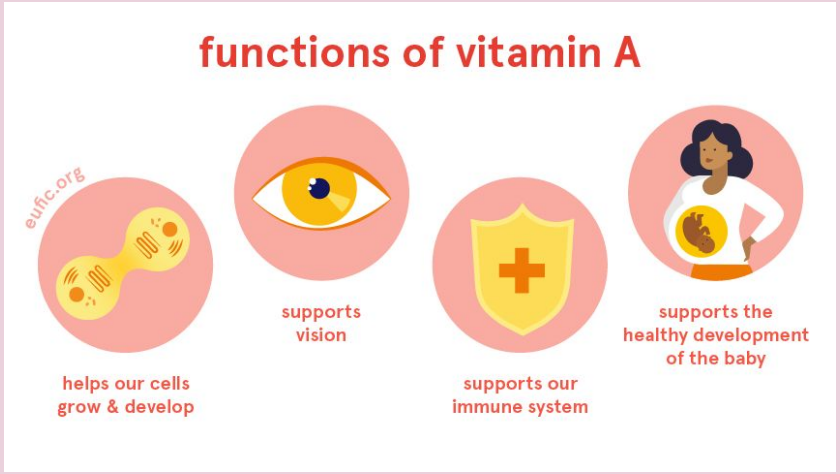
Golden Rice

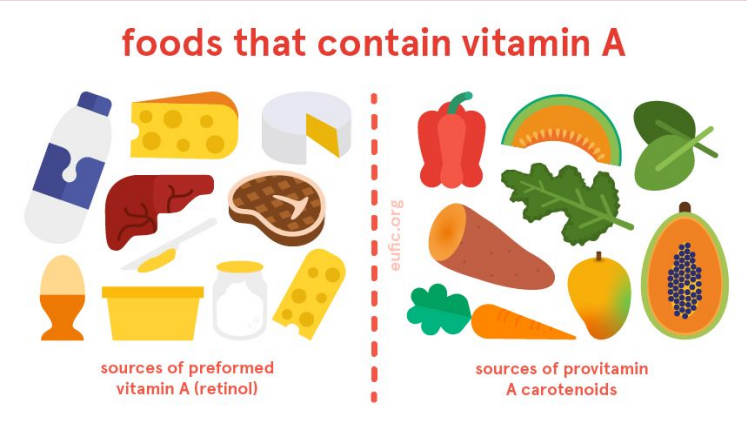
Project by Emily Mahon, Jessica Hall,
Cashe Corbett

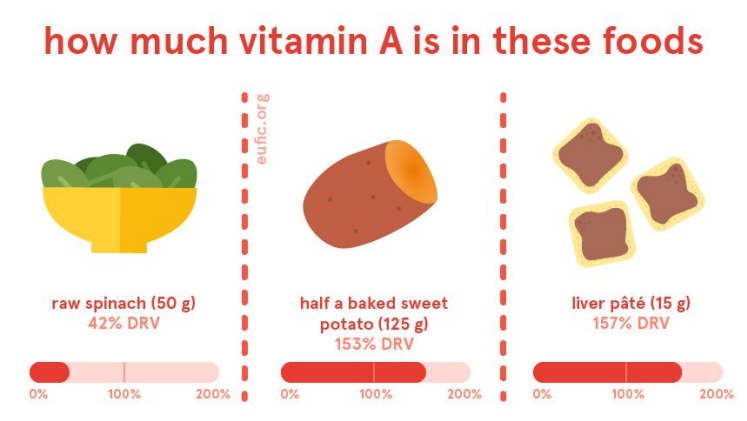
What is Vitamin A

- ★ Fat soluble essential nutrient
  - Carotenoids (plant-based)
    - Beta-carotene → active Vitamin A
  - Retinol (animal-based)
- ★ Multiple functions
  - Vision
  - Immune function
  - Skin health
  - Cell growth
- ★ Antioxidant properties
  - Cancer defence (controversial)
- ★ Issues arise with too little or too much



Where to get Vitamin A





★ Found in many foods
- Spinach
- Eggs
- Dairy products
- Yellow or orange vegetables
- Normally easy to reach required amount

