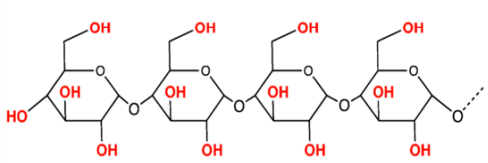
Status of Rice Genetic Improvement for Low GI/GL and Insulinotropic Control

Professor Apichart Vanavichit, Rice Science Center, Kasetsart University

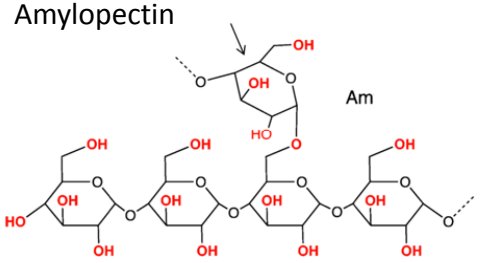
Plant Starch: Solar Energy Storage

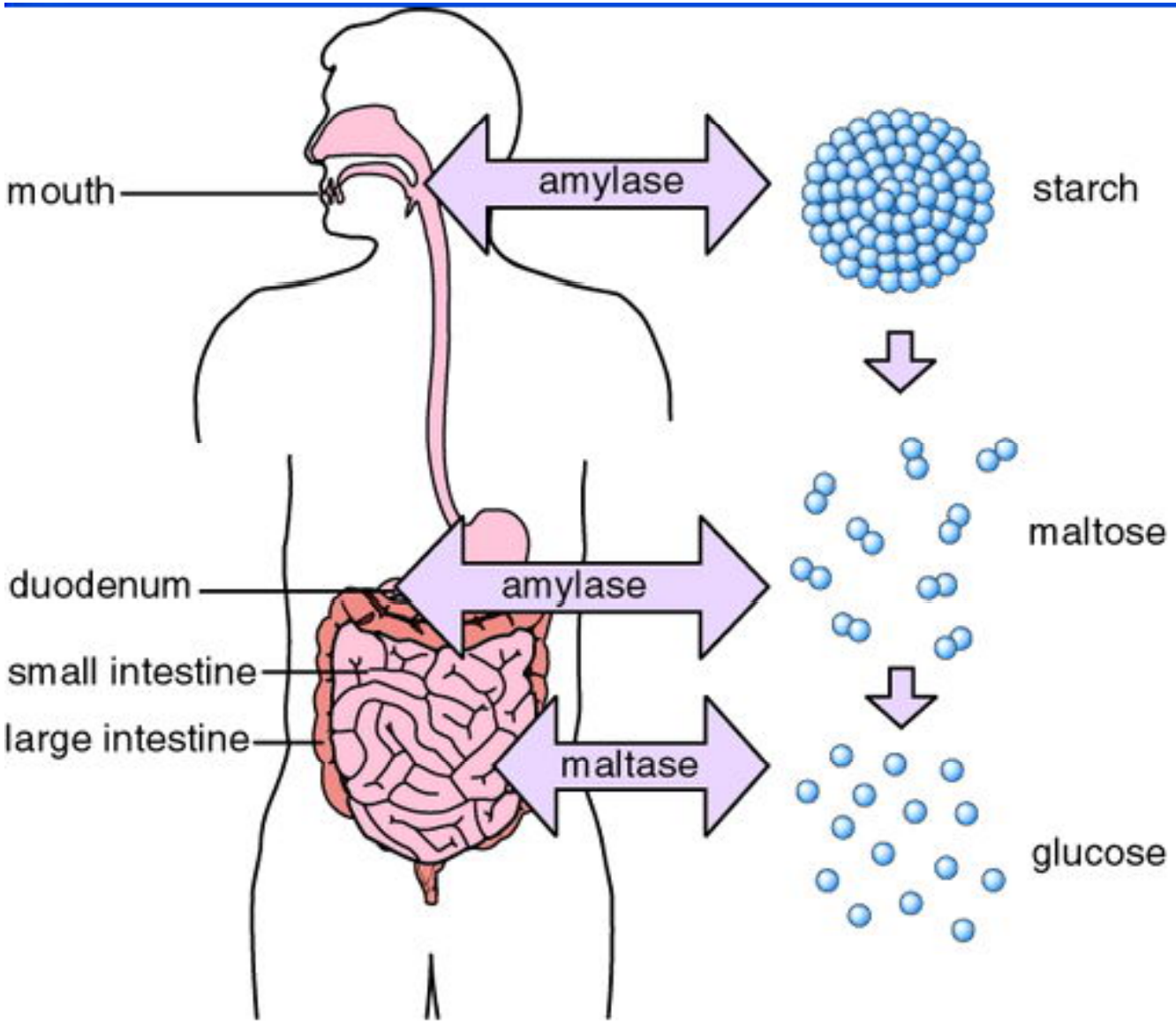
- Plant stores solar energy as starch via photosynthesis
- Animal converts starch to glucose and save extra-blood glucose as glycogen

