

RSC&RGDU  
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# Leaf Protein from Biofortified Rice: A Game Changer

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Rice Science Center

**Population  
10  
Billion**



**More Rice  
Demand**

**56%**

**Climate  
Change**

**Grain Yield  
Reduction**

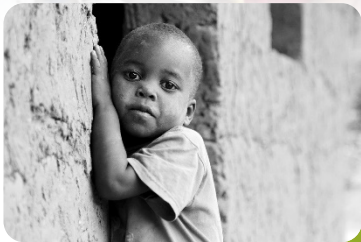
**Starvation**

**Unequal Income &  
Food Distribution**

**Diet High Starch  
Low Fiber**

**Obesity and NCDs**

**BCG  
Biofortified  
Leaf & Grains  
Wellbeing**



**36 Years  
2050**



# Rainbow Rice

- Newly developed biofortified rice orchestrates full spectrum of leaf pigments and patterns created by chlorophyll, anthocyanin and carotenoids



**Solid-Purple Leaf Rice**



X



**White-striped Mutant**

2006



Dr Sulaiman and Jaroon



**Rainbow 01**



**Rainbow 02**



**Rainbow 03**



**Rainbow 04**



**Rainbow 05**



Dr Siriphat

2016

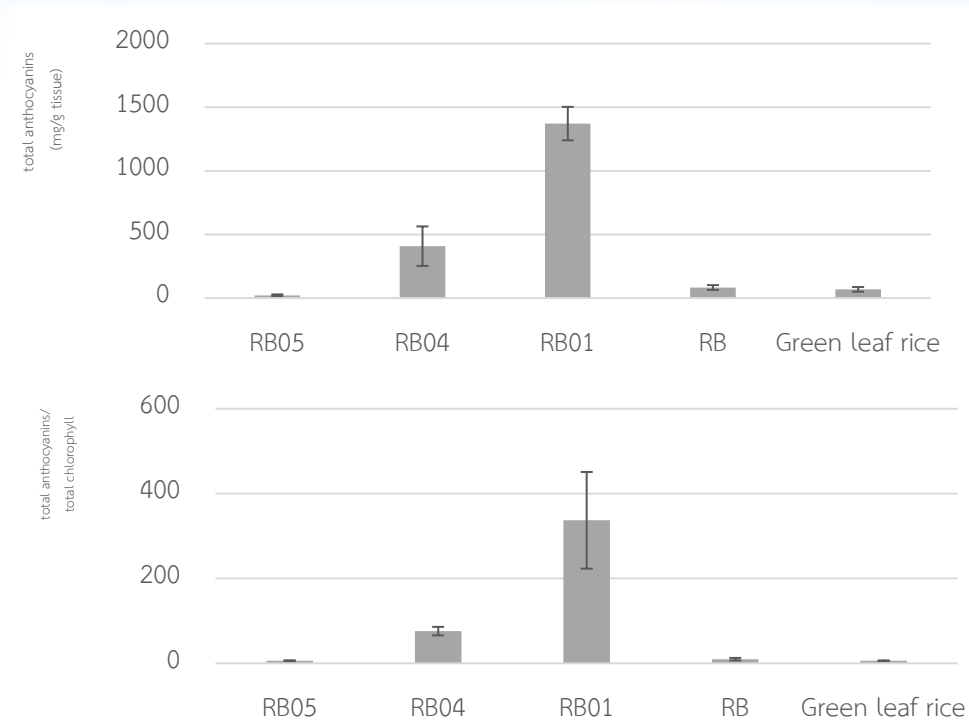
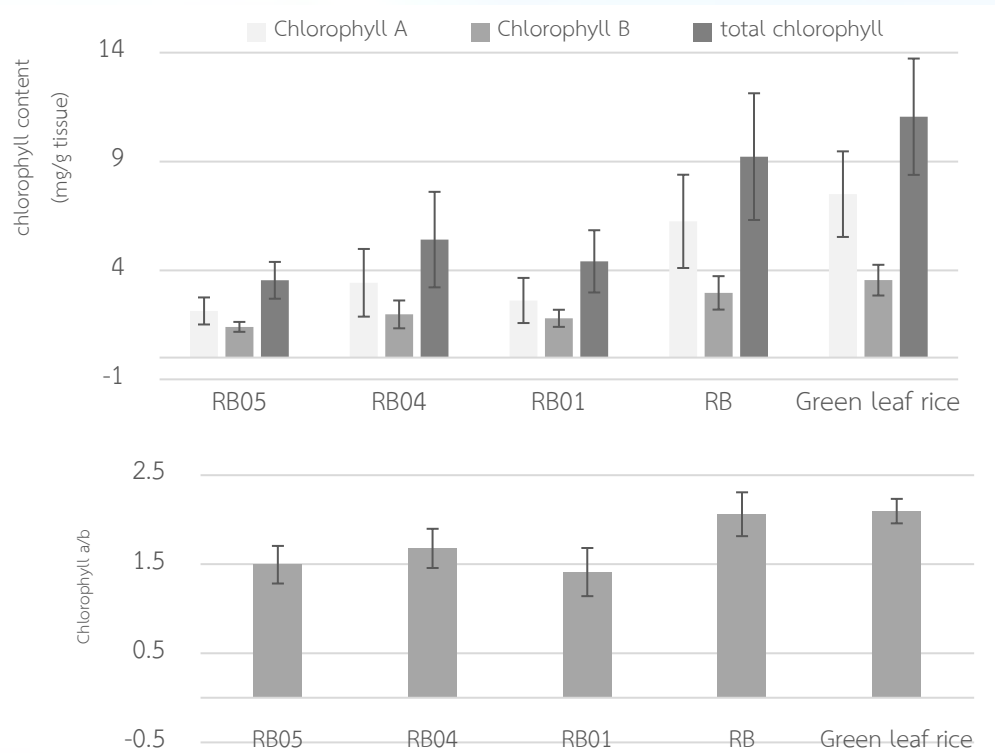
2022



