Golden Rice

Biology term paper
Sarah Amman and Mayu Saphir
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1. Preface

When we were told that we would have to write a paper about Biotechnology we searched for a topic that was not only interesting to read about but also represented a personal interest. Since we both come from Asia the Golden Rice project caught our attention.
Although Golden Rice was invented years ago it is still a rather up to date topic. In 2016, 107 Nobel laureates signed a letter imploring Greenpeace to stop blocking efforts to introduce genetically modified crops, especially Golden Rice (internet source 1).
Golden Rice contains more vitamin A than normal rice. Is the first genetically engineered crop that would not only benefit the farmers but also the consumers, which include at least 3 million children who die of vitamin A deficiency every year and another 350 000 who become blind (internet source 2).
In our paper we will look at how Dr. Ingo Potrykus and other researchers sought to achieve this by genetically engineering rice to synthesize ß-carotene and what potentials lie in Golden Rice.

2. Introduction

Golden Rice is genetically engineered to synthesize ß-carotene which is a precursor of the essential vitamin A. The aim is thus to help to prevent vitamin A deficiency (VAD).
VAD is responsible for 1-2 million deaths worldwide affecting 122 countries. Pregnant women and children are at highest risk (internet source 5).
Rice serves as a staple food for more than half of the world's population. In Asian countries it accounts for 30-70% of the energy intake of the people (internet source 1).
The cooperation between Ingo Potrykus and Peter Beyer resulted in the development of the so called Golden Rice. It is produced to synthesize β-carotene, a precursor of the essential vitamin A. This is achieved by genetically editing the carotenoid biosynthesis pathway in rice. This was eventually achieved using agrobacterium to insert selective genes.

3. Description of the engineering technique used

There are two common ways to create transgenic plants, either with the so-called gene gun method or with the agrobacteria method. In this case the agrobacteria method proved to be successful.

Agrobacterium tumefaciens are a species of soil dwelling bacteria. They have a so-called tumor-inducing plasmid, short Ti-plasmid, which can transfer DNA into a plant. The transferred DNA is called t-DNA. With genetic engineering it is then possible to modify the Ti-plasmid and the t-DNA so that no bacterial DNA is transferred but only the genes of interest (internet source 3).

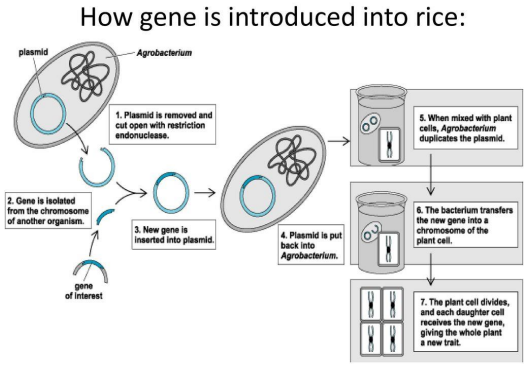

Figure 1: How the new gene is introduced into the rice

First the gene of interest is isolated from the donor organism and the unwanted bacterial genes are removed from the plasmid. Restriction endonucleases are used to make the cuts. After the genes are inserted into the plasmid the plasmid is put back into the Agrobacterium and then mixed with the plant cells. The Agrobacterium duplicates the plasmid. The bacterium then transfers the gene into the chromosome of the plant cell. The plant cell then divides, giving each daughter cell the new genes, thus giving the whole plant the wanted trait. The genes inserted in this case were *psy* (phytoene synthase) from daffodil and *crtI* (carotene desaturase) from the soil bacterium *Erwinia uredovora.* They were inserted together with a marker gene and placed under control of an endosperm specific promotor so that they would be expressed only in the endosperm. The end product of this engineered pathway is lycopene, which is processed into β-carotene in the endosperm. The β-carotene is what gives the rice the yellow color (internet source 5).

4. Interviews

4.1 With Dr. Johannes Wirz, Institute for Contextual Science

Thank you very much for taking the time to meet with me.

What is your current occupation?

