



Mothers and children in the Philippines protest in 2013 against golden rice, which is genetically designed to contain the vitamin A precursor β -carotene.

BIOTECHNOLOGY

Against the grain

Golden rice could help to end a nutritional crisis — but only if researchers can overcome some daunting technical and political hurdles.

BY MICHAEL EISENSTEIN

The years of frustration are audible in Adrian Dubock's voice when he talks about the development of golden rice. "It's been an uphill struggle," he says, "but I think we're winning."

Golden rice was created in response to a nutritional crisis that grips some of the poorest communities in the world. According to the World Health Organization, every year between 250,000 and 500,000 children lose their eyesight because of vitamin A deficiency. Half of them will die within a year of going blind, primarily because their immune systems did not have enough vitamin A to function properly (see 'Prevalence of vitamin A deficiency').

Because golden rice is genetically designed to produce β -carotene — a precursor to vitamin A — it would seem to be an ideal solution to vitamin A deficiency in rice-dependent regions of the world. In many of these areas, including south and southeast Asian nations

such as India, Bangladesh, Indonesia and the Philippines, rice is the primary food source, comprising up to 70% of the daily caloric intake. "In the Philippines, they literally don't call a meal a meal if it doesn't have rice in it," says Dubock, who is manager of the Golden Rice Project.

Rice is relatively affordable and filling, but it has its shortcomings as a staple. For example, it is only a marginal source of many important vitamins and nutrients, including vitamin A. What's more, most grains undergo a polishing process that helps to prevent spoilage, but which also reduces the nutritional value even further, leaving consumers of rice-based diets vulnerable to malnutrition.

However, genetically modified (GM) agriculture remains deeply controversial, and scientific and regulatory setbacks have stopped golden rice from reaching those who need it most. Dubock says that those who want to bring it to the masses

must be ready to wage a multipronged campaign to overcome the research hurdles, win public confidence and inspire government support. It is bound to be a long road. But with so much at stake, Dubock is committed to moving forward, and he is not alone.

A PROMISING START

The seeds of golden rice were sown in 2000, when plant scientist Ingo Potrykus of the Swiss Federal Institute of Technology in Zurich and cell biologist Peter Beyer of the University of Freiburg in Germany first attempted to insert genes that control β -carotene synthesis into rice plants¹. The newly acquired β -carotene gave the rice a distinctive yellow-orange hue that led to its now familiar nickname, but it was unable to address the nutritional needs of vitamin A-deficient consumers. A collaboration with Swiss biotechnology company Syngenta, based in Basel, led to a greatly improved version that could deliver more than half of the recommended daily intake of β -carotene in a single serving. Syngenta

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subsequently transferred control of the product to the Golden Rice Project under the auspices of a humanitarian board of scientists and public-health experts that, under the leadership of Potrykus and Dubock, has been tasked with making golden rice available to low-income farmers and researchers in the public sector throughout the developing world.

Potrykus and Beyer carried out laborious, trial-and-error testing of different combinations of metabolic genes and methods of introducing them into the rice genome. The initial version of golden rice, created with genes extracted from daffodils (*Narcissus pseudonarcissus*) and bacteria, produced only 1.6 micrograms of β -carotene per gram of rice, which was woefully inadequate for use as a dietary supplement. The improved version developed at Syngenta in 2005 replaced the daffodil gene with an equivalent gene from maize (corn)². Golden rice 2, as it became known, delivered far superior β -carotene production — up to 37 micrograms per gram — and safe delivery of β -carotene to human consumers has now been demonstrated in multiple trials.

REALITY BITES

Several technical problems have dogged golden rice's journey from the greenhouse to the field, however. Since 2010, the Philippines-based International Rice Research Institute (IRRI) and the Golden Rice Project have been working with the Philippine Rice Research Institute (PhilRice) to conduct field trials spanning three growing seasons at five sites across the Philippines.

