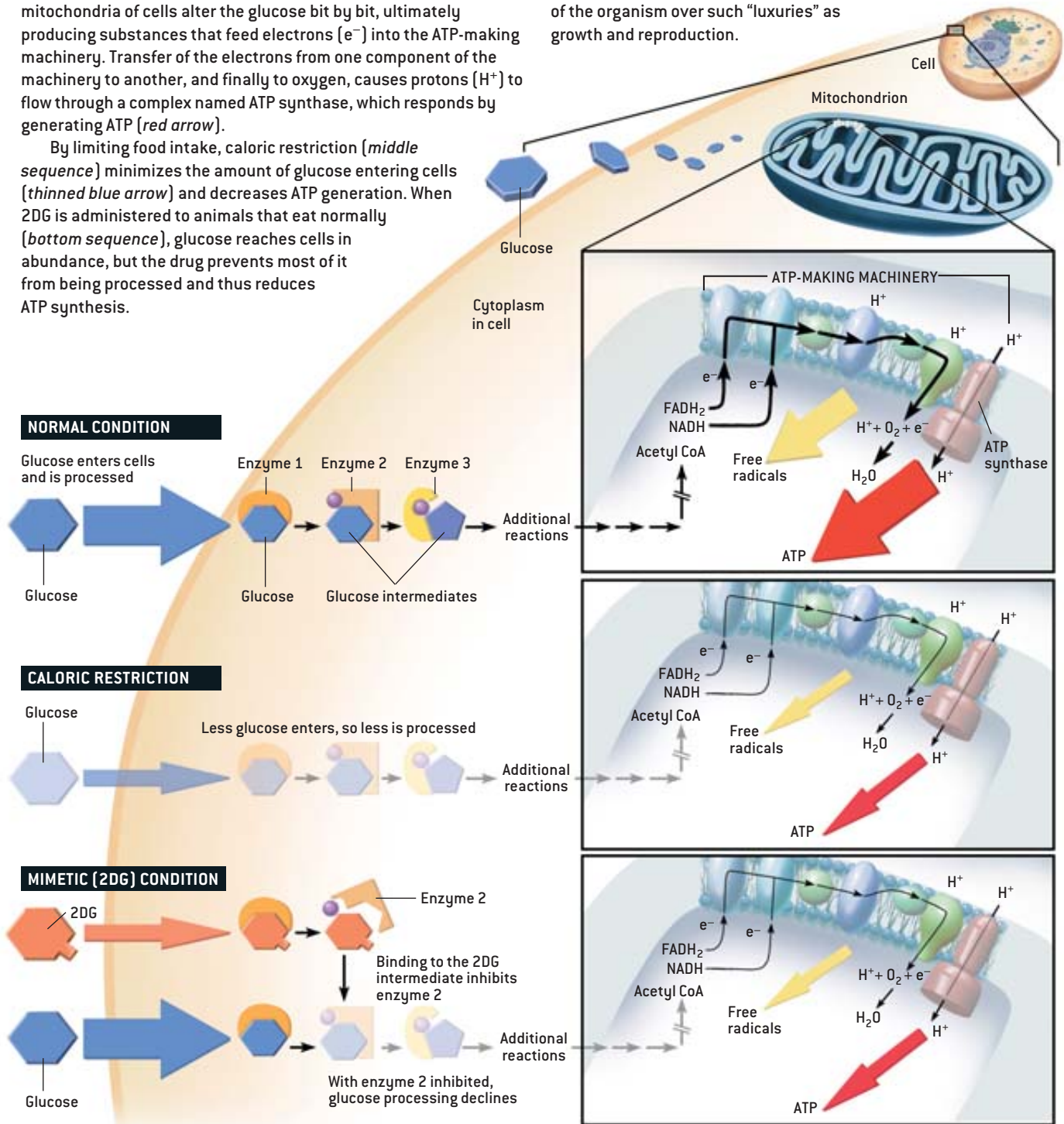
HOW A PROTOTYPE CALORIC-RESTRICTION MIMETIC WORKS

THE BEST-STUDIED CANDIDATE for a caloric-restriction mimetic, 2DG [2-deoxy-D-glucose], works by interfering with the way cells process the sugar glucose. It has proved toxic at some doses in animals and so cannot be used in humans. But it has demonstrated that chemicals can replicate the effects of caloric restriction; the trick is finding the right one.

Cells use the glucose from food to generate ATP [adenosine triphosphate], the molecule that powers many activities in the body [top sequence]. More specifically, after glucose enters cells [blue arrow], a series of enzymatic reactions in the cytoplasm and mitochondria of cells alter the glucose bit by bit, ultimately producing substances that feed electrons (e^-) into the ATP-making machinery. Transfer of the electrons from one component of the machinery to another, and finally to oxygen, causes protons (H^+) to flow through a complex named ATP synthase, which responds by generating ATP [red arrow].