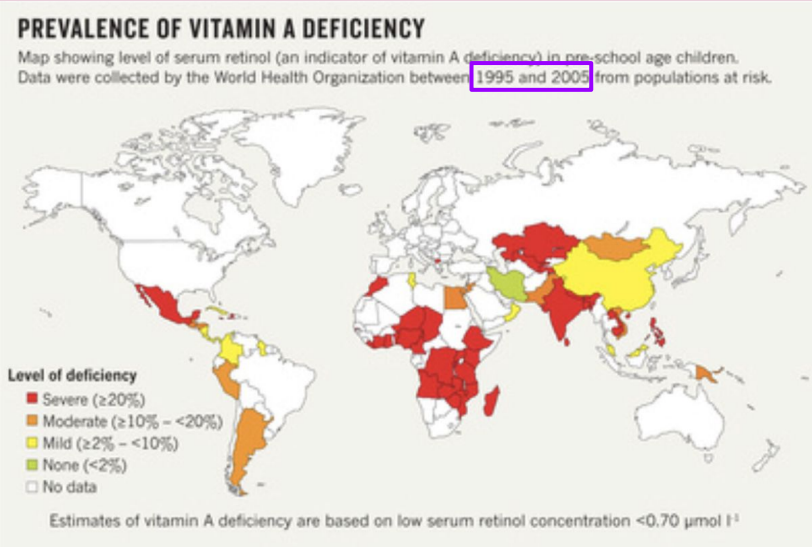
★ Issue arises in lower income countries
- Less access to variety of fresh foods/animal products
- Cystic fibrosis, pregnancy and iron deficiency can worsen the deficiency

Global Issues of World Hunger

- ★ 1 in 11 people worldwide face hunger
  - 1 in 5 in Africa
  - 733 million people
  - ~1/2 of deaths <5 years old linked to undernutrition
- ★ Lack of economic access to healthy diets
  - Affects 1/3 of global population
- ★ Most important in global public health terms:
  - Deficiencies representing major threat to health and development
    - Iodine, **vitamin A,** iron

Why we needed this project

- ★ Severe deficiencies
  - Central Africa, India, Kazakhstan, Mexico
- ★ Moderate deficiencies
  - Argentina, Peru, Mongolia
- ★ Many health impacts due to deficiency
  - Anemia in individuals with low iron
  - Increased risk of birth defects
  - Xerophthalmia
    - Night blindness
    - Severe dryness
    - Spots in the whites of eye
    - Full blindness

