Golden Rice, Part 3: A thoroughly studied, safe and nutritionally enhanced GMO crop approved by Australia, Canada, New Zealand and the US, but vilified by Greenpeace and 'environmental' activists

Golden Rice differs from white rice only in that it contains ?-carotene, that is, provitamin A, which the human body converts to vitamin A. Golden Rice contains no vitamin A itself. So the question about safety relates principally to ?-carotene, which is anyway ubiquitous in a balanced human diet and the environment. At the levels found in food, ?-carotene is a safe source of vitamin A, and classed as 'generally recognized as safe' (GRAS), by the United States Food and Drug Administration (US FDA). At these physiological doses, consumption of ?-carotene over several years has no adverse health effects. The human body only converts to vitamin A, in the form of circulating retinol, the amount of ?-carotene necessary, with the rest being excreted or stored unchanged in body tissues (e.g. fat, liver, etc.). It is impossible to induce vitamin A toxicity by consuming ?-carotene (pers. comm. Dr. R Russell).

In all ?-carotene-containing crops, immediately after harvest the level of ?-carotene reduces. For Golden Rice carotenoid degradation mechanisms have been thoroughly investigated and the products of degradation quantitated. Additionally, 102 plant food items from Philippine markets, together with orange- or yellow-colored soft drinks, as well as non-GMO field grown, in all cases, orange maize cobs and yellow cassava storage roots from Zambia, and orange-fleshed sweet potato tubers from Uganda, were analyzed for the cleavage products of ?-carotene, apocarotenoids.

This article is part three of a four-part series on golden rice based on a book chapter Golden Rice: To Combat Vitamin A Deficiency for Public Health. Golden Rice, Part 1: The story of a GMO crop that could benefit billions of children a year, Part 2: Will nutritionally enhanced rice work and help solve malnutrition in developing countries?; Part 4: Do biofortified crops make economic sense?

The potential risks arising from 'aberrant plant carotenoid synthesis' in genetically modified plants, including Golden Rice, or from non-GMO crops biofortified with pro-vitamin A, have been thoroughly investigated, the authors reporting that 'Our analysis and quantification of ?-carotene derived cleavage products across biofortified and non-biofortified crop plant tissues combined with the calculation of potential exposure document no reason for concern.'

For the formal regulatory approvals for the use of a GMO crop in food, as animal feed or in food or feed processing, on a country by country basis, detailed data sets have to be submitted. For permission to grow a GMO crop in a country, additional data have to be generated and submitted showing environmental safety. The ‘food, feed and processing’ data package developed for Golden Rice GR2E is extensive (42 megabytes of data). It is available without cost to all Golden Rice licensee countries consistent with long-standing Golden Rice Humanitarian Board policy. Here are the key summaries of the regulatory data submission made in the Philippines:

> Rice event GR2E (IR-ØØGR2E-5) was developed using recombinant-DNA techniques to express elevated levels of provitamin A (mainly ?-carotene) in the rice endosperm, which is converted in the body to vitamin A. GR2E rice was produced by Agrobacterium tumefaciens-mediated transformation of embryogenic rice calli with plasmid pSYN12424 resulting in the introduction of the phytoene synthase (psy1) gene from Zea mays (Zmpsy1), the carotene desaturase I (crtI) gene from Pantoea ananatis, and the phosphomannose isomerase (pmi) gene from Escherichia coli as a selectable marker. GR2E rice is intended to complement existing efforts to mitigate vitamin A deficiency by supplying consumers in societies whose diet is primarily rice-based with a portion of the estimated average requirement for vitamin
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> The safety assessment of GR2E rice evaluated information on the history of safe use of rice as a crop, the source of donor genes introduced into GR2E rice, the molecular characterization of the modified plant, the stability of the inserted genetic elements, characterization of new proteins produced in the modified plant and their expression levels, the potential allergenicity and potential toxicity of the newly expressed proteins, and the nutrient composition of GR2E rice compared to conventional rice.
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> Molecular characterization of the introduced DNA within event GR2E confirmed the presence at a single insertion site of one copy of the inserted DNA that was stably inherited over multiple generations as a single genetic locus per Mendelian rules of inheritance. Expression of the ZmPSY1 and CRTI proteins was limited to the rice endosperm with maximum concentrations in mature grain of approximately 0.245 and 0.03 ppm, respectively. The PMI protein was

expressed in all rice tissues measured and accumulated to maximum concentrations of 1.89 and 0.796 ppm in mature grain and straw, respectively.

A tiered “weight-of-evidence” approach was followed in assessing the safety of the ZmPSY1, CRTI, and PMI proteins expressed in GR2E rice. The ZmPSY1 and CRTI proteins did not display significant amino acid sequence similarity with known allergens nor were there any primary sequence structural alerts for potential toxicity based on similarity searches against a database of known and putative protein toxins. Both ZmPSY1 and CRTI were rapidly and completely digested in the presence of simulated gastric fluid containing pepsin, and the enzymatic activity of both proteins was destroyed following treatment at temperatures well below those used during cooking.

