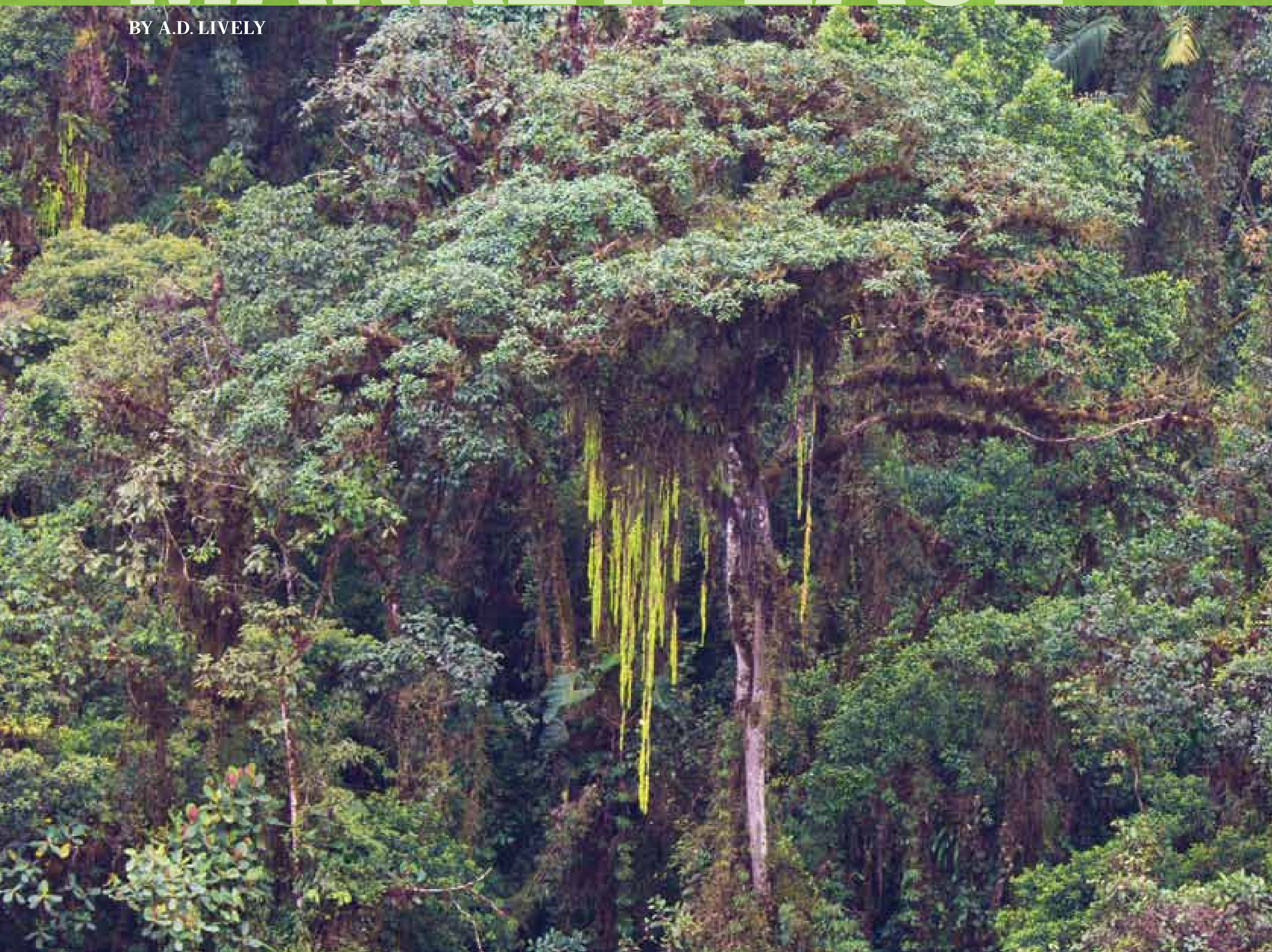
RESEARCH

# FROM MOLECULE TO MARKETPLACE

## Scientists develop new form of Vitamin E to combat radiation

BY A.D. LIVELY



"When you're in the jungle, you've got to remember two things," says Cesar Compadre, PhD, professor of pharmaceutical sciences at the University of Arkansas for Medical Sciences (UAMS) College of Pharmacy. "Look first before you put your feet anywhere, and look first before you put your hands anywhere."

