

Healthier rice: Achievements and Challenges

Christine Bergman, Ph.D., R.D.
Professor
University of Nevada Las Vegas
USA



Healthier Rice



- Achievements
 - Pre-harvest management
 - Post-harvest processing
 - Traditional breeding
 - Mutation breeding
 - Genetic modification



Enhance Nutritional Properties



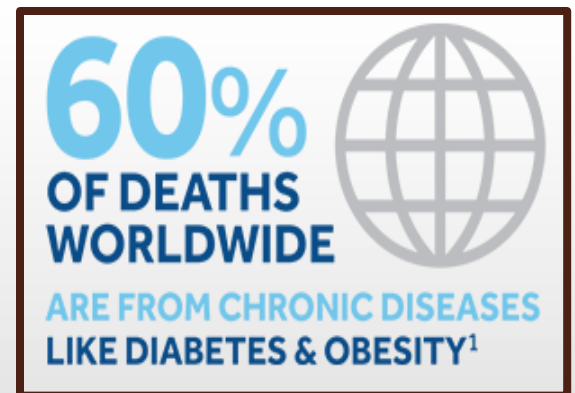
- Decrease chronic disease
- Prevent deficiency disease
- Improve well being



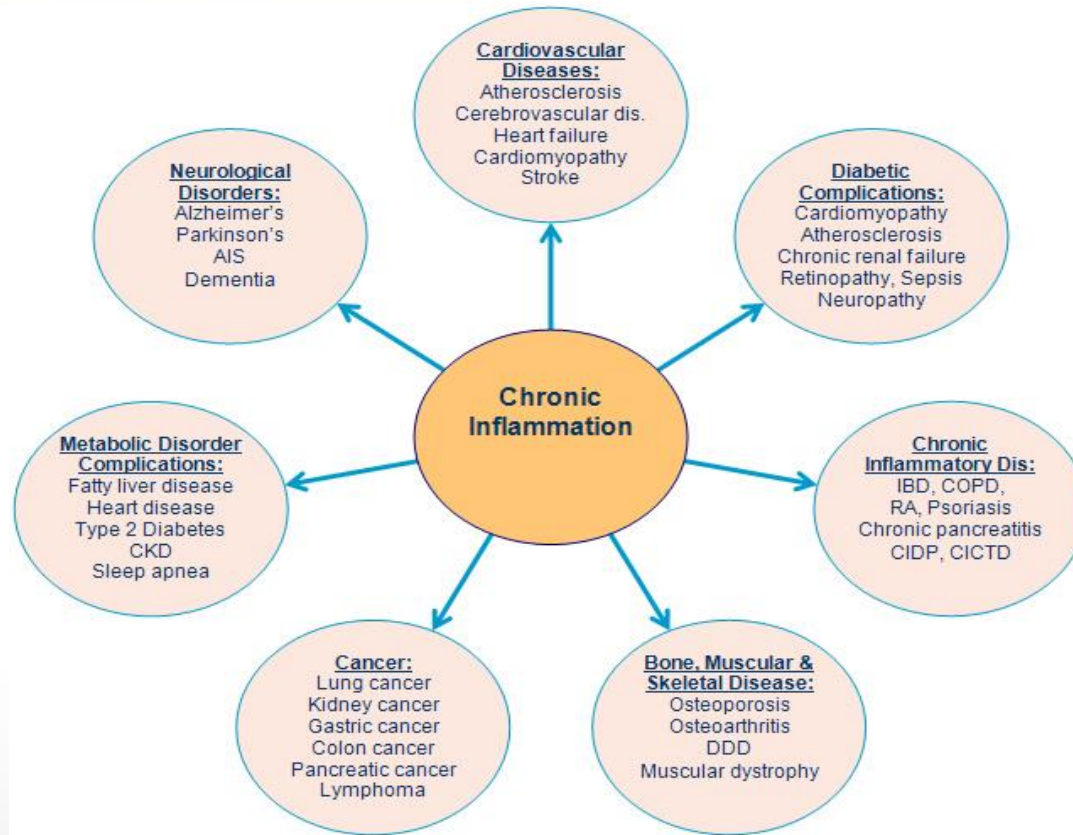
WHO



- **Chronic illnesses have reached global epidemic proportions**
 - Cancer
 - Heart disease
 - Diabetes
- **Cause MORE deaths than ALL other diseases combined including**
 - Malaria
 - HIV
 - Tuberculosis



Cause or Effect?