JW: My current occupation: I have two jobs. One is here at the Goetheanum for 3 days a week. The other is at Mellifera e.V., a non-profit organization in Germany. It is an association for respectful beekeeping in Rosenfeld. So honey bees have become a big issue for me here as well as in Germany.

Have you worked with genetically modified organisms before?

JW: Yes. I did a PhD in molecular genetics and I finished this dissertation in 1987 and I have been manipulating genes myself.

What were the editing methods that you used?

JW: Well in those times there were no editing methods. In bacteria you had plasmids, you could add foreign sequences for instance to produce a protein. This was what I did. A protein from the drosophila fly had been producing [a protein in] bacteria. With this protein I generated antibodies. And with the antibodies I was looking into embryos to see where and when this protein was expressed in the course of development. And for drosophila, the fly itself, there have been a very special transformation technique with so-called P-element. It is like a transposal, a jumping gene, that you could add foreign sequence into it, then inject it into embryos and the embryos took it over into the germ cells.

So you worked with animals and not with plants?

JW: Yes. Although maybe I should say. Then I came here and one of our biggest projects was to investigate non-target effects of a genetic modification in crop plants.

What were the goals and what was achieved?

JW: I will first talk about the project at the university. There the goal was to understand embryological processes on a molecular level. To give you an example: nature sometimes creates flies that instead of antennas have legs growing out of the head. And so – already Goethe said – you must look at monsters, be it plant or animal, and from the monsters you can learn something about natural processes. And so in the lab it was the intention to understand why instead of antennas legs have been growing. So this is what I did at the university. And now with this work on genetically modified crop plants here we didn't want to know whether the genetical modification works (actually it never did - it works fine in the lab). So, we had three crop plants: wheat, tomatoes and potatoes. The potatoes were genetically modified to fight late blight, a disease that killed potatoes in Ireland 100 years ago.

In wheat, colleagues from the University of Zürich wanted to create a resistance against leaf rust, this is a disease in wheat crops. And tomatoes, this had been planned by a friend in Amsterdam. He just added marker genes. Genes that could be activated, and when they were active you could see a color or a fluorescent. And with this project we wanted to show – and actually we did – that if you put in a single foreign gene into a plant the whole plant is changing. Imagine you have a book with 3000 pages: You add 1 sentence and the content of the story is different. And we could see that. That this manipulation had an effect on the growth dynamics, on leaf morphology, on flower morphology and on the fruit. If you want you can download this article from our web page.

Are you familiar with the Golden Rice project?
JW: Yes, yes I'm very familiar with it.

What is in your opinion interesting and what could be problematic in this project?
JW: I think it is interesting to see, that indeed you can introduce the gene for pro vitamin A into plants, into rice. And pro vitamin A is accumulating in the seed or in the grain of rice. This is interesting. Problematic for me is that every single plant is producing pro vitamin A. You just have to eat leaves or whatsoever from plants. So it is like in former times. This method was called silver bullet approach: You shoot and you get a deer. Maybe it would be better to look into the context and see what the people need. So this is one thing, and the other thing is more on the level of social questions. When Potrycus developed Golden Rice he used methods, tools and sequences that were protected by more than 30 patents. And so of course they said small scale farmers will get the rice for free. But when does a small scale farmer become a middle scale farmer? And at this point farmers would have been pushed to buy and to pay for this genetically modified rice.

In your opinion should one integrate it into agriculture or not?
JW: Genetic engineering in general?

In this case specifically Golden Rice
JW: No. It should not be integrated. It's the wrong approach.

And genetically modified crops in general?
JW: It is again for me the wrong approach. Because now I want to make a statement. 90% of all the genetically modified crop plants have a tolerance for herbicides, Roundup or glyphosate. And the problem with weeds in farming is a problem of farming practice. And again I think it is useless to kill all the weeds and not to change the agricultural practice. And by the way soils in the US are dead. No they are really dead. If you find the statistics, look how the yield in arable land is going down.

How do you think the future of GMOs will look like?
JW: Well, as I said farming must do without. I can imagine that in environmental sciences they could be helpful. Maybe microorganisms in a really contained system to get rid of certain toxic substances and in human medicine this technology has been established and it will develop further and further. And I think this makes sense for me. To provide therapeutics for particular diseases and ailments.