**"If you don't make red blood cells you get anemia. If you don't make white blood cells, you get an infection. And if you don't make platelets, you bleed to death. And all three happen simultaneously with radiation poisoning."**

LABORATORY SCIENTISTS do not typically respond to inquiries about their translational research projects with advice on how to safely navigate the Amazonian rainforest. But for Compadre, who grew up in Mexico City watching his grandmother use traditional medicines like aloe and chamomile, the natural world and the drug development laboratory are deeply connected—and success in both arenas is dependent upon forging solid, productive partnerships.

Nowhere is this belief more evident than in his longtime partnership with Philip J. Breen, PhD, associate professor of pharmaceutical science. The two investigators arrived at UAMS 27 years ago, "within about a month of each other," says Breen. They began collaborating almost immediately, with Breen's expertise in pharmacokinetics (the way drugs move through and interact with the body) serving as an excellent complement to Compadre's molecular modeling strengths.

"We are a very good team," says Compadre. So good, in fact, they were able to take their first basic science collaboration successfully from the laboratory into the marketplace as Cecure®, a food processing antimicrobial currently licensed by North Little Rock company Safe Foods.

## A new vitamin E molecule

The duo's latest project, "Analysis of Tocoflexols: A New Class of Radiation Protection Agents with Enhanced Bioavailability," was supported in part by a $50,000 Pilot Award from the UAMS Translational Research Institute (TRI), which they maximized by using cutting-edge computational modeling techniques to discover and synthesize a new, powerful type of vitamin E called tocoflexol.

In addition to possessing the antioxidant qualities of all vitamin E, tocoflexol holds promise for development in a number of directions, including as a statin-like (cholesterol reducing) and possibly even anticancer drug. Most notably, however, it shows tremendous potential as a safe, effective, and widely available treatment for unsafe exposure to any type of radiation—including nuclear.

While there is still much work to do before a clinical application of the molecule is realized, cautions Breen, "without the TRI, and their willingness to take on new, higher-risk projects, we wouldn't have gotten nearly as far as we've gotten."

And so far, they have not only developed a novel, patentable molecule but also started a business, Tocol Pharmaceuticals, along with their clinical partner and radiation research expert Martin Hauer-Jensen, MD, PhD, professor of pharmaceutical sciences, surgery, and pathology and associate dean for research in the UAMS College of Pharmacy.