# Pigment Contents: Chlorophyll and Anthocyanin



Teeraphan



# Create Unseen Agrotourism

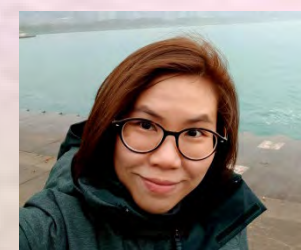
Agro-tourism City

10x on Farm Earning

Therapeutic Products



# Rainbow Rice Leaves are More Nutritious Than the Whole Grains and Brans,

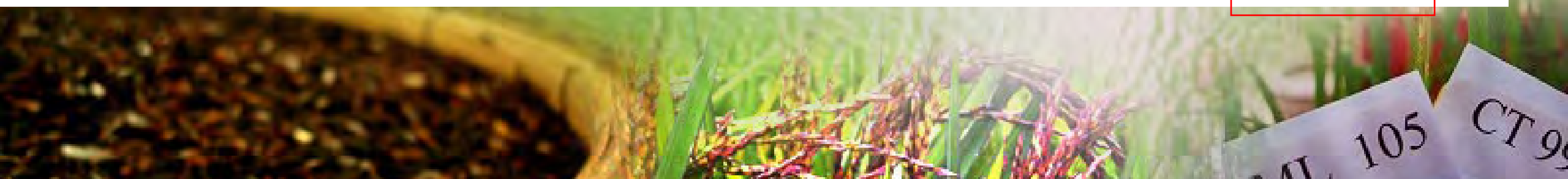


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Teeraphan

Composition		Rice Grain n=17	Rice Bran n=14	Rice Leaf n=5
Carbohydrate	Total starch (%)	82.14 (75.83-85.80)	23.11 (8.88-40.10)	0.56 (0.14-0.97)
	Total dietary fiber (%)	3.61 (2.96-5.43)	32.29 (26.36-42.55)	59.90 (55.82-67.29)
	Insoluble dietary fiber (%)	2.88 (1.65-4.32)	29.10 (23.91-40.33)	57.13 (50.80-66.02)
	Soluble dietary fiber (%)	0.73 (0.26-1.37)	3.19 (1.72-4.36)	2.77 (0.93-5.22)
	Beta-glucan (%)	0.08 (0.03-0.14)	0.26 (0.21-0.40)	0.33 (0.29-0.36)
Protein	Total protein (%)	7.1-8.3*	18.13 (1.68-20.12)	18.22 (14.10-20.48)



70 % BIOMASS 30 %

Leaves

Grains

Starch %	0	70-80
Dietary Fibre	70-85	<10
Protein %	20	8
Antioxidant ORAC	> 90,000	< 40,000
Phytate mg/100 g	12.7	40.3
Fe mg/kg	153.26	9.4
Zn mg/kg	5.39	23.05
Mn mg/kg	325.5	20.65
Mg mg/kg	2,219	1,653
Ca mg/kg	6,791	112
K mg/kg	12,499	1,958
P mg/kg	567	3,485





# Consistency of Leaf Protein Content



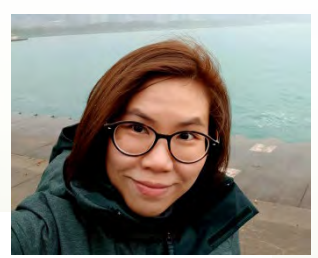
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**Table 2** Protein content in rainbow rice leaf samples (year 2021-2023)

Leaf samples	Year 2021			Leaf samples	Year 2022			Year 2023		
RBR02	19.94	±	0.14 <sup>c</sup>	RB01	16.29	±	0.15 <sub>c</sub>	16.89	±	0.03 <sub>c</sub>
DSK 33	20.48	±	0.03 <sup>d</sup>	RB02	16.65	±	0.39 <sub>c</sub>	18.07	±	0.03 <sub>d</sub>
DSK 104	19.79	±	0.34 <sup>c</sup>	RB03	13.21	±	0.23 <sub>a</sub>	16.54	±	0.13 <sub>c</sub>
RBR 03	16.80	±	0.08 <sup>b</sup>	RB04	15.19	±	0.37 <sub>b</sub>	15.53	±	0.05 <sub>b</sub>
RBR05	14.10	±	0.26 <sup>a</sup>	RB05	13.02	±	0.11 <sub>a</sub>	13.44	±	0.03 <sub>a</sub>

Results are the means ± SD on dry basis and expressed as the percentage of leaf powder. Values with different letters in the same row are significantly different with  $p \leq 0.05$ .