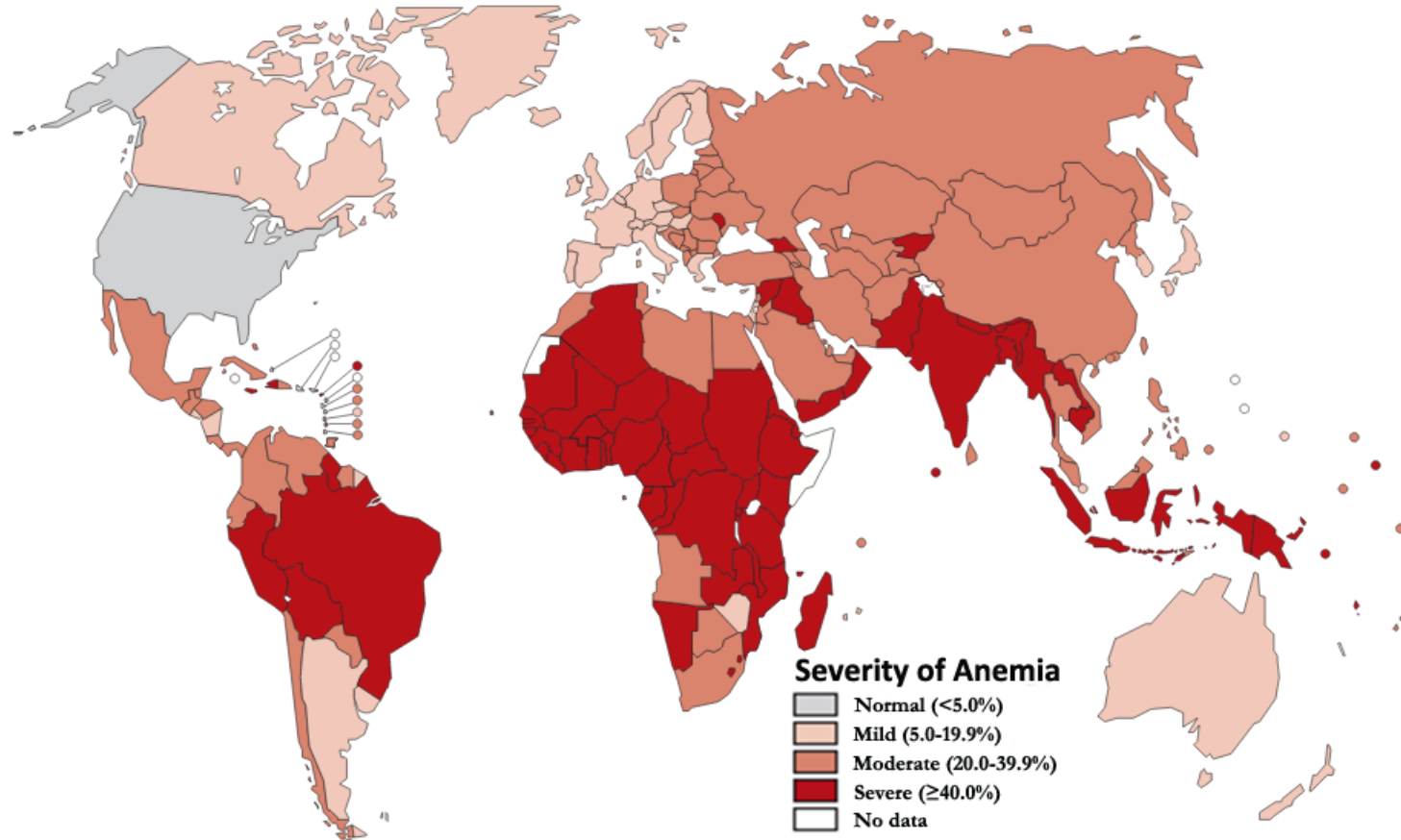


- Diet plays a role – whole grains, fruits and vegetables, fatty fish
- Egger G. 2012. In Search of a Germ Theory Equivalent for Chronic Disease. *Prev Chronic Dis* 9:11.

Healthier rice



Worldwide Prevalence of Anemia, by severity

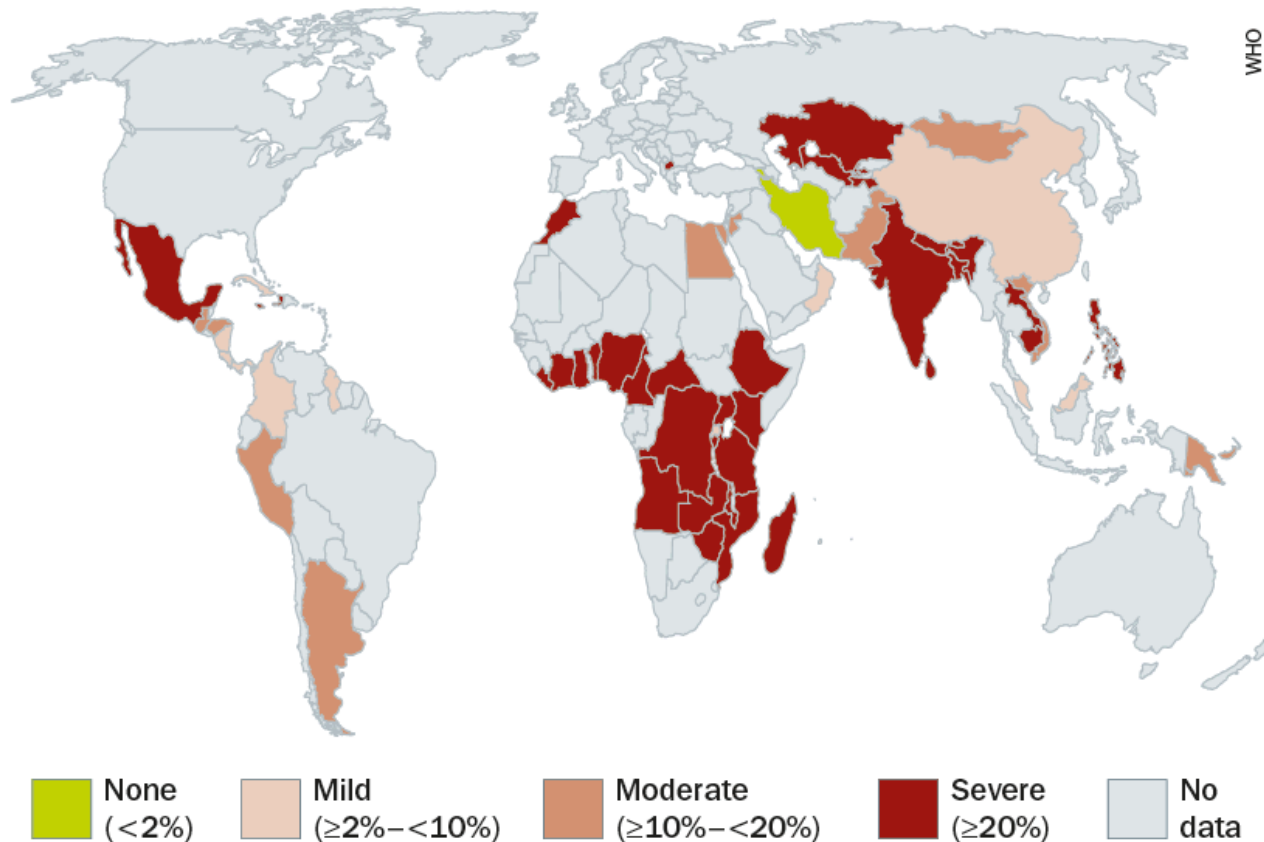


WHO global estimates of anemia prevalence (2008)

Healthier rice



Figure 1. Global extent of vitamin A deficiency as defined by prevalence of serum retinol $<0.70 \mu\text{mol/l}$ in preschool children³