Unfortunately, the golden rice strain selected for field testing does not grow as well as local rice varieties, limiting its appeal to struggling farmers. "The final product has to be so good that it will be readily adopted by farmers in terms of agronomic traits — yield, disease resistance, quality and ability to withstand adverse conditions — as well as β -carotene production," says Antonio Alfonso of PhilRice, who led the trials.

Identifying a gene combination that delivers enough β -carotene is only half the battle. Scientists must insert these genes into the genome of the rice plant in a way that allows them to be expressed without interfering with other genes. As a further complication, crops that work well in the lab may not be the same varieties that people like to eat and grow, so agronomists must perform a lengthy process of 'introgression' in which they breed the GM strain repeatedly with popular strains. The final goal is to produce plants that contain the new trait, but otherwise resemble local strains as much as possible.

Inadequate introgression may have prevented golden rice from thriving, says Inez Slamet-Loedin, who works on transgenic bio-fortified rice at IRRI. "Syngenta was working with an American rice variety that is not suitable for the tropics," she explains. Researchers at IRRI crossed this rice with tropical strains grown in the Philippines, but she estimates that the hybrids acquired only 82% of the local rice's genetic background. "We probably need that to



Golden rice (left) and conventional rice.

be closer to 98%," she says.

The position of the introduced genes may also have been problematic. Syngenta provided IRRI with six different 'insertion events' — individual rice strains with the β -carotene-producing maize genes incorporated at different sites in the genome. The golden rice research team focused on one particular insertion event known as GR2-R, which performed well in greenhouse testing but failed to thrive in the field. Subsequent investigation has suggested that the insertion site could be interfering with the expression of a gene linked to root development.

The golden rice team has access to multiple strains with distinct insertion events, but it focused on GR2-R to streamline the regulatory process surrounding testing. This is partly due to a document called the Cartagena Protocol on Biosafety, which has been ratified by 165

countries and the European Union. The protocol encourages special caution for the regulation of 'living modified organisms,' defined as organisms "that possess a novel combination of genetic material obtained through the use of modern biotechnology". Given the expense and paperwork required to test any particular GM strain, most groups focus on a single event to lead through the regulatory process — essentially betting the house on a single spin of the wheel. "It would be much easier if you could just plant everything in the field and test it," says Matin Qaim, an economist at the University of Göttingen in Germany.

The IRRI researchers are shifting their focus to another event selected from the Syngenta pool, GR2-E. "This was our back-up all along so it's not like we're starting from zero," says Slamet-Loedin. "But we will need to generate some additional regulatory data." This is not as simple as it sounds, and Qaim says the switch has "cost a year or two in terms of further development".

THE GM STIGMA

The scientific problems can be solved, but public fears over GM organisms (GMOs) may be a bigger obstacle. Activists in Europe and North America have shaped the debate by raising doubts and concerns over the environmental impact and health risks of 'unnatural' GMOs, even though scientists have pointed to numerous studies that should assuage these worries. Dubock describes surveys in the Philippines that found that many farmers were interested in golden rice, even when educated about how it was created — until they heard the term GMO. Slamet-Loedin reports similar experiences

IRON RICE

Rice can be used to deliver another key nutrient

Many communities that are short of vitamin A also lack other essential nutrients, such as iron. Scientists see an opportunity to use modified rice as a vehicle for this mineral, too. "Rice has the lowest iron level of any of the major cereals," says food biotechnologist Alexander Johnson⁵ of the University of Melbourne in Australia. Polished rice contains 2–4 parts per million (p.p.m.) of iron, he says, and it takes about 14 p.p.m. to improve the iron status of people who get most of their calories from rice.

Attempts to obtain iron-fortified rice by conventional breeding have yielded only a twofold improvement in iron content. But two genetically engineered strains — one developed by Johnson and his colleagues, the other by Wilhelm Gruissem's group⁶ at the Swiss Federal Institute of Technology in Zurich — could offer an alternative. Both use genes encoding nicotianamine synthase, an enzyme that increases iron transport within

the plant, and ferritin, a storage protein that helps draw iron from the environment.