# High Protein Leaf in other Green and Purple Rice Varieties



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**Table 1** Protein content in rice leaf samples

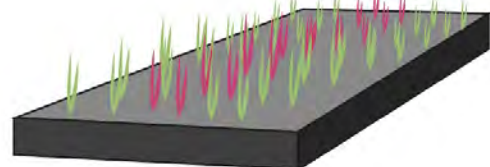
Rice leaf	Protein			Rice leaf	Protein		
RBR01	16.89	±	0.03 <sup>c</sup>	Riceberry (DG)	20.58	±	0.05 <sup>fg</sup>
RBR02	18.07	±	0.03 <sup>d</sup>	Sinlek (G)	20.24	±	0.12 <sup>fg</sup>
RBR03	16.54	±	0.13 <sup>c</sup>	Pitsanulok 2 (G)	19.20	±	0.03 <sup>e</sup>
RBR04	15.53	±	0.05 <sup>b</sup>	White-green JHN (W-G)	16.74	±	0.65 <sup>c</sup>
RBR05	13.44	±	0.03 <sup>a</sup>	Jao Hom Nin (DG)	21.10	±	1.34 <sup>g</sup>
				Klum Doi Saket (Pur)	20.06	±	0.14 <sup>ef</sup>
				Klum Hom Neun (G)	18.06	±	0.27 <sup>d</sup>
				Black Aro Sukothai (G)	14.36	±	0.20 <sup>a</sup>



# Economic Impact



1st Cut



Seedling Sprout Transplanted

60 Days

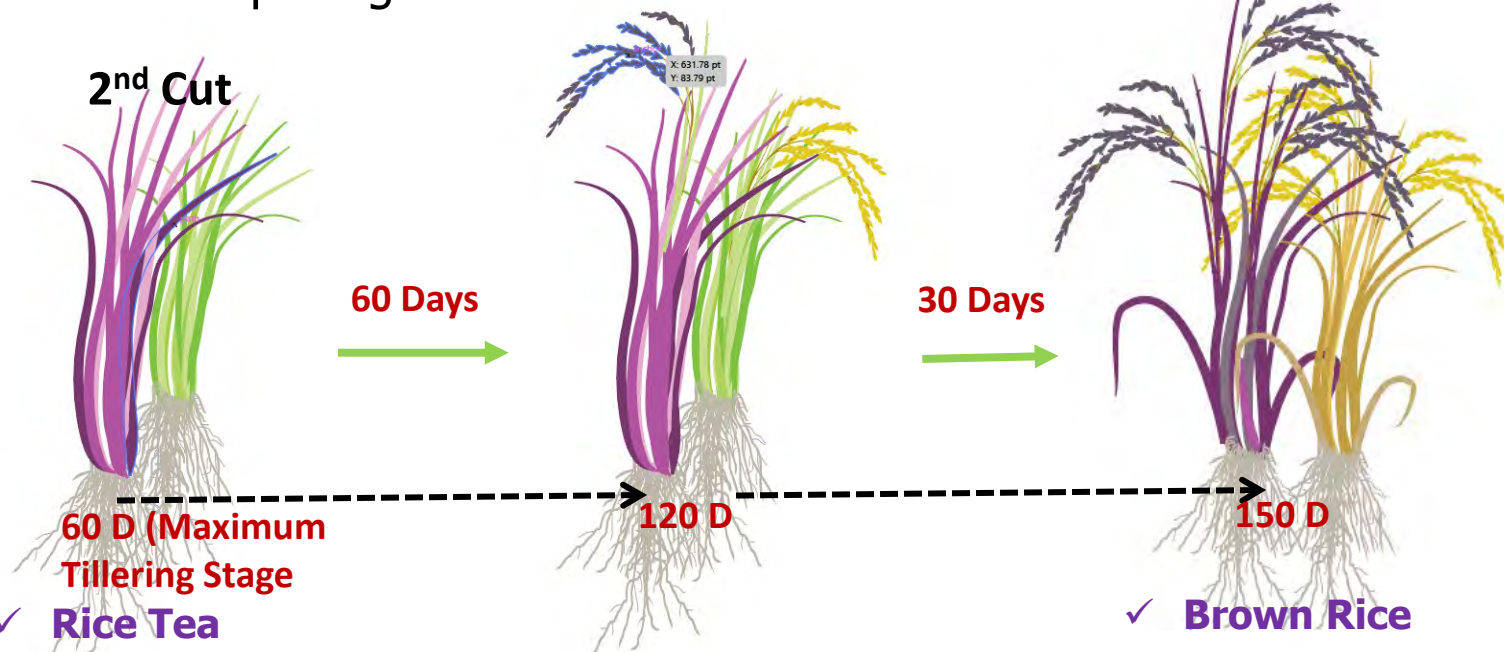
✓ Ricegrass drink

	Wheat Grass	Rainbow Rice (Yong leaves)
	mg/oz	mg/oz
<b>K</b>	42	454
<b>Ca</b>	7.2	28.4
<b>Fe</b>	0.66	479
<b>Mg</b>	8	56.7
<b>Phytate</b>	0.85	0