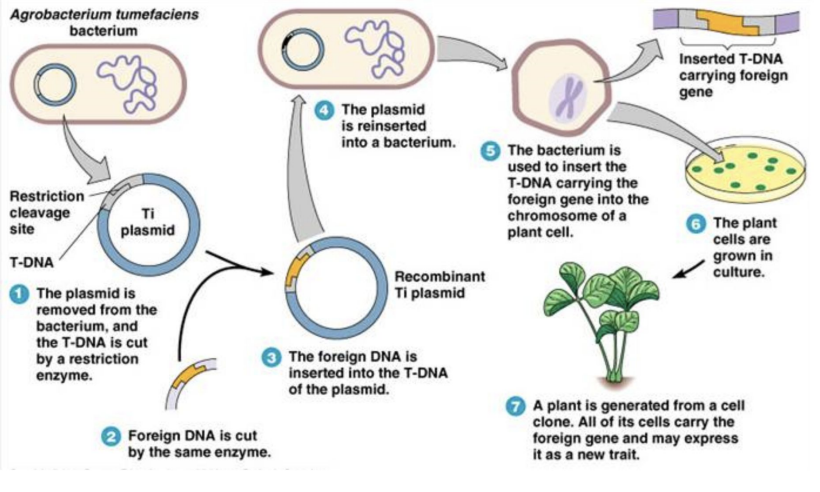


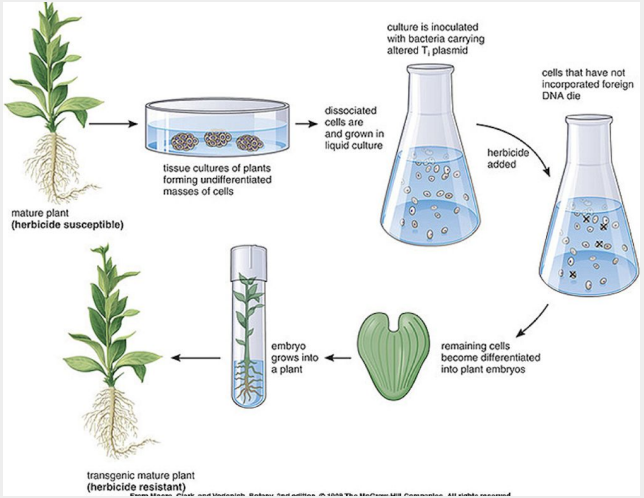
Global Issues of World Hunger

- ★ Zero hunger project- set back 15 years with increasing hunger rates
  - Covid pandemic caused sharp upturn in hunger numbers
  - Increasing food prices
  - Conflict and climate change
- ★ Global malnutrition still a major concern

What is a GMO?

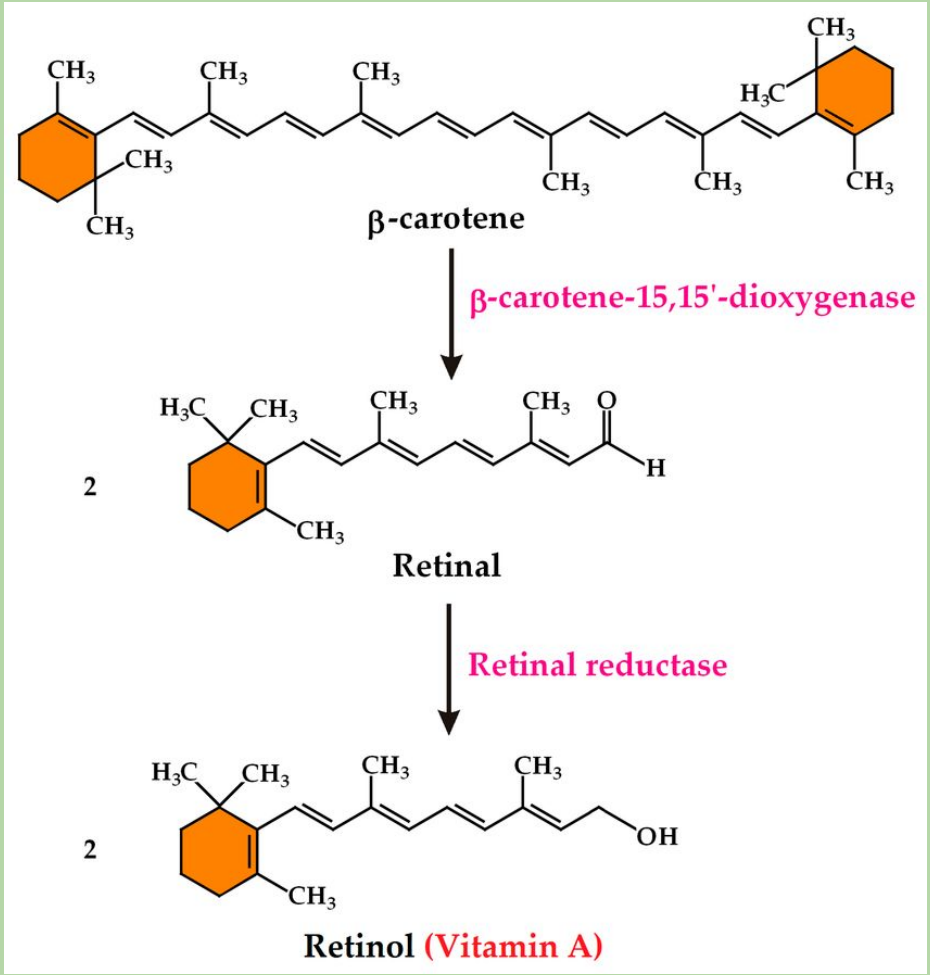
- ★ Genetically modified organism
  - Laboratory procedure
  - Alter genes, introduce desirable traits, delete unfavorable ones
  - Modifications normally unattainable via natural or selective breeding
- ★ Used in agriculture, medicine, and research
  - Plant, animal, or microbe
- ★ Used in Golden Rice project