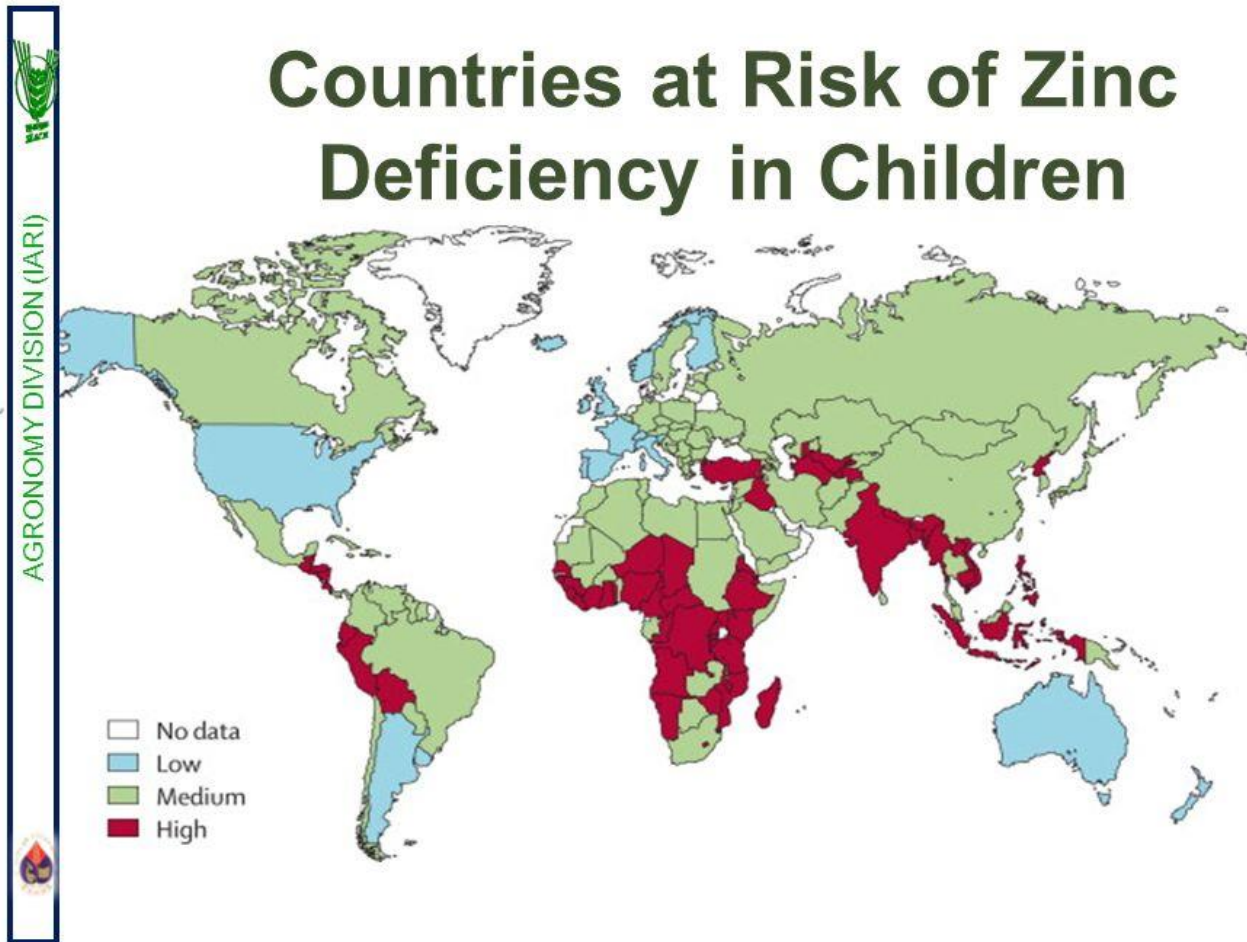


³ World Health Organization (WHO). Global prevalence of vitamin A deficiency in populations at risk 1995–2005. WHO Global Database on Vitamin A deficiency. Geneva, Switzerland: WHO; 2009. Available at: http://whqlibdoc.who.int/publications/2009/9789241598019_eng.pdf

Healthier Rice



Countries at Risk of Zinc Deficiency in Children



Black et al. 2008. The Lancet Maternal and Child Undernutrition Series

Pre-harvest Management

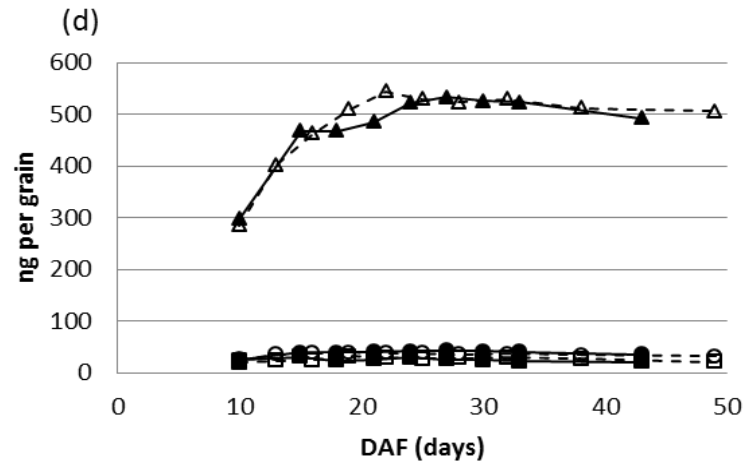
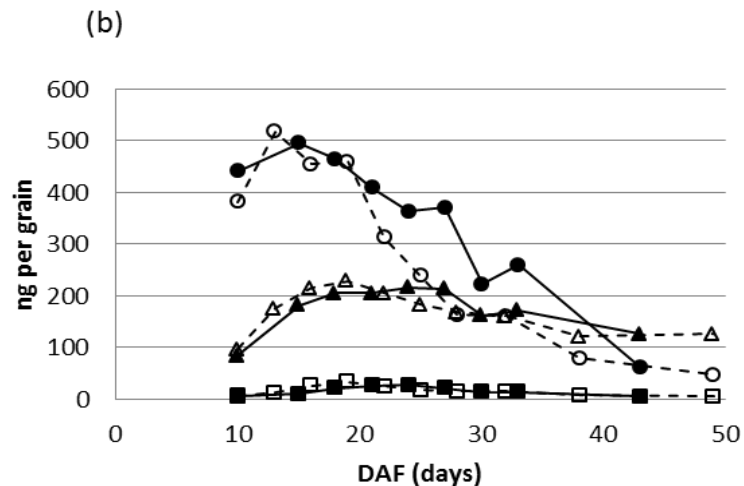
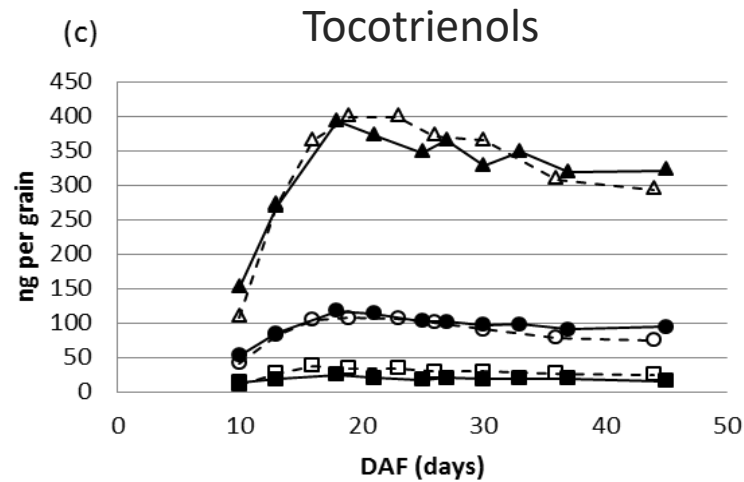
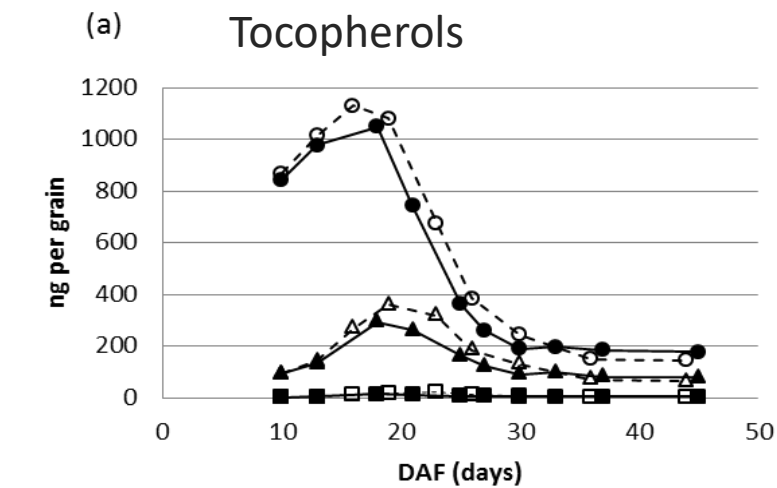