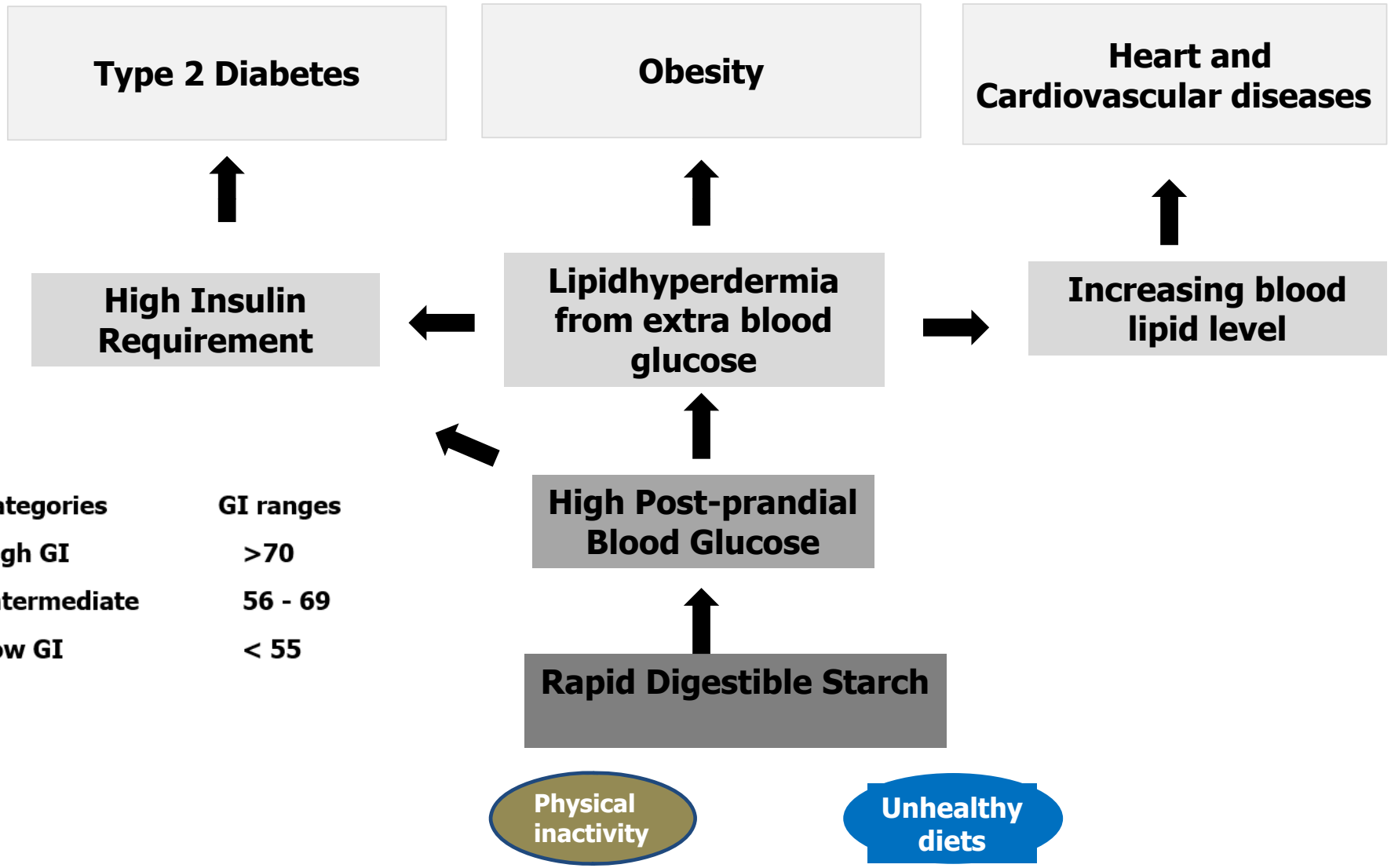
Amylose



Amylopectin

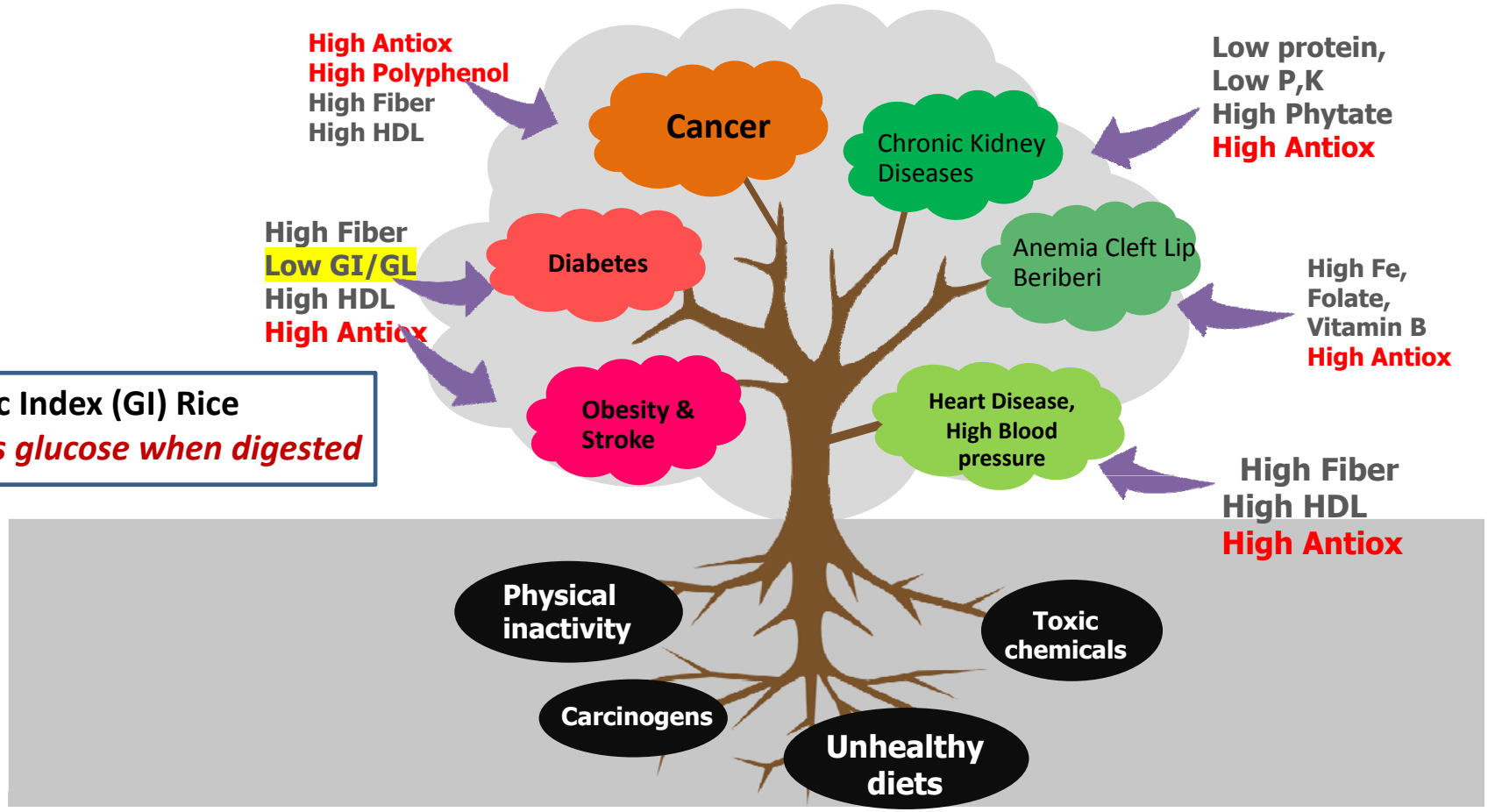



mouth
amylase
starch
maltose
duodenum
amylase
small intestine
large intestine
maltase
glucose



Categories	GI ranges
High GI	>70
Intermediate	56 - 69
Low GI	< 55

Specialty Rice for Non-Communicable Diseases (NCD)