Johnson's group is more than halfway through a five-year field trial in the Philippines and Colombia, and the early data are promising. "Under field conditions, we are obtaining rice with iron at 15 p.p.m. with no yield penalty and good grain quality," says Inez Slamet-Loedin of the International Rice Research Institute in the Philippines, whose team is collaborating closely with Johnson. Gruissem is seeking potential partners to move his rice from the greenhouse to field trials, but he has an even more ambitious goal — to engineer a single, super-nutritious crop with multiple biofortification traits. "We want to stack β -carotene production with iron or vitamin B6 production," he says. "We have the technology and we should try it out — although I would say it's going to be something of a 'regulatory adventure'." **M.E.**

working with nutritionists. “I met one who was so happy when he heard how it would make children healthier, but when I said it was a GMO he suddenly changed his mind,” she says. “People have this image of it as a monster.”

Greenpeace International, a respected non-governmental organization with a long history of standing up for environmental causes, is one of the most prominent opponents of golden rice. As part of a broader campaign against GMOs, the group says that the rice may be unsafe to eat, might be harmful to the environment and could disempower local farmers. As an alternative, the group wants to see ‘traditional’ agricultural methods and conventional strategies of dietary supplementation.

In this climate, any minor misstep by golden rice scientists can become a major setback. For example, a study conducted in China in 2008 by nutrition scientist Guangwen Tang of Tufts University in Boston, Massachusetts, showed that a serving of golden rice was both safe and effective at boosting serum vitamin A levels in young children³. Shortly after the study’s publication in 2012, Greenpeace issued a press release claiming that the investigators failed to disclose that they were testing a GM strain, describing the push for golden rice as “irresponsible and dangerous” and condemning Tang’s team for using “children as guinea pigs”.

The Chinese government subsequently sacked three local scientists for their involvement. A review by Tufts concluded that the researchers had handled consent improperly by failing to adequately inform parents that the rice being tested was GM and inappropriately altering the study protocol after receiving institutional approval. Tang is now engaged in a legal battle with both the university and the American Society for Nutrition to fight the retraction of her study.

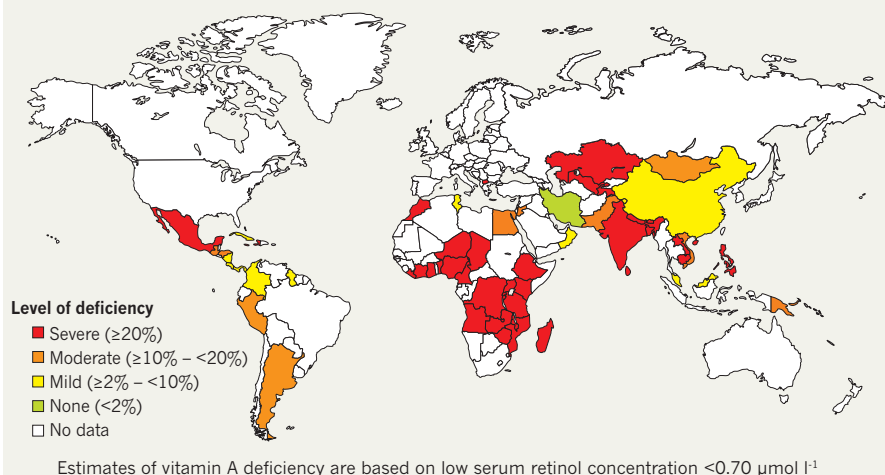
Greenpeace scientist Janet Cotter thinks it is too early to make assertions about the safety of golden rice. “You may very well be able to create β -carotene in rice, but then the question is, what else has changed?” she says. “There’s no way you can test every single compound in a plant, and you still won’t know about the food safety in terms of the wider population.” But proponents of golden rice see this as an unnecessary objection to a promising solution to malnutrition. “We cannot ever say the risk from GM crops is zero,” says Ronald Herring, a political scientist specializing in biotechnology policy at Cornell University in New York. “But I don’t know of any actually authenticated hazard, and I think the science all points in the same direction.”