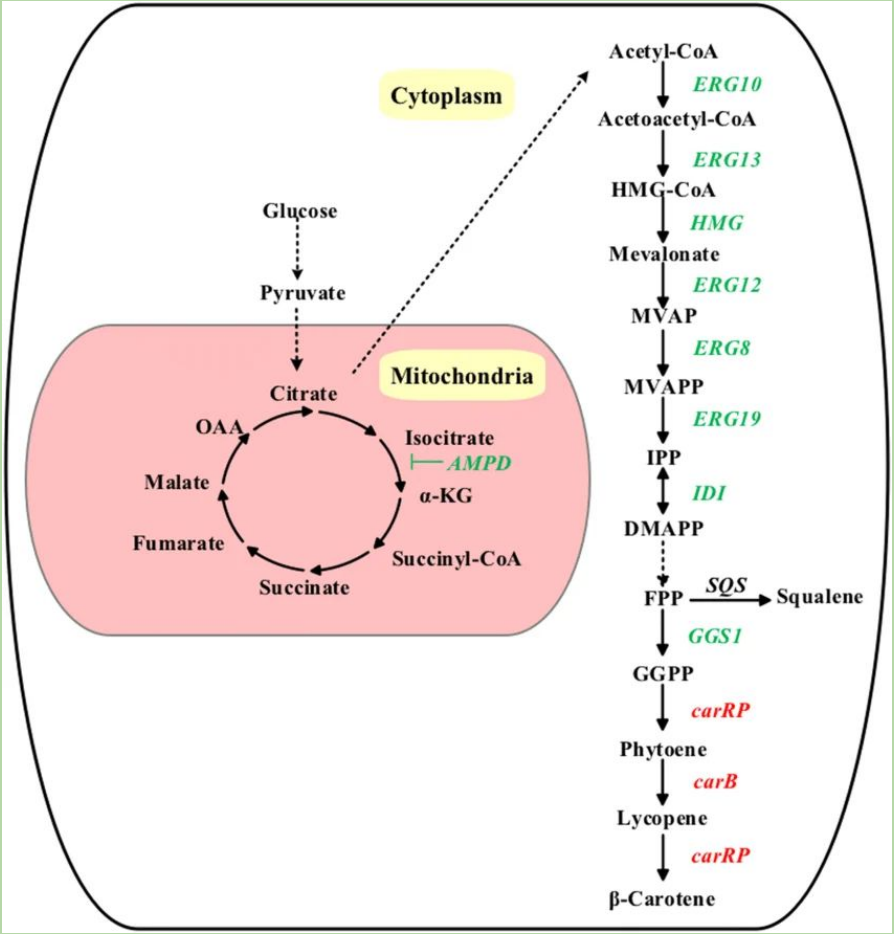




β-Carotene

- ★ Member of the **carotenoids**
- ★ Orange pigment found in plants
  - Responsible for "**golden**" **colour** of golden rice
- ★ **Provitamin A** molecule
  - **Converted** into **vitamin A** (retinol) in the body
  - **Symmetrical** structure with **2** terminal **β-ionone rings** allows production of **2 vitamin A** molecules





β-Carotene Pathway in Rice Endosperm

- ★ Rice plant **contains necessary machinery** to produce **β-carotene**
- ★ **Leaves** ➡ machinery **active** ➡ **β-carotene** production and **accumulation**
- ★ **Endosperm** ➡ some enzymes **inactive** ➡ **no β-carotene accumulation**