Anything additional you would like to add?
And I must tell you when Ingo Potrykus has been one of the professors during my studies. And I liked him a lot because he was not arrogant, he was honest etc., and when Golden Rice came I believed naively that it would be one of the best things to use genetic modification. And then in a next step I went into the internet and I was looking for opinions or for criticisms from NGOs and there it became so evident that this Golden Rice was somehow disguising a deeper problem with nutrition. And I just really would like to ask what kind of a lifestyle it is if you depend exclusively on rice. If you do not have the possibility to eat vegetables and if you do not even have enough fat, and you need fatty acids in order that your organism can metabolize pro vitamin A. So I think something is basically wrong.

4.2 With Dr. Devang Mehta, Department of Biology ETH Zürich

What is your current occupation?
I am a post-doctoral researcher in plant genomics and biotechnology. I am currently at ETH Zurich but will soon be taking a position at the University of Alberta in Canada.

In which way have you worked with genetically modified organisms?
I have worked with GMO bacteria for my Masters (Imperial College London) where I tried to engineer bacteria that could detect the presence of human parasites in water samples. For the last 4.5 years I have been working with GMO plants for my PhD at ETH Zurich.

Did you work on animals and/or plants?
I have worked mainly with plants.

What were the editing methods you used?
I have used RNA silencing and CRISPR-Cas9 in plants.

What were the goals and what was achieved?
The goal was to study the application of these technologies to engineer cassava (Maniok) plants that are resistant to DNA-viruses. We found that RNA silencing in particular was very effective against target viruses. This experiment was conducted in a field trial of GMO plants in Kenya, together with a Kenyan partner university.

Are you familiar with the Golden Rice project?
Yes, Golden Rice was first created in the institute from which I got my PhD.

If yes, what in your opinion is interesting and what could be problematic?
I think Golden Rice was firstly, an important technical advance. Secondly, it showed that it was possible to create a GMO for a public health issue, and to do this in a university rather than a company. The product itself is also impressive since it is able to accumulate a lot of provitamin A. I do not think there is anything problematic with the project apart from the fact that it's release has been delayed far too much.

Should one in your opinion integrate Golden Rice into agriculture? Why?
Yes. For the past hundred years, breeders of crops like rice have mainly focused on yield over improving the nutrient content of plants. As a result most of our staple foods do not provide essential micronutrients at the necessary levels. Golden rice is an attempt to radically change this by producing rice varieties that have high levels of provitamin A, an essential micronutrient that is deficient in most rice-eating populations. I think it's also a cheaper way to fortify diets of people with vitamin A deficiency than the other options, which are pharmaceutical supplements that are both more expensive and not easy to distribute.

If yes, how should it be labeled?
I am not convinced that it should be labeled. For one, it's color is so distinctive! But seriously, I think food labels are very confusing. They are important for safety purposes, for e.g. to label toxins or calories, or allergenic ingredients like nuts. I do not see much value in labelling food with the type of breeding method used to produce it. However, if the population of a certain country wants to label foods with their breeding method, this should be done for all crops, not just GMOs like golden rice. Many of the plants grown today (even by organic farmers) are not GMOs but were still modified using radioactivity in the 1960s. These should also be labelled. For GMOs, I prefer using the term genetically engineered or bioengineered for labels since they are more scientifically accurate.

What are your ethical concerns regarding Golden Rice and/or GM crops in general?
I do not have any ethical concerns regarding Golden Rice or GMOs specifically.
I do have many concerns about how GMOs are treated by NGOs in Europe, who tend to conflate the science with their world-views about seed companies like Monsanto and Syngenta. These are two different issues and should be treated as such.
For my PhD, I worked on making GMO virus-resistant cassava which would be patent-free and available to small-holder farmers for free. African scientists have also created their own GMO bananas for their farmers. All of these projects have been publicly funded, without the involvement of private industries. However due to the misinformation spread by European-funded NGOs in Africa, many countries there have banned GMO cultivation, even if the GMO in question was developed by their publicly-funded scientists. To me, this is appalling. It is, in my opinion, wrong to prevent the use of safe, effective technology due to misinformation.