# Three-step Harvesting

- 1) Sprouting leaves
- 2) Vegetative leaves at 60-DAG
- 3) Seed Harvest

Whole crop silage



- ✓ Rice Tea
- ✓ Non-caloric carbohydrate
- ✓ Dietary fiber
- ✓ Natural pigment



Harvesting

✓ Brown Rice

Only 50% grain yield  
Of solid-green rice

# Harvesting and Preparation

## Challenges

### How to preserve antioxidants?

- **Manual**
  - Cut manually using scissors or sickles
  - Quickly Preserved on dried ice in ice boxes
  - Cool air-dried and cut in small pieces
  - Add liquid nitrogen and grind to powder
  - Freeze in -80 C for months
- **Mechanization**
  - Leaf Collector/Cooler Machine
  - Industrial Freeze-Dry Technology



**Riceberry**



**Rainbow 01**

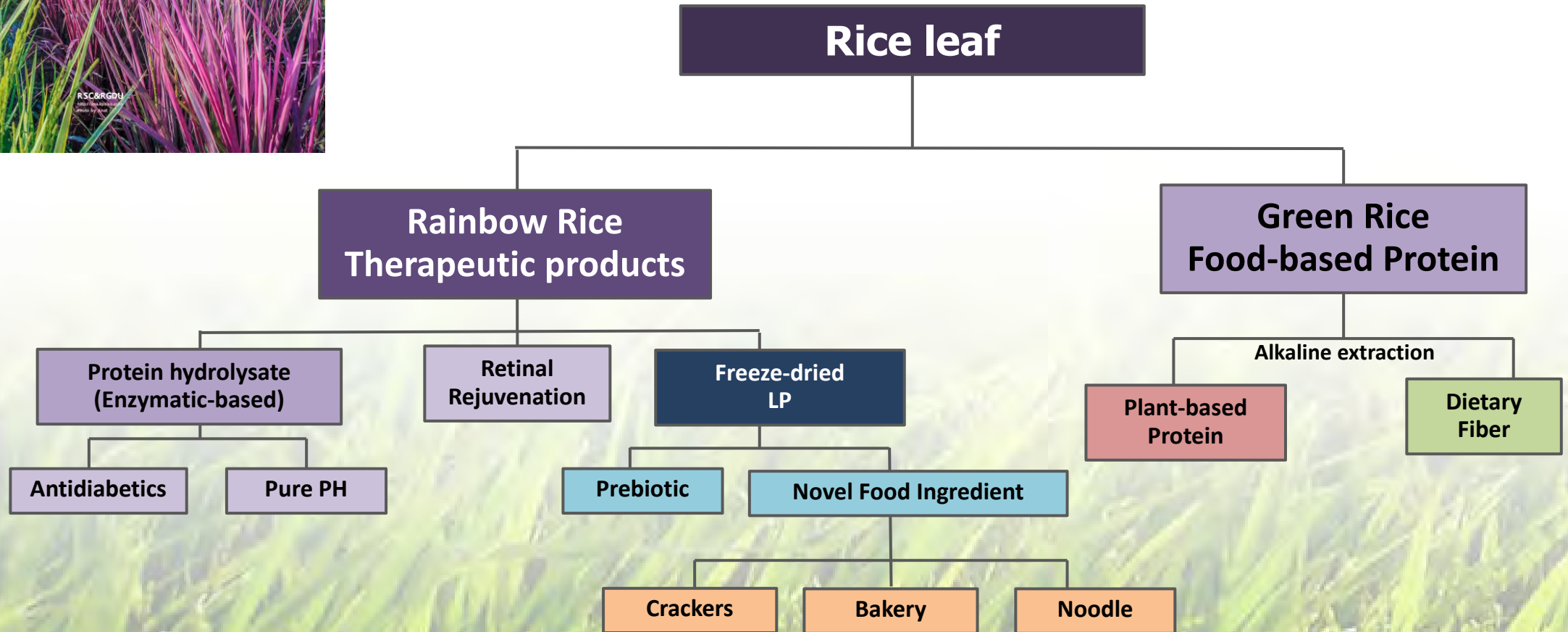


**Rainbow 02**



**RBL: Rainbow Rice Leaf**

**RBLP: Rainbow Rice Leaf Powder**



# Therapeutic Functions of Riceberry Bran Protein Hydrolysate (RBPH) from Organic Farming

*RBPH has therapeutic effects against NCD for the elderly.*

## Composition

19.24% protein,  
49.44 mg GAE/g  
phenolic content,  
11.13 mg of CE/g  
flavonoid content  
0.43 mg Cyn-3 Glu/  
g anthocyanin