Due to the non-food source of the crtI gene, acute oral toxicity testing of CRTI protein in mice was conducted as a further assurance of safety and demonstrated a lack of any observable adverse effects at a dose of 100 mg/kg body weight, which represents at least a 115,000-fold margin of exposure relative to any realistically conceivable human dietary intake from GR2E rice. Based on its presence in a wide range of food and feed stuffs derived from genetically engineered maize lines, and on the extensive history of prior regulatory reviews in the Philippines, additional characterization of the PMI protein was unnecessary.

Previously submitted safety studies reviewed in the context of other genetically engineered plant events are directly applicable to the safety assessment of PMI protein expressed in GR2E rice. The genetic modification resulting in GR2E rice was only intended to increase levels of provitamin A (primarily ?-carotene) in the rice endosperm. To confirm the intended effect and the lack of any meaningful unintended consequences of the genetic modification, compositional parameters were compared between GR2E rice and control, unmodified, rice. Compositional analyses were performed on samples of rice grain and straw obtained from PSB Rc82 rice containing event GR2E and near-isogenic control PSB Rc82 rice that was grown at four separate sites in the Philippines during 2015 and again in 2016.

The compositional assessment included analyses for proximates, fiber, and minerals in samples of straw, and analyses for proximates, minerals, vitamins, amino acids, fatty acids, vitamins, and key anti-nutrients in grain samples. Samples of processed bran derived from GR2E and control rice were also analysed for proximates, fiber, and minerals. Among the 69 compositional components that were tested for in samples of GR2E and control PSB Rc82 rice grain, and 10 components that were assessed in derived bran and straw samples, the only statistically significant difference observed from the multi-year combined-site analysis was for stearic (C18:0) acid, a minor fatty acid component, measured in grain samples (not including the intended difference in provitamin A levels). Except for ?-carotene and related carotenoids, the compositional parameters measured in samples of GR2E rice, including stearic acid, were within or similar to the range of natural variability of those components in conventional rice varieties with a history of safe consumption.

Overall, no consistent patterns emerged to suggest that biologically meaningful changes in

composition or nutritive value of the grain or straw had occurred as an unexpected, unintended consequence of the genetic modification. Collectively, the studies performed for GR2E rice have not identified potential health and safety concerns, and support the conclusion that food and/or livestock animal feed derived from provitamin A biofortified GR2E rice is as safe and nutritious as food or feed derived from conventional rice varieties."

Although it is hard to imagine that such golden grains of polished Golden Rice could be included in commercial shipments of white rice by accident, in the modern world, any such inclusion could be damaging to international trade. To prevent even such an unlikely situation, the Golden Rice regulatory data have been submitted to regulatory authorities in countries which import rice, where VAD is not a public health issue. As a result of these data submissions, Golden Rice GR2E has been confirmed as safe for use as food, in feed, and for processing by the government's regulatory authorities in Australia, Canada, New Zealand and USA. The regulatory deliberations and decisions are publicly available: Australia and New Zealand, Canada and the USA.

Because in these industrialized countries rice forms only a tiny proportion of standard diets which already contain ample sources of vitamin A, the amounts of ?-carotene in Golden Rice would have no significant additional nutritional benefit there. Comments to this effect by the US regulatory authorities were implied by anti-GMO crop opponents to be applicable also in developing countries where the dietary situation is completely different. Such implication has been rebutted by the US FDA .

The regulators in these industrialized countries concurred with Tufts University's statement issued after their investigation of the 'Chinese children' research: … Golden Rice….could significantly improve health outcomes if adopted as a dietary regimen.' Further regulatory submissions have been made, and registrations are expected, in countries where VAD is a public health problem. In the Philippines the process is not yet complete; nevertheless various government departments have already expressed their support.

GMO crops have been vilified by activist groups since the 1990s. 'Frankenstein foods' were used in a letter in the New York Times on June 16, 1992. The Daily Mail, a UK newspaper, headlined the same

phrase in February 1998 and subsequently and extensively used ‘Frankenfoods’. The ‘anti-GMO groups,’ in various guises, have been critical of Golden Rice, a GMO crop, since 2001.

Notwithstanding this opposition, all independent scientific institutions globally have determined, for many years, that there is no inherent danger to crop plants, or the human use of crops plants, or the environment from transferring genes from one organism to another, to create GMO crops, also known as genetically engineered (GE) crops, including transfer of genes between species which cannot sexually reproduce to transfer the genes ‘naturally.’

[Cornell University Alliance for Science fellow Daniel] Norero provides a list of more than 280 independent science institutions from all over the globe that have made comments endorsing the safety of the techniques of genetic modification. A particularly clear reference comes from the heart of the geography politically most opposed to GMO technology, the European Commission of the European Union:

> The main conclusion to be drawn from the efforts of more than 130 research projects, covering a period of more than 25 years of research and involving more than 500 independent research groups, is that biotechnology, and in particular GMOs, are not per se more risky than, for example, conventional plant breeding technologies.

At the time of writing, 141 Nobel Laureates, of about 290 living, have signed an open letter dated June 29, 2016, addressed to the leaders of Greenpeace, the United Nations and governments around the world calling for the campaign against Golden Rice specifically, and crops and foods improved through biotechnology in general, to cease: ‘Opposition based on emotion and dogma contradicted by data must be stopped.’ The letter also has the support of more than 13,000 other scientists and citizens.

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