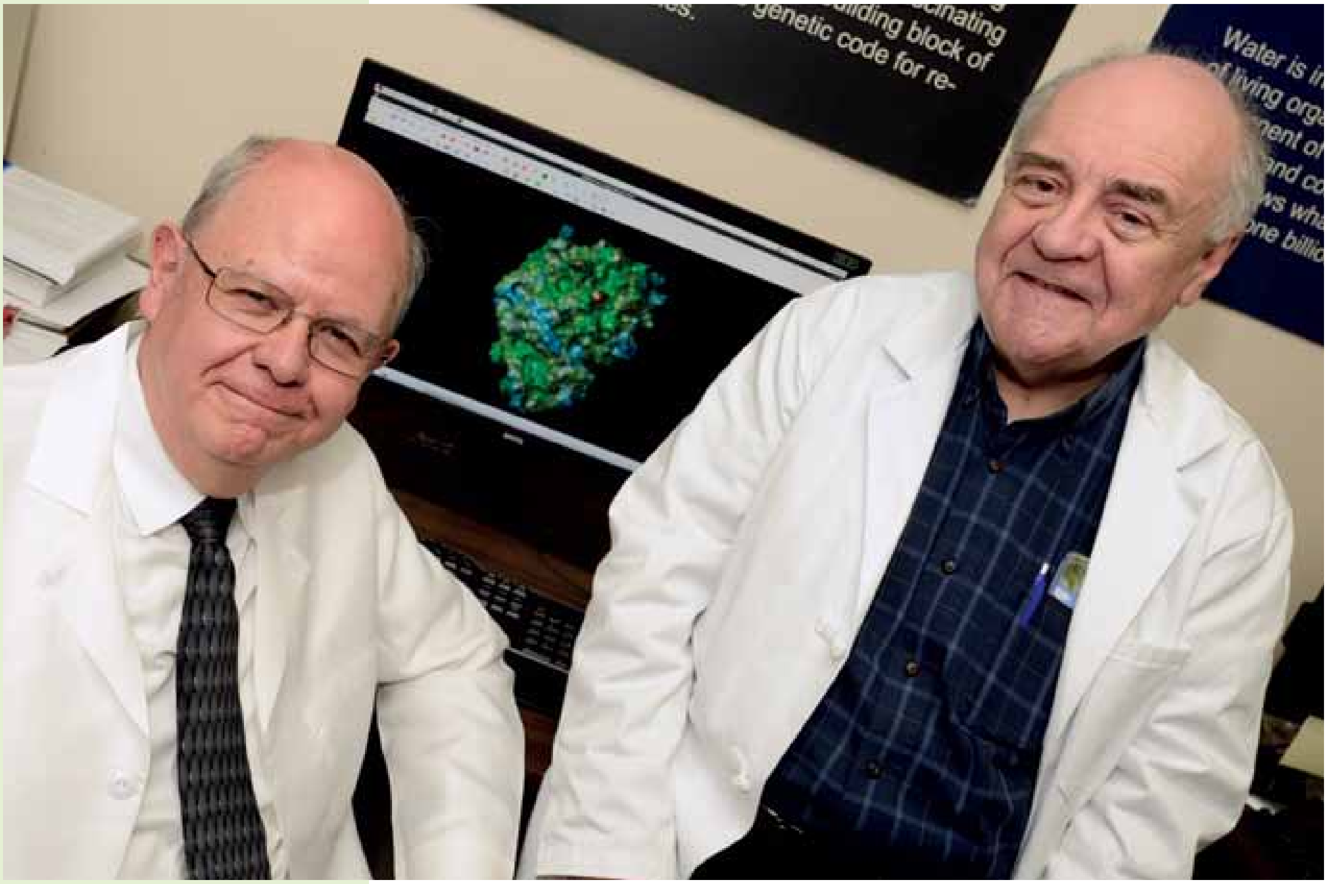
## Radioprotection and Radiomitigation

Given the ever-present possibility of a radiological emergency, be it accidental or terrorist-initiated, there is an urgent need for therapeutic agents with minimal side effects that can protect from and/or mitigate the damage of radiation exposure.

Not only would such a treatment have a lifesaving impact in the event of a large-scale disaster, but it might also be able to help in other ways: by lessening the side effects of radiation therapy for cancer patients, for example, or providing ongoing protection for people who regularly work with or near radiation.

"It is very nasty to see a death from a high dose of radiation," says Breen. The lining of the entire gastrointestinal tract sloughs off, including the lining of the mouth and throat. Radiation also severely damages bone marrow, where the constituents of blood are manufactured.

"If you don't make red blood cells you get anemia," he continues. "If you don't make white blood cells, you get an infection. And if you don't make platelets, you bleed to death. And all three happen simultaneously with radiation poisoning."




**"We are a very good team,"** says Cesar M. Compadre, PhD, professor of pharmaceutical sciences at the University of Arkansas for Medical Sciences (UAMS) College of Pharmacy, left, with Philip J. Breen, PhD, associate professor of pharmaceutical science.

Breen, who served in the Army during the Vietnam War as a pharmacy specialist, first became aware of the need for an effective radioprotectant when he learned in military training that the only real defense from radiation was to stay away from it. At that time, iodine was the sole treatment available, and its protection was limited to the thyroid. And since then, attempts to address this gap in the pharmacopeia have only resulted in drugs with nearly intolerable side effects.

## A flash of light

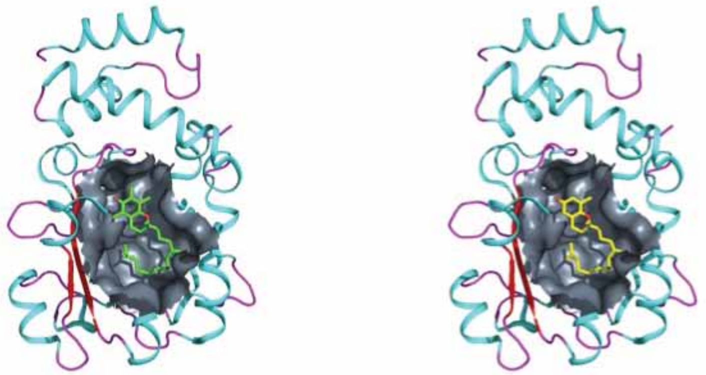
Compadre and Breen's inspiration for the tocoflexol molecule began when they attended a seminar by K. Sree Kumar, PhD, a senior research scientist from the Armed Forces Radiobiology Research Institute and long-term collaborator of Hauer-Jensen.