Pre-Harvest Management



| Sources of Variation | $T + T_3^a$ | Gamma-Oryzanol |
|---------------------------------------|-------------|----------------|
| Genetics (<i>Oryza sativa</i>) | 3.6 | 2.4 |
| Genetics (other <i>Oryza</i> species) | 10.1 | 5.8 |
| Growing Conditions | | |
| Seeding Rate | No effect | No effect |
| Nitrogen Application | No effect | No effect |
| Planting Date (w/ in a Field) | 1.55 | 1.28 |
| Between Years | 1.33 | 1.0 |

^a Fold variation in the trait (i.e., maximum value divided by minimum value). Amount of starch in bran used to represent milling degree or the degree to which kernels broke-up during milling, thus diluting the amount of antioxidants measured.



Contents of tocopherol and tocotrienol homologues in developing grains of Cypress (a and c, respectively) and Teqing (b and d, respectively). The filled and non-filled symbols represent planting date 1 and 2, respectively. Symbols of circles, triangles and squares are α -, γ - and δ -homologues, respectively. DAF, days after flowering.

Chen and Bergman, 2005

'Cypress'

'Teqing'

Limitations

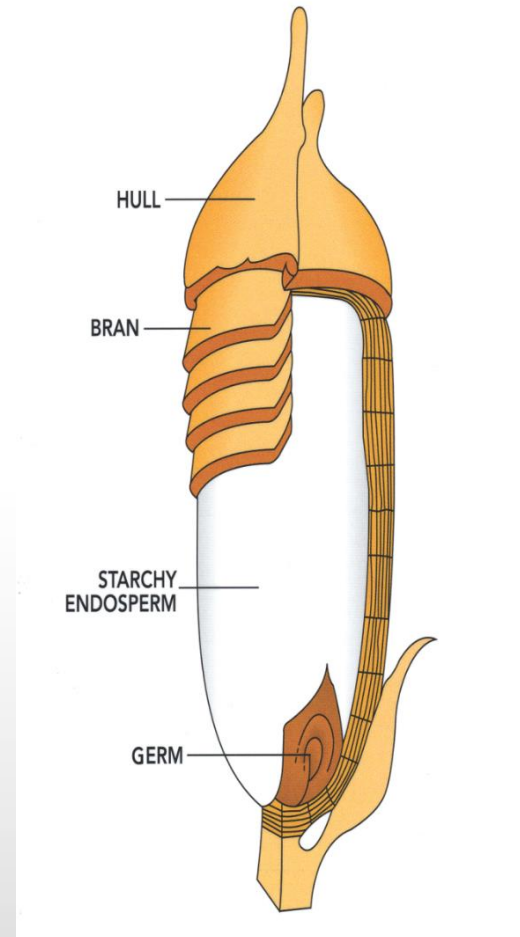


- There is more to know about pre-harvest effects on nutritional properties of rice
- e.g., organic production effects on mineral content
- Consumers are requiring more labeling information – difficult with variation coming from environmental effects
- Harvesting prior to maturity benefits yes but cons = different harvest and processing methods; costly.

Post-Harvest Management



- Not milled (brown)
- Under milled
- Pre-germinated Rice



Nutrient differences



| | Brown Rice (one cup) | White Rice (one cup) |
|-----------------|----------------------|----------------------|
| Calories | 232 | 223 |
| Protein | 4.88 g | 4.10 g |
| Carbohydrate | 49.7 g | 49.6 g |
| Fat | 1.17 g | 0.205 g |
| Dietary Fiber | 3.32 g | 0.74 g |
| Thiamin (B1) | 0.176 g | 0.223 g |
| Riboflavin (B2) | 0.039 mg | 0.021 mg |
| Niacin (B3) | 2.730 mg | 2.050 mg |
| Vitamin B6 | 0.294 mg | 0.103 mg |
| Folacin | 10 mcg | 4.1 mcg |
| Vitamin E | 1.4 mg | 0.462 mg |
| Magnesium | 72.2 mg | 22.6 mg |
| Phosphorus | 142 mg | 57.4 mg |
| Potassium | 137 mg | 57.4 mg |
| Selenium | 26 mg | 19 mg |
| Zinc | 1.05 mg | 0.841 mg |

FDA Health Claims: Brown Rice



Cardio-Disease & Some Cancers

- 1997/2008
- "Diets rich in whole grain foods and other plant foods and low in total fat, saturated fat and cholesterol may reduce the risk of heart disease and some cancers."