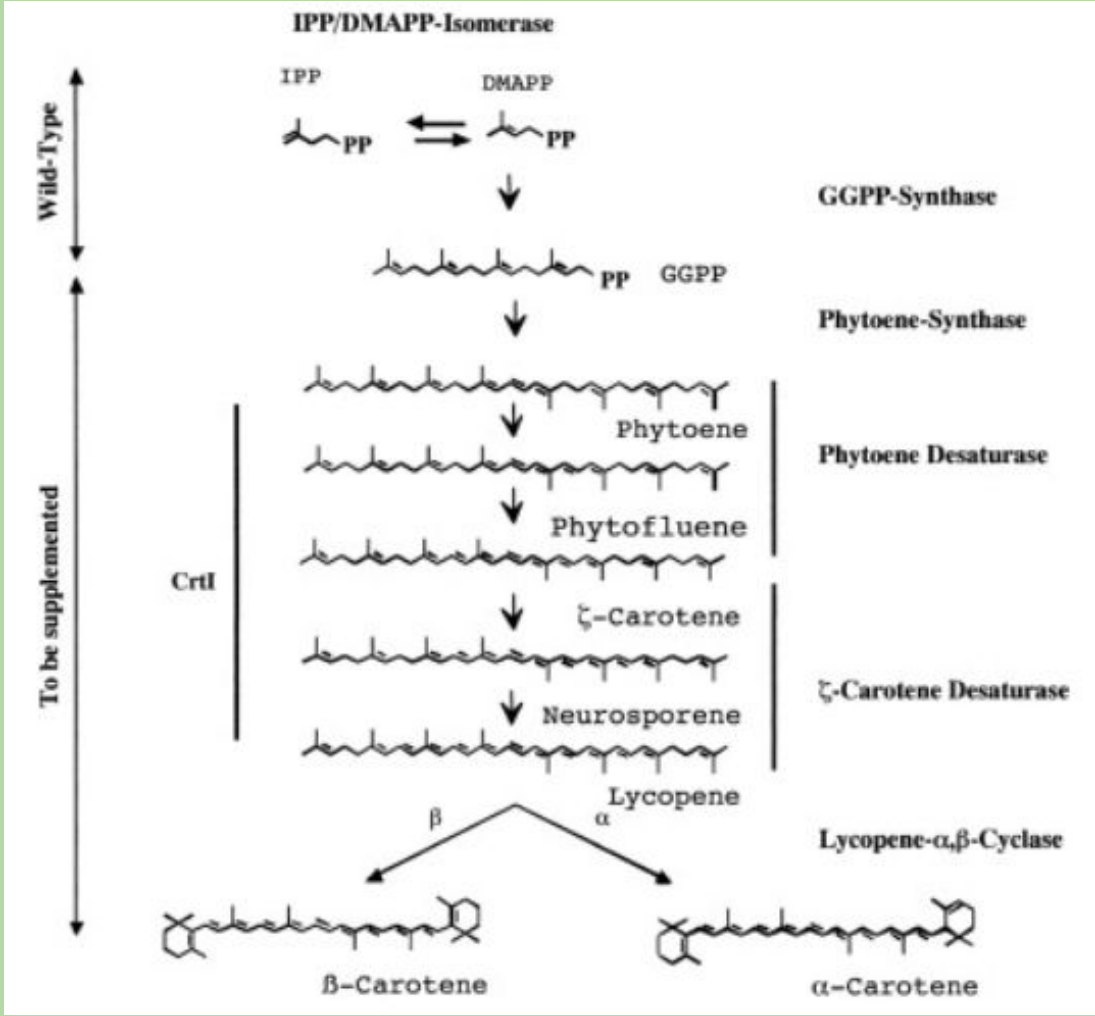
AIM: **Introduce** the **minimal** number of genes sufficient to **complete** the **β-carotene pathway**

Missing Enzymes

(1) GGPP ➡ Phytoene
- Via **Phytoene Synthase**

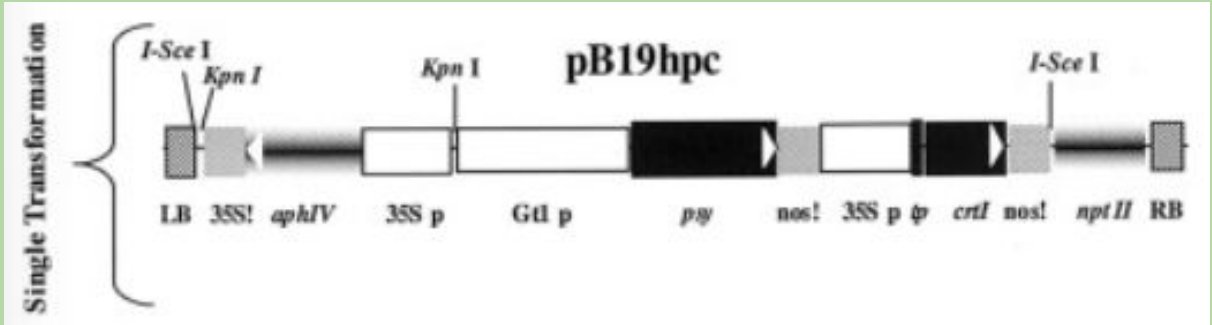
(2) Phytoene ➡ Lycopene
- Via **Phytoene Desaturase & ξ-Carotene Desaturase**