Kumar already knew that certain types, or vitamers, of vitamin E had special qualities. But what he didn't know was why some were active and some were not.

**c. 500** For Christians, disease is still widely believed to be a result of sin or evil spirits leading to some rather random and seldom helpful treatments. Some lay physicians do, however, use examination, palpation, percussion, and examination of feces, urine, and semen, as well as dream interpretation in making diagnoses.

**c. 1300** European physicians regularly use uroscopy to determine illness.

**1592** The first thermometer is invented by Galileo Galilei



Alpha-tocopherol

Tocoflexol

Comparison of the binding of AT (green, left) and tocoflexol (right, yellow) in the binding cavity of ATTP.

> **Maybe, they thought, it would be possible to make a version of vitamin E that combined the half-life and bioavailability of the tocopherols with the "superpowers" of the tocotrienols to create something the body could actually use.**

"Not all vitamin E is the same," says Breen.

The most widely-studied vitamers of vitamin E are the tocopherols, with alpha-tocopherol being the form most frequently included in supplements, cosmetics, and other commercial products. Complete tocopherol deficiency can cause pregnant women to miscarry. But otherwise "it's kind of a general antioxidant, but it doesn't do much else."

As Kumar's presentation outlined, molecules in the lesser-known tocotrienol group are also antioxidants, but they exhibit many other qualities that the tocopherols do not, including statin-like behaviors and radioprotection. But they have a problem: they have such a short elimination half-life (two hours compared to the twelve-hour duration of alpha-tocopherol) that they don't stay in the body long enough to be therapeutic.

"As we listened to Kumar's presentation," says Compadre, "we got a flash of light."

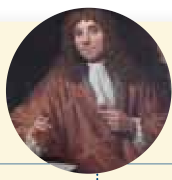
Maybe, they thought, it would be possible to make a version of vitamin E that combined the half-life and bioavailability of the tocopherols with the "superpowers" of the tocotrienols to create something the body could actually use.

### New tail, new half-life

All vitamin E is lipophilic, or poorly soluble in water (and therefore blood). Tocopherol molecules address this by binding in the liver with a fatty acid called ATTP (alpha-tocopherol transfer protein), which allows them to go back into the blood and recirculate. This "recycling" is responsible for their long half-life. Tocotrienol, however, does not obtain solubility by binding with ATTP and so is rapidly eliminated by the liver.

Why doesn't tocotrienol bind with ATTP? The answer, Compadre and Breen

**1612** Italian physician Sanctorius adapts the thermometer for clinical use.



**1660** Anton van Leeuwenhoek creates the light microscope and later publishes drawings of the bacteria he observes.

**c. 1700** Stephen Hales creates a device to measure blood pressure in horses.

hypothesized, was in the structure of the molecules themselves.

Tocotrienol molecules have a "tail" containing three double bonds, making it inflexible, while the tocopherol tail contains no double bonds. Using advanced computational imaging techniques, they were able to confirm that this flexible tail was responsible for tocopherol's ability to curl up and "nestle" within the ATTP molecule, while the stiff tail on tocotrienol prevented bonding.

Using predictive molecular dynamics simulations to test out different configurations, Compadre discovered that eliminating the middle of the three double bonds in the tail created a flexible "joint" that allowed it to bond with ATTP and stay in the body much longer. He named the new molecule tocoflexol, in honor of its flexible tail.

## Back to nature

The commercially available tocotrienol that serves as the starting point for tocoflexol synthesis is currently $10,000 a gram, a price tag that makes more extensive laboratory


Visiting Dr. Cartuche's Medicinal Plant Garden

1714 The mercury thermometer is created by Gabriel Fahrenheit.

1754 Chest percussion becomes an accepted diagnostic tool.

1816 The stethoscope is invented by R.T.H. Laennec.

Dr. Compadre's guide treats him with traditional medicine after an accident in the jungle.

testing challenging and puts mass production entirely out of reach.