## Antioxidative activities

### Anti-inflammatory activities

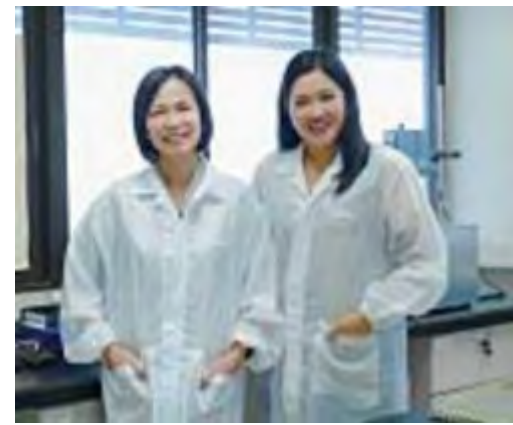
- Inhibiting NO

### Antidiabetic (1 mg/mL) via

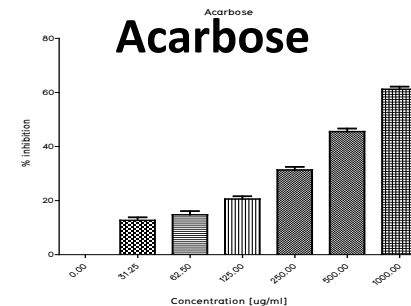
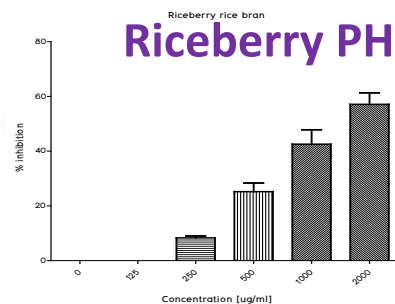
- inhibiting alpha-amylase (30.99%)

- alpha-glucosidase (57.11%)

Inducing cancer inhibitory

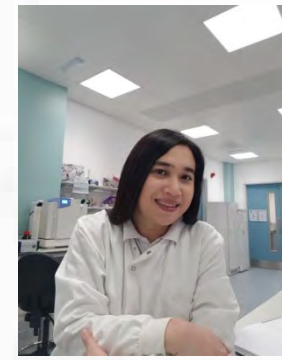


**Dr Sukuntharoj and  
Dr Pornphimon  
KU Agric Product Insti**



# Can Rainbow Rice Leaf Extract Improve Vision Loss?

Institute of Nutrition Mahidol University (INMU)



Dr Chawanrnpat



Assoc Prof.Ratcha

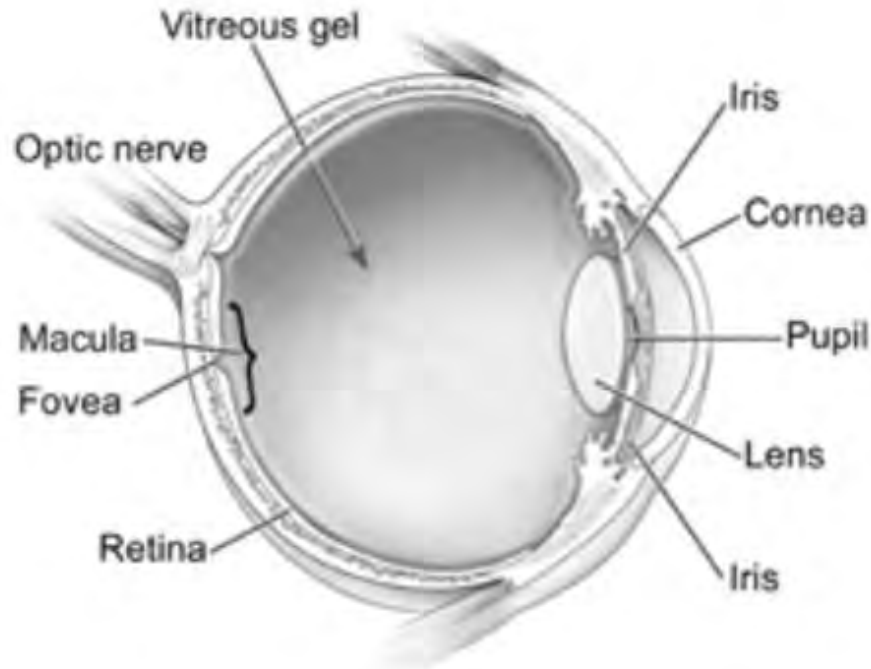
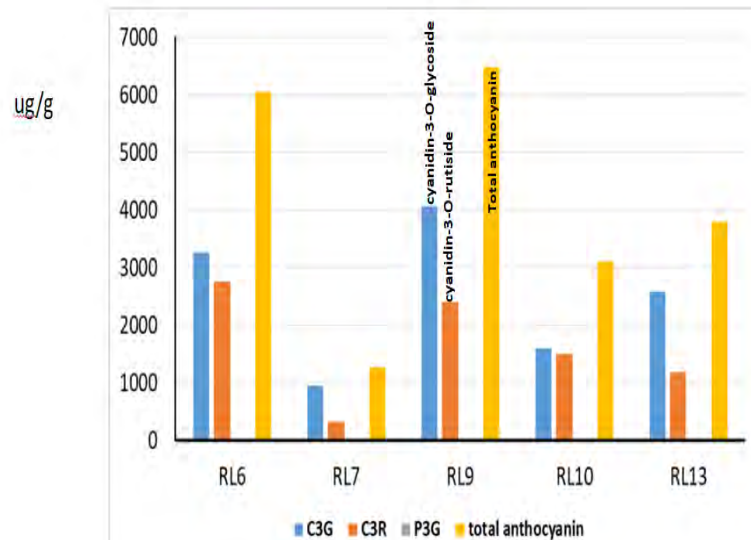