What do you think the future of GM crops will look like in 10 years?
I am optimistic for the future of GM crops in countries that currently allow their cultivation like Canada, USA, South Africa. GM crops have been found to reduce pesticide use by 37%, and reduce CO2 emissions in to the atmosphere and increase farmer profits. There are new GM crops being made right now in labs around the world that are able to do photosynthesis more efficiently, able to grow well with less water, produce and store more nutrients (like vitamin B6/B9 and iron) etc. I think we will also see a change in public perception of GMOs as more products with direct benefits for consumers make it to the market. One example, is the Impossible Burger, a meat-free burger made from genetically engineered plant proteins that is now very successful in the USA. (I've tried it and it's better than a 'real' burger!) However, I am also pessimistic that any of these fantastic products will ever be sold in Europe.

5. Discussion

The original aim of Golden Rice was to give it to third world countries for free. The inventor thought they could patent the rice and then do whatever they want with it. This proved to be a wrong assumption since 70 different trade mark rights and 32 companies and universities were involved in the invention. After time-consuming negotiations they were able to give the rice to local research institutes. Golden rice fulfils all requirements for an "ideal" transgene plant.

- It wasn't developed by industry
- It doesn't cause any high costs for the farmer and the farmers can continue developing their seeds themselves.
- The farmers don't become dependent
- The rice doesn't require any specific pesticides or fertilizers
- No negative impact for either humans or environment are known.

Although Golden Rice is supported by multiple humanitarian organizations there are a lot of NGOs which are vehemently against the use of Golden Rice. They fear that it could serve as a Trojan horse for GMOs in the agriculture in third world countries. One of the main arguments against Golden Rice is that the amount of β-carotene isn't high enough. They also worry that other methods to fight vitamin A deficiency are getting neglected. For instance it is cheaper and more sustainable to teach people to each enough vegetables to cover the required amount of vitamin A (internet source 4).

5.1 Alternative methods to fight vitamin A deficiency

The most important strategies are a varied diet, food supplements and the distribution of vitamin A pills. These pills can for instance be distributed as a part of vaccination programs. Since vitamin A is stored in the liver one pill in 4-6 months suffice. However, such programs usually only reach part of the population and are dependent on outside financing. Adding vitamin A in food is an approved way of fighting VAD. The biggest problems with this method are usually political. Sometimes laws have to be adjusted and long-term financing is necessary. An additional problem is that the food with added vitamin has to be consumed by the affected population. Without genetic engineering it is not possible to enrich rice with vitamin A. The methods vary according to the country (internet source 4).

5.1 Future of GMOS

The future of genetically modified crops lies in CHRISPR-Cas. Projects like waxy corn are already in progress. One hope is that with CHRISPR there will be a new chance to introduce GMOs and to avoid the sort of public backlash that rocked Monsanto in the late 1990s and still plagues agriculture two decades later. Since Monsanto blotched the introduction of GMOs the term GMOs has been treated like a dirty word. Great efforts have been made to clean up its image in recent years.

CHRISPR unlike other conventional genetic engineering methods works directly on the DNA of the plant or animal being bred. GMOs as we know them involve inserting target DNA from a different species (internet source 6).

6. Summary

Golden Rice was originally developed by Ingo Potrykus. It is genetically engineered to synthesize β-carotene. This was achieved by inserting two selected genes.
Both interviewed researchers state that genetic engineering is an interesting technique. As to the applications in agriculture their opinions vary. Dr. Mehta seems to believe in a silver bullet approach while Dr. Wirz has a more differentiated view on the issue and seems more familiar with broader implications that defy a simple answer.
Researching our topic and conducting the interview with Dr. Wirz made us aware that the issues surrounding Golden Rice are much more complex than we imagined when we began. In particular the interplay between research, patent law, nutrition, agriculture and economics defies simple answers. We do hope that a viable and sustainable solution to VAD will be found in the future.

7. References

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