(3) Lycopene ➡ β-carotene
- Via **Lycopene-α,β-Cyclase**



Agrobacterium-Mediated Transformation
Single Transformation

- ★ Transformation of precultured rice **immature embryos**
- ★ **Single transformation**:
  - **pB19hpc** vector
    - Plant **phytoene synthase** (psy)
    - Bacterial **phytoene desaturase** (crtI)
    - Selectable marker (aphIV) - **hygromycin resistance**
- ★ Produced grains with **β-carotene** and to some extent, **lutein** and **zeaxanthin** (deficiency leads to **macular degeneration**)



pB19hpc - SIngle Transformation

(1) Plant **phytoene synthase** (**psy**)
- Originating from **daffodil** (Narcissus *pseudonarcissus*)
- Under control of the **endosperm-specific glutelin** (Gt1) promoter
- cDNA previously shown to contain **5'-sequence** coding for a functional **transit peptide**

(2) Bacterial **phytoene desaturase** (**crtI**)
- Originating from **Erwinia** ***uredovora***
- Under control of the constitutive **CaMV 35S** promoter
- **Fused** to **transit peptide** sequence of the **pea Rubisco small subunit** (tp)

★ Transit peptides direct the enzymes to the **plastids** where **GGPP** is formed

Agrobacterium-Mediated Transformation
Cotransformation

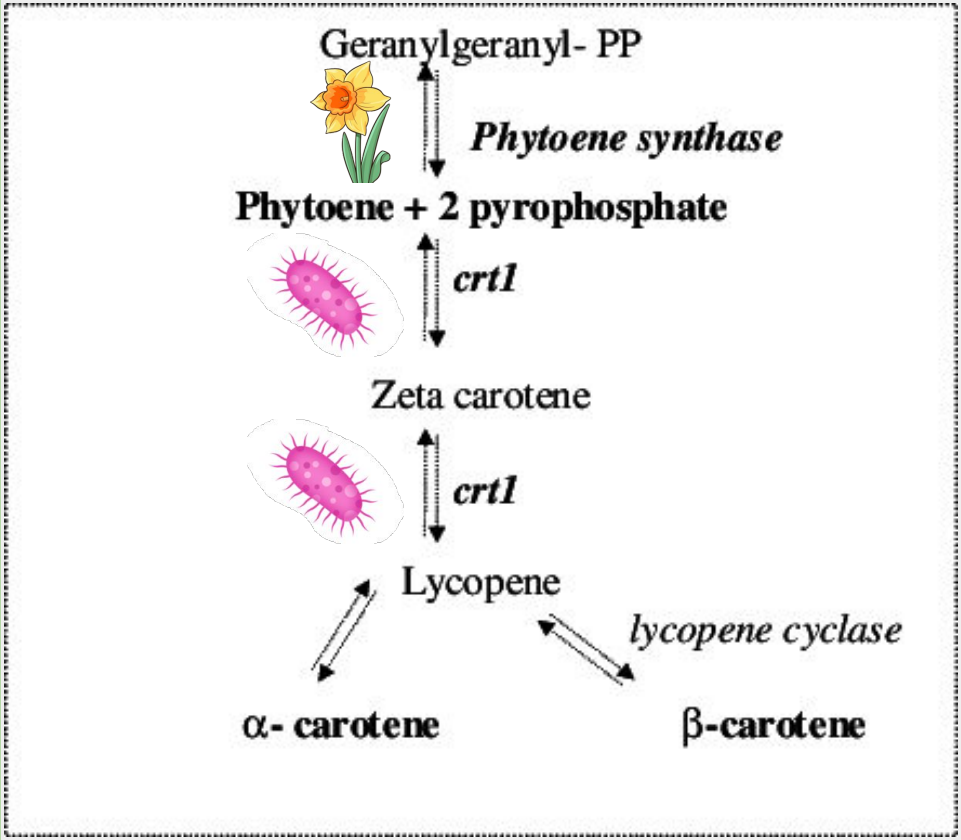
- ★ Transformation of precultured rice **immature embryos**
- ★ **Cotransformation:**
  - **pZPsC** vector
    - Plant **phytoene synthase** (psy)
    - Bacterial **phytoene desaturase** (crtI)
  - **pZCycH** vector:
    - Plant **lycopene β-cyclase** (lcy)
    - Selectable marker (aphIV) - **hygromycin resistance**
- ★ Produced rice with β-carotene as almost the exclusive carotenoid