Diagram of the eye, viewed from the side

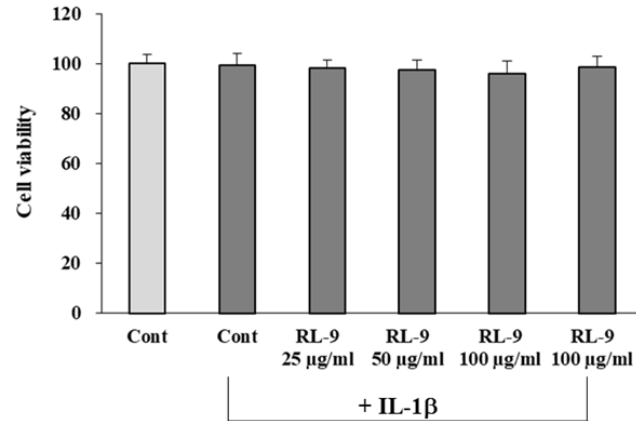
- Loss of vision is a retina disease affecting the macular area
- Oxidative stress on retinal pigment is the major cause of **Macular Degeneracy (MD)**
- Often found in elderly population
- Need high antioxidant supplements to prolong MD



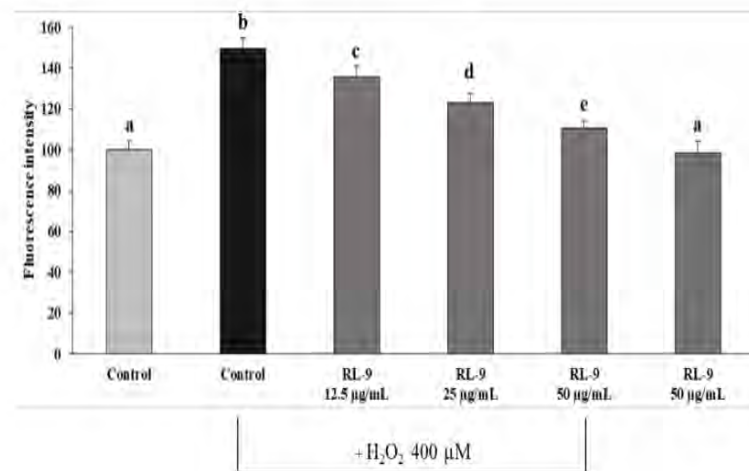
Ethanol extracts of Rainbow rice leaf is rich in flavonoids, carotenoids, lutein, xanthophyll, phenolic, and anthocyanins, particularly, Cyanidin-3-O-rutinoside. In the cherry- red Leaf



**No toxic effects or changes in cell viability of ARPE-19 cells treated with Rainbow Rice leaf extracts**



12-50 microgram of ethanol extracts were added to H<sub>2</sub>O<sub>2</sub>-treated ARPE-19. The leaf extract RL-9 showed a significant rate-dependent response to 400 micromole H<sub>2</sub>O<sub>2</sub>





# Effective Prebiotics

- Human intestinal system contains a large-diverse population of **Gut Microbiota (Probiotics)**.
- Their functions are important for the host well-being by improving immune system, digestion, detoxification, and preventing non-communicable disease (NCDs).
- **Prebiotics** : Specific substrates to protect and grow gut microbiota in our digestive system: **Dietary fiber**