By limiting food intake, caloric restriction [middle sequence] minimizes the amount of glucose entering cells [thinned blue arrow] and decreases ATP generation. When 2DG is administered to animals that eat normally [bottom sequence], glucose reaches cells in abundance, but the drug prevents most of it from being processed and thus reduces ATP synthesis.

Researchers have proposed several explanations for why interruption of glucose processing and ATP production might retard aging. One possibility relates to the ATP-making machinery's emission of free radicals [yellow arrows], which are thought to contribute to aging and to such age-related diseases as cancer by damaging cells. Reduced operation of the machinery should limit their production and thereby constrain the damage. Another hypothesis suggests that decreased processing of glucose could indicate to cells that food is scarce (even if it isn't) and induce them to shift into an antiaging mode that emphasizes preservation of the organism over such "luxuries" as growth and reproduction.



TERESE WINSLOW

gest that primates respond to caloric restriction almost identically to rodents, which makes us more optimistic than ever that CR mimetics could help people.

The monkey projects—initiated by our group at the National Institute on Aging in the late 1980s and by a separate team at the University of Wisconsin–Madison in the early 1990s—demonstrate that, compared with control animals that eat normally, caloric-restricted monkeys have lower body temperatures and levels of the pancreatic hormone insulin, and they retain more youthful levels of certain hormones (such as DHEAS, or dehydroepiandrosterone sulfate) that tend to fall with age.

The animals also look better on indicators of risk for age-related diseases. For example, they have lower blood pressure and triglyceride levels (signifying a decreased likelihood of heart disease), and they have more normal blood glucose levels (pointing to a reduced risk for diabetes, which is marked by unusually high blood glucose levels). Further, we have shown that rhesus monkeys kept on caloric restriction for an extended time (nearly 15 years) have less chronic disease, just as the risk data suggested. They and the other monkeys must be followed still longer, however, before we will know whether low food intake can increase both average and maximum life spans in monkeys: rhesus monkeys typically live about 24 years and sometimes up to 40; squirrel monkeys typically live about 19 years but may live for 28.

The Journey Starts

BY 1995 WE WANTED to know how the many physiological and biochemical changes induced by caloric restriction led to delaying aging in mammals. We suspected that changes in cellular metabolism would be key. By “metabolism” we mean the uptake of nutrients from the blood and their conversion to energy usable for cellular activities. We focused on metabolism in part because the benefits of caloric restriction clearly depend on reducing the overall amount or temporal pattern of fuel coming into the body for processing. Also, caloric restriction affects the aging of a wide va-

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riety of tissues, which implies that it alters biological processes carried out by all cells. Few processes are more fundamental than metabolism.

We specifically wondered whether changes related to metabolism of the sugar glucose would account for the benefits. Glucose, which forms when the body digests carbohydrates, is the primary source of energy in the body—that is, it is the main material used by cells for making ATP, or adenosine triphosphate, the molecule that directly powers most cellular activities. We also wanted to know whether alterations in the secretion and activity of insulin, which influences glucose use by cells, would be important. Insulin is secreted as glucose levels in the blood rise after a meal, and it serves as the key that opens cell “doors” to the sugar. We concentrated on glucose and insulin because reductions in their levels and increases in cellular sensitivity to insulin are among the most consistent hallmarks of caloric restriction in both rodents and primates, occurring very soon after restriction is begun.

Others began publishing data showing that metabolic processes involving glucose and insulin influence life span. For instance, a number of investigations achieved remarkable extensions of life span in nematode worms by mutating genes similar to those involved in molecular responses to insulin in mammals. More recently researchers have found that lowered intake of glucose or disruption of glucose processing can extend life span in yeast. And in fruit flies, genes in-

involved in metabolism, such as INDY (I’m Not Dead Yet), have been implicated in life-span control.

An “Aha!” Moment

AROUND THE TIME the nematode work came out, we began to scour the scientific literature for ways to manipulate insulin secretion and sensitivity without causing diabetes or its opposite, hypoglycemia. Our search turned up studies from the 1940s and 1950s mentioning a compound called 2-deoxy-D-glucose (2DG) that was being tested in rodents for treating cancer but that also reportedly lowered insulin levels in the blood. As we perused the literature further, we had a true “aha!” moment.

The compound apparently reproduced many classic responses to caloric restriction—among them reduced tumor growth, lowered temperature, elevated levels of glucocorticoid hormones and reduced numbers of reproductive cycles. If 2DG really could mimic many aspects of caloric restriction in animals, we thought, perhaps it would do the same for people.