Type II Diabetes

- 2013
- "Whole grains may reduce the risk of type 2 diabetes, although the FDA has concluded that there is very little scientific evidence for this claim"

All rice bran isn't created equal



- Total phenolics, flavonoids and anthocyanins, and in diverse purple rice brans. More than 13-, 12-, 25- and 8-fold variations
 - Chen et al. 2017. J Cereal Science 77: 110-119
- Polyphenols reduce obesity-related oxidative stress and inflammation
 - Calcot et al. 2018. Journal of Applied Biomedicine (In press)

Whole Grain Consumption



- Indonesia, Philippines, Iran, Nigeria and a few European countries
- Brown market is projected to expand at a higher pace compared to rice market; shifting preference towards healthy eating habits
 - Diabetics
 - Fitness
 - Healthy eaters
- Increasing per capita consumption in countries where rice is not a staple expected to support the growth of the brown rice market

Whole Grain Consumption

- Due to increasing demand, manufacturers are creating new brown products such as quick cooking, ready to eat and more restaurants are offering

| Indian Breads | | Accompaniments | |
|--|-----------------|--|------|
| Naan | 3.90 | Indian Side Salad | 5.90 |
| Flatbread made with plain flour, yoghurt & milk. | | Sliced red onion, tomato, cucumber, green chili, coriander & chaat masala. ♦ | |
| Garlic Naan | 4.50 | Lime Pickle | 3.00 |
| Naan with fresh garlic butter. | | (Home-made) ♦ | |
| Peshawri Naan | 4.90 | Mango Pickle ♦ | 3.00 |
| Naan stuffed with a blend of cashew nuts, raisins and desiccated coconut. | | Pappadums (4pcs) ♦ | 2.50 |
| It's Mirchi Special Naan | 5.50 | Cucumber Raita ♦ | 3.00 |
| Stuffed with Mushroom, Spinach, Avocado & Onion. | | Mango Chutney ♦ | 3.00 |
| Spinach & Feta Naan | 4.90 | | |
| Tandoori Free-Range Chicken Naan | 5.50 | | |
| Cheese Naan | 4.90 | | |
| Garlic & Cheese Naan | 4.90 | | |
| Avocado & Onion Naan | 4.90 | | |
| Mushroom & Onion Naan | 4.90 | | |
| Onion Kulcha | 4.90 | | |
| Stuffed with diced onion, green chillies & fresh coriander. | | | |
| Extra-Long Rice | | | |
| Steamed Basmati Rice | \$3.00 / L 4.00 | | |
| Plain boiled rice. Basmati rice is a unique species of rice originating from India. ☆ ♦ ♦ | | | |
| Brown Rice | \$4.50 | | |
| Boiled brown rice. A healthier choice. ☆ ♦ ♦ | | | |
| Saffron Rice | \$3.50 / L 4.50 | | |
| Yellow Basmati rice. Crushed saffron and a good pinch of sea salt. ☆ ♦ ♦ | | | |
| Vegetable Rice | \$4.50 / L 6.50 | | |
| Basmati Rice and tender morsels of vegetables, cooked with ginger and garlic, finished with fresh coriander. ☆ ♦ ♦ | | | |
| Cumin Rice | \$3.50 / L 4.50 | | |
| Basmati rice sautéed with ghee and cumin seeds. ☆ ♦ ♦ | | | |
| Coconut Rice ☆ ♦ ♦ | \$3.50 / L 4.50 | | |
| Kashmiri Rice | \$4.50 / L 6.50 | | |
| A popular North Indian style Basmati rice cooked with sultanas, almonds, and cashews, flavoured with cardamom and sugar. ☆ | | | |
| Kids Zone | | | |
| Kids Fish | 9.90 | | |
| Chicken Nuggets | 9.90 | | |
| Butter Chicken ♦ ☆ | 9.90 | | |
| Mango Chicken ☆ ♦ ☆ | 9.90 | | |
| Beef / Chicken / Lamb Korma ♦ ☆ | 9.90 | | |
| Free-Range Chicken (+ \$2) / Organic Lamb (+ \$3) | | | |
| Desserts | | | |
|  | | Gulab jamun | 5.50 |
| | | Solid milk balls soaked in sugar syrup and flavoured with cardamom and rose water. | |
| | | Kulfi (Mango / Malai / Pistachio / Paan) | 5.90 |
| | | Home-made Indian ice cream. | |



Limitations



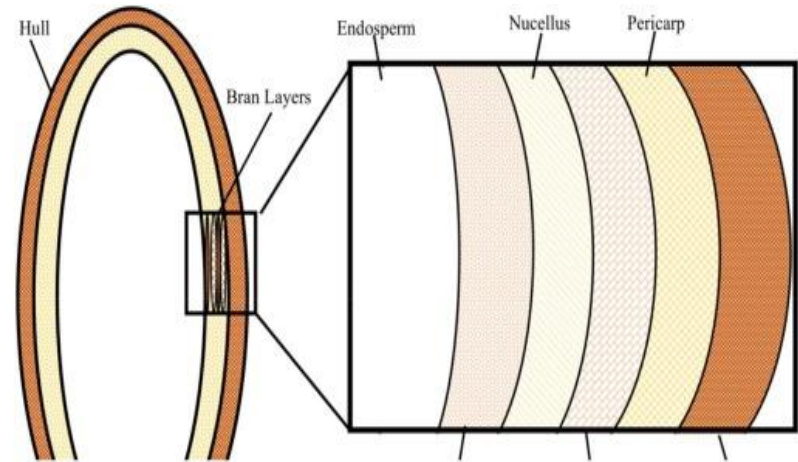
- Consumers in less developed countries see it as poor people's food, etc.
- Developed world consumers think of it as poor tasting and don't like the texture, etc.
- More social marketing campaigns
- Teach consumers, chefs and school foodservice operators how to best prepare



Partially Milled



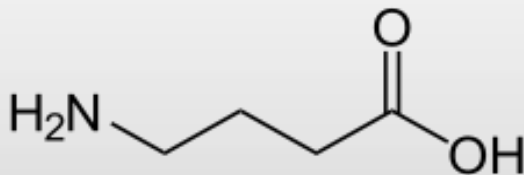
- “Beige rice”
- “Gently milled”
- “Pink rice”
- Softer texture, less cooking needed
- Compromise between sensory and nutritional properties
- India – rice for diabetics
- Japan – consumers can request particular degree of milling!!!