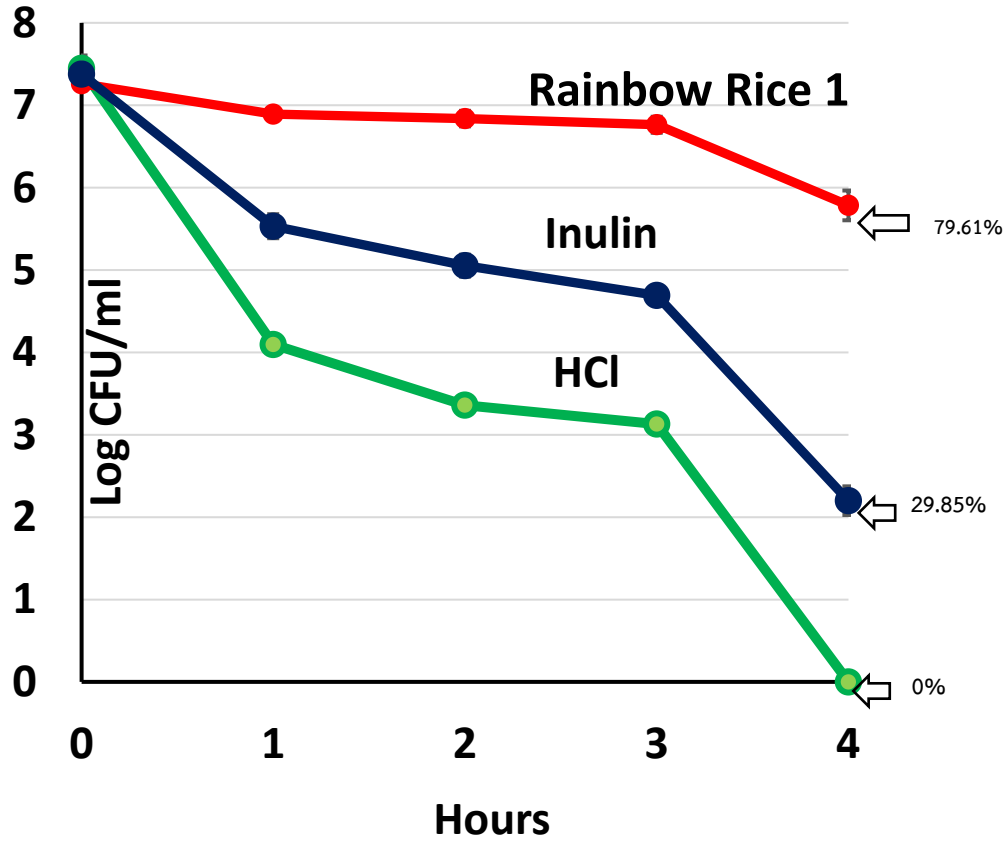
Pop Nanthanat



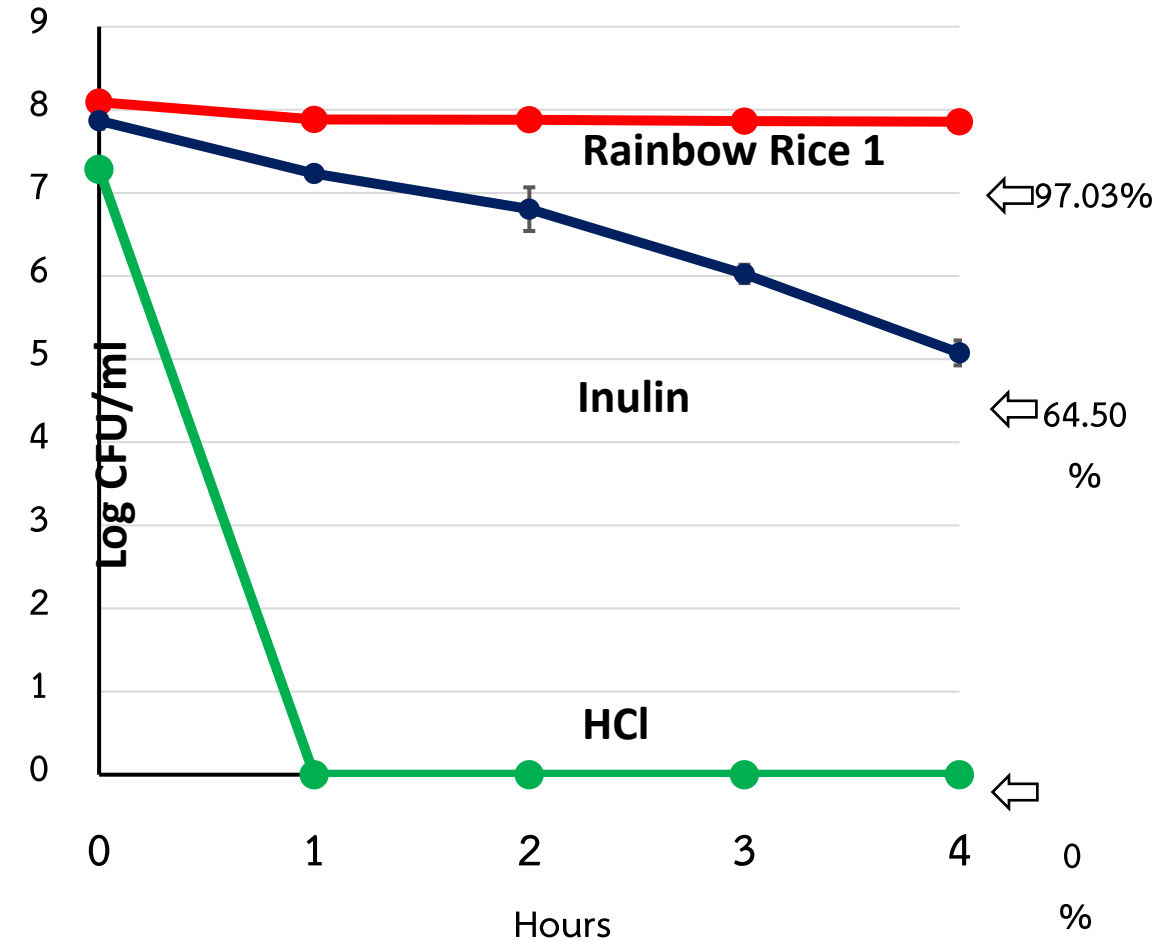
Serving size 1 capsule (0.23 g)

