Biofortified orange maize is as efficacious as vitamin A supplement in Zambian children even in the presence of high liver reserves of vitamin A: a community-based, randomized placebo-controlled trial

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Current dimensions of major forms of malnutrition

Intra-uterine growth retardation
- 30 million/year (23.8% of all births)

Protein-energy malnutrition
- 159 million under-5 children

Iodine deficiency disorders
- 740 million

Vitamin A deficiency
- 190 million under-5 children

Anaemia including iron deficiency
- 2 billion

Obesity
- >300 million people

Low birth weight
Brain damage
Increased morbidity & mortality
Brain damage
Blindness
Increased morbidity & mortality
Increased morbidity & mortality
Impaired motor development
Reduced physical activity
CVD & Diabetes
Functions of vitamin A

Methods to alleviate vitamin A deficiency

- Targeted supplementation with preformed retinyl palmitate (with coverage >80%)
- Fortification of sugar (median VA concentration of 8.8 µg/g)
- Promotion of dietary diversity through inclusion of more provitamin A carotenoid sources
- Biofortification of staple crops with β-carotene and/or β-cryptoxanthin
Advantages of biofortification

- Targets the poor
- It’s rural based
- Cost effective
- Sustainable
- Negative feedback mechanism of β-carotene absorption and bioconversion to VA mitigate hypervitaminosis issues associated with preformed VA
Biofortified orange maize has shown bioefficacy in maintaining VA liver reserves in Mongolian gerbils and in providing VA in single meals to humans.
Main objective of the randomized controlled trial

- To evaluate the bioefficacy of orange maize with the use of 13C-RID test.
Good choice for Zambians
Assessing the efficacy of biofortified (orange) maize in rural Zambian children

**Timeline for Studies in Eastern Province of Zambia**

- **2009**: Population survey, serum retinol and MRDR test (Hotz et al 2012)
- **2010**: RCT with biofortified maize, serum retinol and MRDR test (Bresnahan et al 2014)
- **2012**: RCT with biofortified maize, serum retinol and isotope dilution test (Gannon et al 2014)
Hiring nutritionists
Grinding the maize
Setting up kitchens
Cooking the maize
Stirring and stirring....
If you don’t have help!
Eating!
Weigh and record all uneaten food for each subject
Cleaning up the mess
Retinol isotope dilution

$^{13}$C-retinol and
Gas chromatography-combustion-isotope ratio mass spectrometry
Isotope dilution technique to assess total body vitamin A pool size

• The isotope dilution technique provides an accurate quantitative estimate of mean vitamin A pool size for groups of subjects in a target community

• The isotope dilution technique detects quantitative changes in vitamin A pool size in response to supplementation with known amounts of vitamin A

• The paired-isotope dilution technique can be used to estimate both the impact of vitamin A interventions and the relative vitamin A equivalency of β-carotene
Principle of isotope dilution

Dose with $A^*$

Baseline

Body pool

Enriched body pool

Blood for $A^*/A$
Recruited (n = 143)
- 1 did not receive anthelminthic treatment
- 2 did not meet minimum blood draw

Randomly assigned (n = 140)
- Assigned to VA – group (n = 47)
  - 1 withdrawn by parents unwilling to have more blood drawn
  - Began intervention 46
  - Completed intervention 46
  - Completed study 45
  - Successfully analyzed 44
- Assigned to orange – group (n = 46)
  - 1 withdrawn by parents unwilling to have more blood drawn
  - Began intervention 46
  - Completed intervention 44
  - Completed study 44
  - Successfully analyzed 44
- Assigned to VA – group (n = 47)
  - 1 withdrawn by parents unwilling to have more blood drawn
  - Began intervention 46
  - 1 left study area with family
  - 1 withdrawn by parents
  - Completed intervention 46
  - Completed study 46
  - 1 sample lost due to severe hemolysis
  - Successfully analyzed 44

Completed study 46
- 1 lost in transit to lab
  - Successfully analyzed 45
The “blue” group was administered the recommended daily allowance for vitamin A, which is 400 μg retinol activity equivalents/day, six days/week. The other groups received placebo oil (214 μL/d) with or without orange maize.

Gannon et al., Am J Clin Nutr 2014
Calculation of Total Body Reserves

- Mass balance equation
  \((F_a * a) + (F_b * b) = (F_c * c)\)

- Assumptions:
  - 80-90% dose absorbed
  - 80% stored in liver
  - Catabolism of dose during equilibration

- Paired study design
  - See direct effect of intervention on each child
Daily dosing with the RDA or placebo oil
Our children grew up!

A total of 133 children were successfully enrolled and analyzed.
Changes in total body stores

$P = 0.0034$

When values are ranked

Gannon et al., Am J Clin Nutr 2014
Degree of concern for baseline status

- Scientific published finding
- Assumes 80% is stored in the liver
- 59% of the children have hypervitaminotic liver stores corrected for CRP
- Defined as 1.0 μmol/g liver

Biofortification could be a safe way to mitigate the hypervitaminosis that is likely in part caused by combined high dose supplementation and fortification efforts in the country.

Gannon et al., Am J Clin Nutr 2014
Release of results to the community

Photo credit: B. Gannon
Overall successes

• Synthetic maize was traditionally bred for the second efficacy study that had 20 μg β-carotene/g at harvest.

• Compliance was excellent in both trials >90%.

• Dietary recalls helped to displace some of the negative claims associated with the menu.

• Information sharing from the trials occurred with HarvestPlus-Zambia, district officials, and the communities that participated in the trial.

• Distribution of orange maize seed and fertilizer to the caregivers of participants added value to the trials.

• Erection of one school in the district will have long-lasting effects in the community.
Where we are at now

Photo credit: K. Pixley
Truck advertising and free samples at outreach activities

Photo credits: E. Simpungwe & N. Palacios
Summary

• The current science has demonstrated that these biofortified crops improve vitamin A intake and status of consumers or enhance circulating concentrations of β-carotene, which may have other health benefits in addition to vitamin A nutriture.

• Demand creation and nutrition messaging has made careful links with the vitamin A messages promoted by the Ministries of Health so that there is no contradiction about the benefits of vitamin A.

• Future effectiveness studies may show improvement in optimal health through healthier food choices.
Overcoming the color of the food that we eat!!!
Really?? Orange hands??

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