Pre-Germinated Rice



- Whole grain rice soaked in water and allowed to begin germination
- Tremendous physicochemical changes
- “The most nutrient rice rich”
- Popularity growing in S. Korea, Japan, US
- ↓ phytic acid = ↑ mineral bioavailability
- Higher levels of phytochemicals including the neurotransmitter gamma-aminobutyric acid (GABA)



Patel et al. 2011. J Food Sci Technol. 48: 661–667.
Yoto et al. 2012. Amino Acids. 43:1331-7.

Pre-Germinated Rice



- Pre-germinated rice or purified GABA
 - in-vitro (mice): High-fat diet-induced metabolic syndrome parameters improved: BP, blood glucose, serum lipid profile, inflammation indicators.
 - In-vitro (63 human, single blind, cross over): ↓ brain waves indicating stress after mental stress tasks

Pre-Germinated Rice



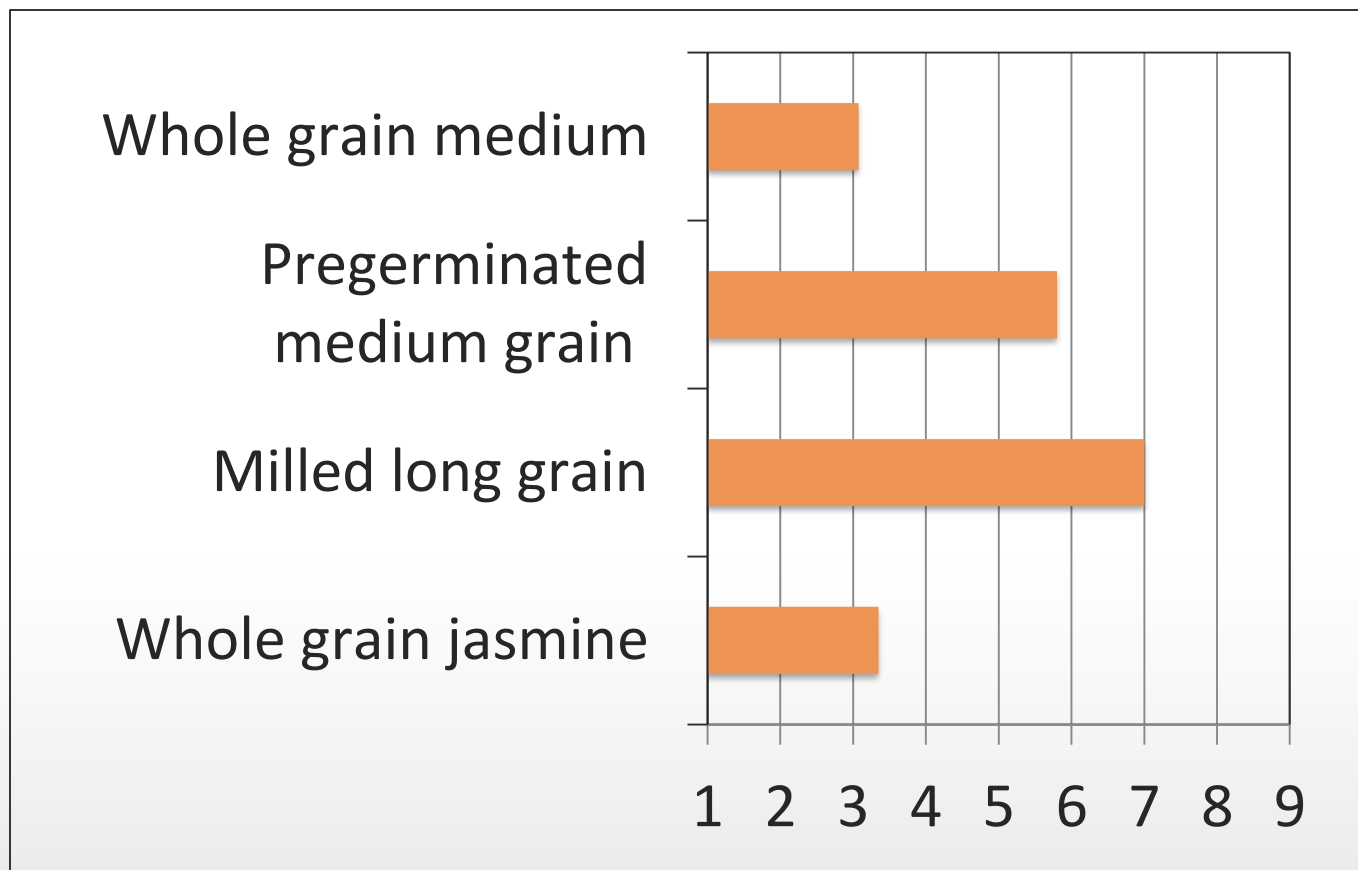
Table 1A. Changes in the major phytochemical composition of Black rice during germination.

| S. No. | Phytochemicals | Concentration/g of Rice (Mean \pm SD) | |
|--------|---------------------------------|---|------------------------------|
| | | Before germination | After germination |
| 1. | <i>Phenolic compounds</i> | | |
| | Protocatechuic acid | 16.77 \pm 0.85 μ g | 31.06 \pm 1.58 μ g** |
| | Chlorogenic acid | 23.42 \pm 1.78 μ g | 43.38 \pm 2.21 μ g*** |
| | Vanillic acid | 126.2 \pm 6.32 μ g | 133.70 \pm 6.82 μ g* |
| | <i>p</i> -coumaric acid | 54.14 \pm 2.72 μ g | 90.27 \pm 4.60 μ g*** |
| 2. | <i>Anthocyanins</i> | | |
| | Cyanidin 3-glucoside | 8.12 \pm 0.42 mg | 15.03 \pm 0.77 mg |
| | Peonidin 3-glucoside | 276 \pm 18.34 mg | 311.11 \pm 15.87 mg* |
| 3. | <i>Tocols</i> | | |
| | δ -tocotrienol | 5.01 \pm 0.03 μ g | 5.21 \pm 0.27 μ g |
| | γ -tocotrienol | 11.05 \pm 0.06 μ g | 11.82 \pm 0.60 μ g |
| 4. | Gamma-Amino Butyric acid (GABA) | 4.4 \pm 0.23 μ g | 202.90 \pm 4.35 μ g*** |

* $P \leq 0.05$; ** $P \leq 0.005$; *** $P \leq 0.001$.