Antioxidant	Antioxidant (ORAC)	93,008.70 ORAC μmoles TE/100 g
	Antioxidant (FRAP)	17,088.31 FRAP μmoles TE/100 g
Carbohydrate (%DM)	Total dietary fiber (TDF)	
	Insoluble dietary fiber (IDF)	0.1518 g
	Soluble dietary fiber (SDF)	0.0029 g
	Beta-glucan	0.0007 g
	Soluble carbohydrate	0.0071 g
	Soluble sugar	0.0042 g
	Total Starch	0.0012 g
Protein (% DM)	Total protein	0.0458 g

## RBLP protected Probiotics from a gut enzyme



Survival rate of probiotic, *L.rhamnosus* KUKPS6007 in acid conditions (pH2, HCl) at 0, 1, 2, 3, and 4 hours



Survival rate of probiotic, *L.reuteri* KUKPS6103 in acid conditions (pH2, HCl) at 0, 1, 2, 3, and 4 hours

# Adding RBLP to Bakery Products to Reduce Glycemic Index (GI)



Pann Bakery SME



**RBLP**  
**Rainbow Rice Leaf Powder**



- **1.5-10 % RBLP**
- **20% whole Riceberry flour**
- **wheat flour**



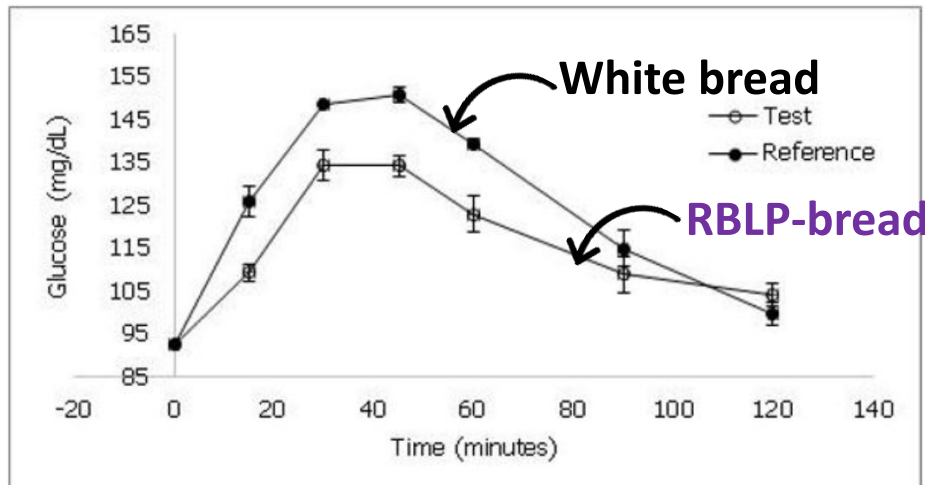
# Can RBLP affects Glycemic Responses in Bread?

## In vitro analysis of Glycemic Index (GI)

Amount of **glucose** released from **50 g carbohydrate** foods within 20-180 minutes after digestion with pepsin, invertase, and pancreatic  $\alpha$ -amylase enzymes at pH 1.5,  
Comparing **Area Under Curve (AUC)** of the released glucose from RBLP-bread vs white bread.



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**Adding 1.5% RBLP can reduce 10 units of GI in white bread**

RAG = Rapid Available Glucose (20 min),  
SAG = Slowly Available Glucose (20-40 min)  
 $C_{\infty}$  = Equilibrium concentration; k = Kinetic constant;  
HI = Hydrolysis Index;  
GI = Glycemic Index =  $39.71 + 0.549HI$

Bread Sample	%Available glucose						In vitro kinetic of starch digestion			
	RAG			SAG			Ca	k	HI	GI
White bread	50.5	$\pm$	3.1	17.4	$\pm$	0.1	60.87	0.06	100	95
1.5% Rainbow Rice Powder	42.3	$\pm$	1.4	16.4	$\pm$	0.2	51.72	0.06	84	86

# Conclusions

- Biofortified Rainbow Rice leaf enriched with dietary fibre, antioxidants, protein, and micronutrients Fe, Mg, and Mn, and starch-free has a solid potential bioresource for inventing various therapeutic products against NCDs.
- Agro-tourism developed from Rainbow Rice will make an immediate economic impact on farms.
- On a full scale, this novel food concept can be expanded to capture any high leaf-protein rice cultivar to scale up plant-based protein production for food and nutrition security.
- Lastly, we hope to improve farmers' attitudes against open-field burning while saving whole rice plants for the future of future foods.

THANK YOU



# Stop Open-field Burning!

- Burning seasons cause serious air pollution and health problems in India, Thailand, and Vietnam
- From farmers' misconception about eradicating life cycles of insect pest and obstruction of soil preparation
- Fixed Carbon lost