The vigour of the opposition may seem surprising, given that golden rice was created to keep impoverished children healthy. According

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PREVALENCE OF VITAMIN A DEFICIENCY

Map showing level of serum retinol (an indicator of vitamin A deficiency) in pre-school age children. Data were collected by the World Health Organization between 1995 and 2005 from populations at risk.



to Qaim, this is precisely the problem: many in the anti-GM movement perceive golden rice as a Trojan horse that, if made widely available, will fundamentally alter the discourse about agricultural biotechnology. “They’re looking for propaganda to show that we need GM crops,” says Cotter.

Golden rice does sidestep many of the arguments typically marshalled against GM crops. A common environmental concern is that genetically engineered traits will disperse into other plant species, potentially resulting in undesirable consequences such as hardier weeds. But β -carotene is only beneficial for human consumers and offers no clear advantage for the plant itself. “It’s not going to make those plants fitter,” says Qaim. “It’s not going to spread far.”

Furthermore, the trait is being introduced into the rice strains normally grown by local farmers, so adopting golden rice will not leave growers beholden to biotech firms to purchase new seeds each season, nor should it alter their agricultural practices. Many opponents of golden rice argue that greater dietary diversity would be a better solution, and this is certainly true. Unfortunately, many of Asia’s poorest and most malnourished people lack — and are unlikely to acquire — reliable access to mangoes, carrots or other fruits and vegetables rich in vitamin A. Golden rice, in contrast, could easily be integrated into local diets.

Unfortunately, each year of delay translates directly to lives lost through malnutrition. Qaim and his colleagues have analysed the potential health and economic benefits of adopting golden rice and estimate that the crop could potentially save up to 40,000 lives per year worldwide in a highly cost-effective manner⁴. The results from the human consumption trials in China suggest that, in some conditions, the return on the investment could be even greater. “Even our optimistic assumptions may be on the pessimistic side,” says Qaim. “Not every consumer

will grow or eat it, and this is not a ‘magic bullet’, but it is another potential instrument in the fight against malnutrition.” The early results with golden rice have been so encouraging that other research groups are now investigating the use of rice as a vehicle to combat deficiencies of other essential nutrients, such as iron (see ‘Iron rice’).

GOLDEN FUTURE

The setbacks have been unfortunate, but the golden rice researchers are confident they will find a winning combination in the years ahead. The regulatory environment will be a decisive factor, and both Alfonso and Slamet-Loedin praise the support their project has received from the Philippine government. They are hopeful that their country will back commercialization once the safety and efficacy data roll in from the field trials. If the crop is good, they say, the benefits should sell themselves.

The governments and protestors are important, of course, but the tipping point for GM rice may come from the farmers themselves. As farmers start to recognize the advantages of some of these new rice strains, the seeds are bound to find their ways to paddies around the world, with or without official government sanction. Herring predicts that golden rice — and GM foods in general — will someday revolutionize agriculture in much the same way that MP3 players changed the music industry.

One way or another, good ideas — and good crops — will eventually take root. ■

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1. Ye, X. *et al.* *Science* **287**, 303–305 (2000).
2. Paine, J. A. *et al.* *Nature Biotechnol.* **23**, 482–487 (2005).
3. Tang, G. *et al.* *Am. J. Clin. Nutr.* **96**, 658–664 (2012).
4. Stein, A. J., Sachdev, H. P. S. & Qaim, M. *World Dev.* **36**, 144–158 (2008).
5. Johnson, A. A. T. *et al.* *PLoS ONE* **6**, e24476 (2011).
6. Wang, M. *et al.* *Front. Plant Sci.* **4**, 156 (2013).