Optimized Process for GABA: 12 hr soaking, pH 7, 36 hr sprouting
 Chaiyasut et al. 2017. Food Sci. Technol, Campinas, 37: 83-93

Mexican style rice – Degree of Liking



Bergman et al. 2014. AACC International Annual Meeting.

Pre-Germinated Rice



- Challenges
 - Water needed for growth
 - Energy use for drying
 - Differences in functional and sensory properties
 - GABA levels are maintained at cooler temperatures
 - Consumers need to learn about its benefits and how to prepare it

Limitations



- Studies frequently don't control for differences in degree of milling – genotype differences can be confounded
- Brown rice is susceptible to rancidity
- Many studies focused on antioxidant activity: yet this hypothesis has yet to be well substantiated – anti-inflammatory effects are better supported but still not as strong as the “germ theory”. Is it cause or effect?
- Most traits haven't been studied in-vivo in well designed human based clinical trials
- Those designed well are short in duration – is there a physiological adaptation unknown?

Traditional Breeding Techniques



- Compounds with the potential for enhancement
 - Fatty acid profile
 - Simple phenolics types and quantities
 - Polyphenolics types and quantities
 - Tocopherols and tocotrienols percentages and amounts
 - Gamma oryzanol compounds and amounts
 - **Resistant starch amount**

Resistant Starch



- Medical community recommending reduced CHO consumption by pre- and diabetics
- Not all starch is the same
- Resistant starch isn't hydrolyzed in small intestine but passes to large:
 - no effect on BS
 - Increased satiety
 - ↓ risk of colon cancer
- AOAC method agrees well with RS values obtained through ileostomy studies

Resistant Starch



- 40 high amylose rice genotypes
- 1.9-fold variation in RS found
- Some had > 2 fold more RS typical US long grains
- Amylose and pasting temperature were strong predictors of resistant starch
- Cooking methods did not alter resistant starch concentrations (1:2 ratio and excel water)

Resistant Starch



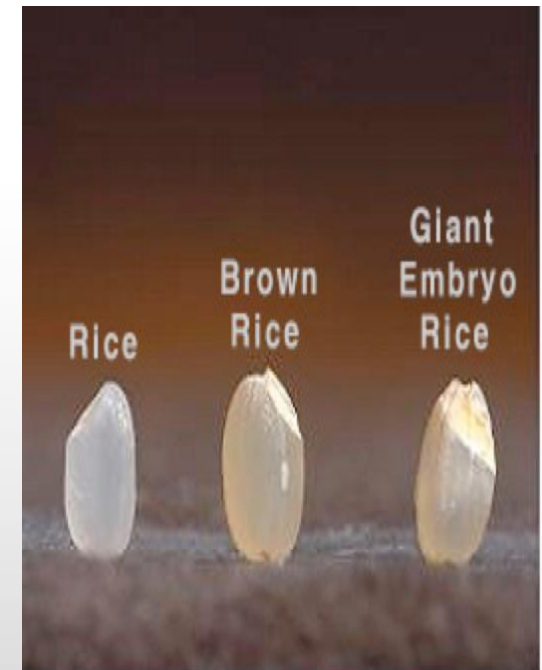
Table 1. Resistant starch concentration in cooked rice using a 1:2 (w:v) rice to water ratio and a 20-min cooking time.

| SampleName ¹ | COUNTRY | Structure | Apparent amylose type ² | Viscosity type ³ | Gel temp type ⁴ | Viscosity_GT haplotype | Resistant starch (%) | |
|-------------------------|-------------|-----------|------------------------------------|-----------------------------|----------------------------|------------------------|----------------------|-------|
| DALIDAO | China | IND | High | Strong | Low | Strong_Low | 4.46 | A |
| Santa Julia | Colombia | IND | High | Weak | Int | Weak_Int | 4.33 | AB |
| KN-1 B-361-BLK-2 | Indonesia | IND | High | Strong | Int | Strong_Int | 4.28 | ABC |
| Ghoal Champa | Iran | IND | High | Weak | Int | Weak_Int | 4.13 | BCD |
| Tsipala 421 | Madagascar | IND | High | Weak | Int | Weak_Int | 4.13 | BCD |
| Vary Vato 462 | Madagascar | ADMIX_I | High | Weak | Int | Weak_Int | 4.07 | BCD |
| Jaya | India | IND | High | Strong | Low | Strong_Low | 4.04 | BCDE |
| CNTRLR80076-44-1-1-1 | Thailand | IND | High | Weak | Int | Weak_Int | 3.96 | CDEF |
| Rojofotsy 738 | Madagascar | ADMIX_I | High | Weak | Int | Weak_Int | 3.93 | DEF |
| Sapundali Local | India | IND | High | Weak | Int | Weak_Int | 3.84 | DEFG |
| Ghati Kamma Nangarhar | Afghanistan | AUS | High | Weak | Int | Weak_Int | 3.74 | EFGH |
| EL Paso L-144 | Uruguay | IND | High | Strong | Low | Strong_Low | 3.73 | EFGHI |
| ECIA76-S89-1 | Cuba | IND | High | Strong/Wea | Low | Strong/Weak_Low | 3.67 | FGHI |