While we were planning our first studies of 2DG, we scanned the literature for details of how it works at the molecular level, learning that it disrupts the functioning of a key enzyme involved in processing glucose in cells. The compound structurally resembles glucose, so it enters cells readily. It is also altered by an enzyme that usually acts on glucose itself. But the enzyme that completes the next of several steps involved in glucose processing essentially chokes on the in-

THE AUTHORS

MARK A. LANE, DONALD K. INGRAM and GEORGE S. ROTH researched caloric restriction for many years at the National Institute on Aging of the National Institutes of Health. Lane is now a project manager at Merck in Rahway, N.J., and continues to collaborate with Ingram and Roth as a guest investigator at the NIA. Ingram is chief of the Behavioral Neuroscience Section at the institute’s Laboratory of Experimental Gerontology. Roth, who spent nearly 30 years as a full-time researcher at the NIA, is now a senior guest scientist there. He is also chief executive officer of GeroSciences, a biotechnology venture devoted to antiaging strategies.

CALORIC RESTRICTION'S VARIED EFFECTS

RODENTS AND MONKEYS on caloric restriction differ from their more abundantly fed counterparts in many ways, some of which are listed below (1–3). Although the influence of these shared changes on aging remains to be clarified, the close similarities in the responses of rodents and monkeys encourage hope that the health-promoting and antiaging effects long seen in rodents (1–4) are universal among mammals, including humans. If so, caloric-restriction mimetics should help people live well longer. The effects marked by capsules (below) have been reproduced in rats by the compound 2DG.



CALORIE-RESTRICTED MONKEY (left) is shorter and leaner than its nonrestricted counterpart (right).

1 EFFECTS INDICATIVE OF ALTERED GROWTH, DEVELOPMENT OR METABOLISM

- Lower body temperatures
- Later sexual maturation
- Later skeletal maturation

2 EFFECTS INDICATIVE OF IMPROVED HEALTH

- Lower weight
- Less abdominal fat

3 EFFECTS INDICATIVE OF REDUCED RISK FOR AGE-RELATED DISEASES (SUCH AS DIABETES AND HEART DISEASE)

- Greater sensitivity to insulin
- Lower fasting insulin levels
- Lower fasting glucose levels
- Lower cholesterol and triglyceride levels
- Lower insulinlike growth factor 1 levels
- Higher levels of “good” (HDL) cholesterol
- Slower decline in levels of the hormone DHEAS

4 EFFECTS FOUND IN RODENTS BUT STILL UNDER INVESTIGATION IN MONKEYS

- Later onset of age-related diseases (including cancer)
- More cell suicide (which may help limit tumor growth)
- Longer average life span
- Longer maximum life span (a strong sign of slowed aging)

intermediate produced from 2DG. When it tries to act on this intermediate, it fails; in addition, its ability to act on the normal glucose intermediate becomes impaired [see box on page 38].

The net result is that cells make smaller amounts of glucose’s by-products, just as occurs when caloric restriction limits the amount of glucose going into cells. Certain of these products serve as the raw material for the ATP-making machinery, which is composed of a series of protein complexes located in intracellu-

lar compartments called mitochondria. Deprived of this raw material, the machinery makes less ATP. In essence, 2DG tricks the cell into a metabolic state similar to that seen during caloric restriction, even though the body is taking in normal amounts of food. As long as the amount of ATP made meets the minimum requirements of cells, this diminished operation of the ATP-making machinery is apparently beneficial.

Why might reduced functioning of the ATP-producing machinery help com-

bat aging? We can’t say with certainty, but we have some ideas. A long-standing theory of aging blames the production of molecules called free radicals. The lion’s share of free radicals in the body are emitted as the ATP-making machinery operates. Over time these highly reactive molecules are thought to cause permanent damage to various parts of cells, including the protein complexes responsible for generating ATP. Perhaps by reducing the rate of ATP production, 2DG and caloric restriction slow the rate at which free radicals form and disrupt cells.

The lack of glucose’s by-products might retard aging in another way as well. Certain of those substances help to induce cells in the pancreas to secrete insulin after an organism eats. Reductions in the amount of those by-products would presumably limit insulin secretion and thereby minimize insulin’s unwanted actions in the body. Aside from indirectly promoting excessive operation of the ATP-making machinery and thus boosting free-radical production, insulin can contribute to heart disease and to undesirable cell proliferation.

We also suspect that cells interpret reduced levels of raw materials for the ATP-making machinery as a signal that food supplies are scarce. Cells may well respond to that message by switching to a self-protective mode, inhibiting activities not needed for cell maintenance and repair—such as reproduction—and pouring most of their energy into preserving the integrity of their parts. If that idea is correct, it could explain why caloric restriction has been shown to increase production of substances that protect cells from excess heat and other stresses.

This adoption of a self-preservation mode would mirror changes that have been proposed to occur on an organismic level in times of food scarcity. In the generally accepted “disposable soma” theory of aging, Thomas Kirkwood of the University of Newcastle in England has proposed that organisms balance the need to procreate against the need to maintain the body, or soma. When resources are plentiful, organisms can afford both to maintain themselves and to grow and reproduce. But when food is

limited, the body invokes processes that inhibit growth and reproduction and takes extra care to preserve the soma.