Conclusion: cotransformation is not necessary

★ Psy and crtI are sufficient to activate β-carotene production

★ Single transformation permits the production of other essential carotenoids

Transformed β-Carotene Pathway



Golden Rice Controversies - Regulatory Approval

➔ Safety approvals by national regulators
- Australia, NZ, Canada, U.S., Philippines, Bangladesh?

➔ Long approval time & very costly
- 1st developed: 1999
- 1st safety approval: 2017 (Australia - NZ)

➔ **Philippines** regulatory process
- Food & feed approval: 2019
- 1st to approve commercial propagation of GR: 2021
- **Revoked** approval: 2024

Golden Rice Controversies - Public

Greenpeace: environmental advocacy org. known for opposition to GM crops

➔ **Nutritional impact** not fully understood / *unknown*
- Ineffective in treating VAD: insufficient beta-carotene levels

➔ **Environmental impact**
- “Opening the door”
- Risk to environment (cross-pollination): threaten biodiversity
- Diverse diet - no need to GM

★ Local anti-GMO groups
- Destroyed a field of GR

Golden Rice Controversies - Counter-Arguments

Counter-arguments to **Greenpeace** (& others) claims:

1) Studies prove GR effectively addresses VA daily requirements (⇧ total intake)
2) No proven studies show harm from GR; approved in multiple countries
3) Studies prove rice primarily self-pollinating → cross-pollination % very low
4) Purpose of GR → developing countries with limited access and/or poor

➢ **Philippine cease and desist:** *speculation* - NOT scientific proof

Defeating their own purpose?

GREENPEACE

GMO controversies - General Debates

- ➔ **Environmental** concerns
  - "**Superweeds**" : Herbicide - resistant weeds
  - "**Superpests**" : Pesticide - resistant pests
  - Biodiversity loss on non-targets
- ➔ **Human health** concerns
  - Long-term effects *unknown*
- ➔ **Ethical** concerns
  - Corporate control of seeds
  - Patents on GMOs

Other GMOs (reported from US)

- ➔ **Corn** ~ 92%
- ➔ **Soybeans** ~ 94%
- ➔ **Canola** (oil) ~ 95%
- ➔ Other commons GMOs:
  - Cotton, Sugar beet, Potato, *many more*

Conclusion Question

Do YOU think the benefits of GR outweigh the potential risks?
Would you approve this if you were living in a developing country?

Benefits	**Risks Proposed**
Treat VAD in millions of (mainly) children	Low levels of beta-carotene: being ineffective (proven wrong)
Reduce blindness in children	Potential cross-pollination (0 - 1% chance)
Reduce the millions of deaths by VAD - mainly in children	Could “open the door” to other & more GMOs (already in our everyday diet)
Cost - effective	Unknown impacts of consuming GR (proven healthy and approved by many countries)
★ **No proven concerns for health or efficiency - only benefits and pro-efficiency**	

References

Beyer, P., Al-Babili, S., Ye, X., Lucca, P., Schaub, P., Welsch, R., & Potrykus, I. (2002). Golden Rice: Introducing the β-carotene biosynthesis pathway into rice endosperm by genetic engineering to defeat Vitamin A deficiency. The Journal of Nutrition, 132(3). https://doi.org/10.1093/jn/132.3.506s

European Food Information Council. (2021, January 11). *Vitamin A: Foods, functions, how much do you need & more*. https://www.eufic.org/en/vitamins-and-minerals/article/vitamin-a-foods-functions-how-much-do-you-need-and-more

Mayo Clinic Staff. (n.d.). *Vitamin A*. https://www.mayoclinic.org/drugs-supplements-vitamin-a/art-20365945

Zero Hunger Project. (n.d.). *Zero Hunger Project*. https://www.zerohungerproject.com/

World Health Organization. (2014, December 20). *Global nutrition targets 2025: Policy brief series*. https://www.who.int/publications/i/item/WHO-NMH-NHD-14.2