Mutation Breeding

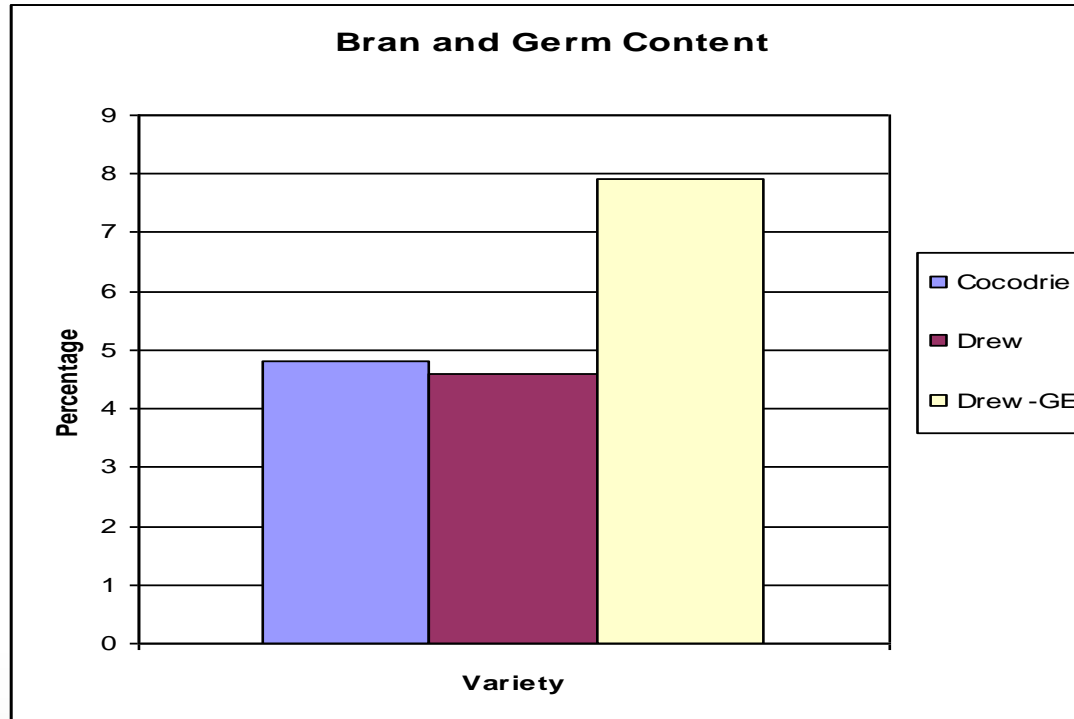


- Gamma ray mutation
- Giant embryo mutant developed from the US cultivar Drew
- Study evaluated Drew, its mutant and 'Cocodrie'



Bergman et al. 2012. Rice Technical Working Group Abstract 4487.

Giant Embryo



- ~ 2x more bran and germ as a % of endosperm
- Closer in proportion to the amount of bran and germ in wheat

Limitations



- Agronomics need improvement
- Segregation/identity preservation techniques
- An industry partner is likely needed to support the development of nutritionally enhanced genotypes
- Such partnerships have occurred. e.g., Industry partners and the USDA
 - Quality suitable for canning and steam table stability
 - Brown rice that combined with a patented process results in quick cooking rice

Genetic Engineering



- First generation
 - Disease, insect resistance
- **Second generation**
 - End-use and nutritional quality traits
 - Traits with limited potential for enhancement using genetic variation
 - Folate, beta carotene, Zn, Fe
- Third generation
 - Pharmaceuticals, other industrial compounds

High Beta-Carotene Rice Update



- **Substitution Study**

- Substitution of biofortified rice for white rice in an optimistic scenario (70% substitution)
- would ↓ the prevalence of vitamin A inadequacy 78% in women and 71% in children in Bangladesh
- De Moura et al. 2016. Amer J of Clinical Nutr, 104: 769–775

- **Safety trial**

- Standard toxicology evaluation – histopathology, hematology and blood biochemistry
- Lack of biologically meaningful unintended effects
- Wu et al. 2017. Regulatory Toxicology and Pharmacology 88: 66-71



High Fe and Zn



- IR68144 (indica): insertion of soybean ferritin gene under the control of the endosperm specific glutelin promoter
- Increased Fe and Zn in the endosperm - only changes identified
- GRAS in the US

Table 2
Mineral content of ferritin rice and IR68144 rice.

| Mineral (mg/100 g) | Brown rice | | | Milled rice | | |
|--------------------|--------------------------|---------------|-------------------------|--------------------------|---------------|-------------------------|
| | IR68144 | Ferritin rice | Ref. range ^b | IR68144 | Ferritin rice | Ref. range ^b |
| Sodium | 2.65 ± 0.08 | 2.63 ± 0.17 | 2–40 | 1.65 ± 0.11 | 1.77 ± 0.05 | 0.6–10 |
| Potassium | 290.83 ± 6.56 | 289.5 ± 1.04 | 70–320 | 251.73 ± 5.24 | 252.8 ± 3.08 | 80–150 |
| Copper | 0.32 ± 0.01 ^a | 0.35 ± 0.004 | 0.1–0.7 | 0.29 ± 0.004 | 0.29 ± 0.02 | 0.2–0.3 |
| Manganese | 1.77 ± 0.01 ^a | 1.98 ± 0.05 | 0.2–4.2 | 1.22 ± 0.01 | 1.29 ± 0.03 | 0.7–2.0 |
| Magnesium | 110.5 ± 1.60 | 104.7 ± 1.66 | 20–170 | 82.2 ± 1.80 | 83.6 ± 0.87 | 20–60 |
| Iron | 1.57 ± 0.02 ^a | 2.01 ± 0.04 | 0.2–6.0 | 0.65 ± 0.01 ^a | 1.6 ± 0.01 | 0.2–3.3 |
| Zinc | 3.01 ± 0.02 ^a | 3.35 ± 0.03 | 0.7–3.3 | 2.34 ± 0.06 ^a | 2.75 ± 0.02 | 0.7–2.7 |

Values are mean ± SEM. n = 3.

^a IR68144 rice values were significantly different from ferritin rice at $p < 0.05$.

^b Source: OECD (2004).

Limitations



- Genetics is moving faster than our understanding of health beneficial phenotypes and our ability to quantify them
 - Proteomics, metabolomics, etc.
- Understanding of rice components is ahead of our confidence in their effects on human health
- We need more clinical trials!!!
- Consumer acceptance of genetically modified rice needs to increase
- The Nobel Laureates' (n=129) Campaign Supporting GMOs for the benefit of the developing world, specially addressing Green Parties and consumers
 - Roberts 2018. Journal of Innovation & Knowledge (In press)

Future



- With CRISPR Cas9 genome editing technology “the sky is the limit”
- Increase a trait, decrease it, or modify it
- Multi-biofortification approach more likely to occur in the future
- Geneticists, nutritionists, food scientists and public health educators must work together to create and promote *healthier rice*

Collaborators



- Ming Chen
- Anna McClung
- Shannon Pinson
- Jeff Shen
- Andrew Moreo



- Thank you conference organizers for the kind offer to come and speak with you about Healthier Rice

What will rice look like in 50 years? Similar to today.

But, hopefully, however, healthier to be able to meet the nutritional needs of our growing human population.....