Recent research has indicated another potential pathway for mimicking CR. A National Institute on Aging group led by R. Michael Anson showed that a regimen of intermittent fasting—in which mice were allowed free access to food on alternating days—resulted in beneficial effects similar to those of caloric restriction, including reduced serum glucose and insulin levels and increased resistance of neurons in the brain to toxic stresses. Surprisingly, the food intake and body weight of Anson's mice did not diverge substantially from control mice that had unlimited access to food. These data suggest that an absolute reduction in caloric intake may not underlie all of CR's effects; rather, hormonal changes related to the stress of intermittent fasting may play an important role.

Testing Begins

IN OUR FIRST experiments on 2DG's effectiveness, we delivered low doses to rats by adding it to their feed for six months. The treatment moderately reduced fasting blood glucose levels (measured after food was removed for 12 hours), body weight and temperature and robustly reduced fasting insulin levels—findings consistent with the actions of caloric restriction itself. Interestingly, after an initial adjustment to the novel diet, the 2DG group did not eat significantly less food than the controls. Thus, these exciting preliminary analyses revealed that it was possible to mimic at least some sequelae of caloric restriction without reducing food intake.

Shortly after we published these results, in 1998, other groups began identifying more ways that 2DG imitates caloric restriction. We are in the midst of conducting long-term rodent trials of 2DG. Initial results confirm our previous findings that 2DG slightly reduces blood glucose and body temperature. We also examined whether 2DG reduces the incidence of cancer and increases life span when fed to animals at low doses from the time they are weaned until they die. Contrary to our expectations, the high-

The task becomes finding other substances that yield 2DG's benefits but are safer.

est dose of 2DG in this experiment did not extend life span. In fact, there was some evidence of early deaths in this group, apparently from toxicity. Interestingly, a lower dose, which had also resulted in some biological effects similar to CR in our early tests, did not show a statistically significant effect on average or maximal life span.

The work so far clearly provides a “proof of concept” that inhibiting glucose metabolism can re-create many effects of caloric restriction. Regrettably, 2DG has a fatal flaw preventing it from being a “magic pill.” Though safe at certain low levels, it apparently becomes toxic for some animals when the amount delivered is raised just a bit or given over long periods. The narrowness of the safety zone separating helpful and toxic doses would bar it from human use. We hope this is not a general feature of CR mimetics.

Moving On

ASSUMING OUR long-term studies confirm that inhibiting metabolism can retard aging, the task becomes finding other substances that yield 2DG's benefits but are safer over a broader range of doses and delivery schedules. Several candidates seem promising in early studies, including iodoacetate, being investigated by Mark P. Mattson's group at the NIA's Laboratory of Neurosciences. In animals, this agent appears to protect brain cells from assaults by toxic substances, just as 2DG and caloric restriction do. Treatment with antidiabetic medications

that enhance cellular sensitivity to insulin might be helpful as well, as long as the amounts given do not cause blood glucose levels to fall too low.

A great deal of research implicates glucose metabolism in regulating life span, yet other aspects of metabolism can also change in reaction to caloric restriction. When the body cannot extract enough energy from glucose in food, it may shift to breaking down protein and fat. Pharmaceuticals that targeted these processes might serve as CR mimetics, either alone or in combination with drugs that intervene in glucose metabolism. Some compounds that act in those pathways have already been identified, although researchers have not yet assessed their potential as CR mimetics. Drugs that replicate only selected effects of caloric restriction could have a role to play as well. In theory, antioxidant vitamins might fit that bill. Research conducted to date, however, indicates that this particular intervention probably will not extend longevity.

Unlike the multitude of elixirs being touted as the latest antiaging cure, CR mimetics would alter fundamental processes that underlie aging. We aim to devise compounds that fool cells into activating maintenance and repair activities that lead to greater health and longevity of the organism. If scientists can develop agents that offer the benefits of 2DG without its drawbacks, they will finally enable people to have their cake—a longer, healthier life—and eat it, too. **SA**

MORE TO EXPLORE

2-Deoxy-D-Glucose Feeding in Rats Mimics Physiological Effects of Caloric Restriction.

Mark A. Lane, George S. Roth and Donald K. Ingram in *Journal of Anti-Aging Medicine*, Vol. 1, No. 4, pages 327–337; Winter 1998.

Caloric Restriction in Primates and Relevance to Humans. George S. Roth, Donald K. Ingram and Mark A. Lane in *Annals of the New York Academy of Sciences*, Vol. 928, pages 305–315; 2001.

A scientific position statement on human aging mentioned at the start of this article is available at www.sciam.com/agingstatement.cfm