While the group continues to refine the tocoflexol synthesis process to increase the yield per gram of tocotrienol, Compadre is turning to another important collaboration, with the Universidad Técnica Particular de Loja in Ecuador, for a possible solution.

After a 2007 visit to the country with his daughter, he returned so inspired by the wide array of plant life in the Amazonian jungle that he forged a "memorandum of understanding" involving knowledge and student exchange between UAMS and the Universidad. "I could spend my whole life just studying the ecology of the plants there," he says.

Now he is hopeful of finding a plant source for tocotrienol that can be incorporated into the production of tocoflexol—but only if it can be done ethically and cooperatively.

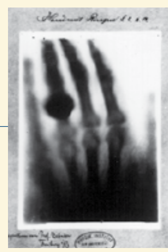
"You know, it's a very delicate situation. There is the concept that the knowledge belongs to the community. Because for a long, long time, people have just gone into the rainforest and grabbed things. There are a lot of biopirates."

The collaboration with Loja also offers

1817 British physiologist Marshall Hall publishes On Diagnosis.

c. 1850 Lab tests are developed for common diseases such as tuberculosis, cholera, typhoid, diphtheria, though the cures lag far behind.

1895 Wilhelm Conrad Roentgen discovers x-rays and creates an image of his wife's hand.



Compadre the opportunity to interact with traditional community healers "to see how I can learn from them, and how I can help."

"It's all in plants," he says. "And you have to learn how the plants themselves survive."

Which is exactly what he was doing when he learned—the hard way—about the importance of careful hand and foot placement in the jungle:

"Well, I got distracted and made a double mistake. I didn't watch where I was putting my feet, and I slid. And then I was falling, so I didn't see where I was putting my hands."

Fortunately, he was able to grab a nearby vine to slow his fall; unfortunately, it was a vine studded with giant thorns, a dozen of which were embedded in his hand by the time he slid to a stop. The pain was excruciating, and he dreaded the infection that would likely result from multiple puncture wounds in such an unsterile environment.

Then one of his guides obtained a handful of leaves from a nearby tree, quickly removed the thorns from Compadre's hand, and applied the juice from the leaves to the punctures.

"We didn't see any infection from any of these places," says Compadre. "But there was one small place we missed, and the next day—oh, it was infected, and painful, and we had to use Western medicine to cure it. Whatever was in that leaf, it worked."

**IT'S ALL IN PLANTS. AND YOU HAVE TO LEARN HOW THE PLANTS THEMSELVES SURVIVE.**

### The Future of Tocol Pharmaceuticals

"There's the science, and then there's the commercialization," says Breen. "It's time to put our business hat on."

Another critical partner on the path to the commercialization of tocotrienol is technology licensing office and life science business incubator UAMS BioVentures, which also supported earlier commercialization of Cecure®.

"We've been working closely with Dr. Compadre and his colleagues on a number of fronts, including helping him license some of the patents that he has developed here," says BioVentures Director Nancy Gray, PhD.

With tocoflexol, says Compadre, "we invented something that wasn't there before. So now we patent the compound; we patent the way to make it; and we patent the use of these compounds for radiation."

Business models involving the development of molecules into therapeutic agents have "a very long-term horizon to commercialization, and the capital requirement is large," says Gray. "I think, however, what the marker of success for this small company will be is to progress research and get more proof-of-concept studies in hand."

At that point, she continues, Tocol Pharmaceuticals "will either need to partner with a larger company or raise a lot of money" in order to meet what is almost certain to be a colossal demand for a safe, effective treatment against the damage of radiation exposure.

"We were able to keep Cecure® in Arkansas, and license it to what is now a multi-million dollar Arkansas company, and it is a big provider of jobs," says Breen, "And I think the same, maybe bigger, will happen with Tocol Pharmaceuticals."

"We want people to know that, with the right partners, this kind of research and this kind of business-building can happen here in Arkansas," says Compadre. "We are proving that it's possible." ■

**1896** Improving on Hale's manometer, S. Riva Ricci invents the sphygmomanometer for measuring blood pressure.

**1900** Chest x-rays allow for early diagnosis of tuberculosis

**c. 1905** Radiology becomes a sub-specialty