Low Glycemic Index (GI) Rice
releasing less glucose when digested

Improved Rice Cultivars became higher GI than Traditional Varieties

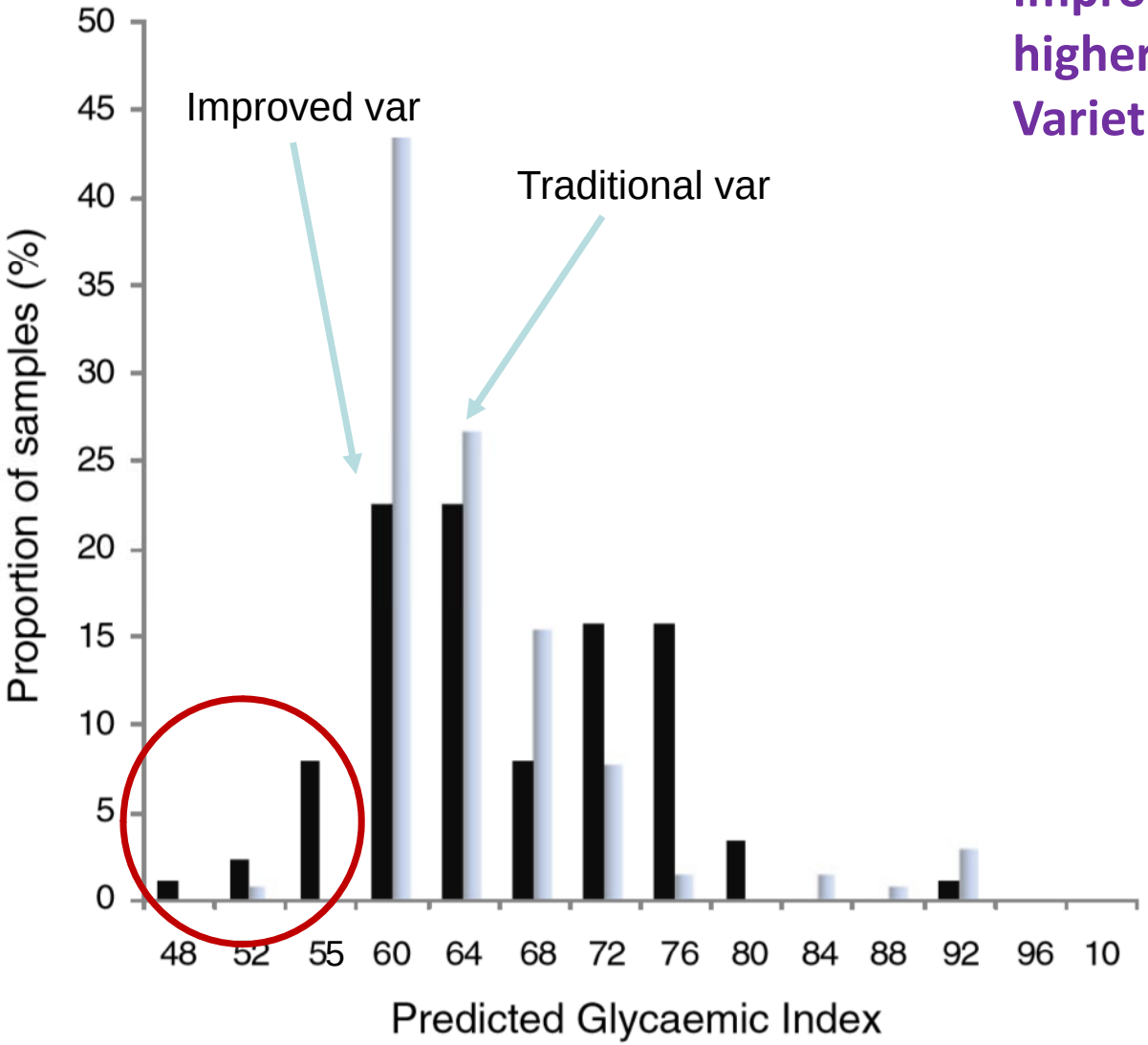


Fig. 4 Frequency histogram showing that the distribution in GI of improved varieties of rice (*black bars*) contains more high GI rices than the distribution of the traditional varieties (*grey bars*).

Fitzgerald et al., 2011. Rice J. 4:66

GLYCEMIC INDEX FOUNDATION
Making healthy choices easy

Glycemic Index Testing & Research

Sydney University Glycemic Index Research Service (SUGiRS)

The Sydney University GI Research Service (shortened to SUGiRS) was established in 1995 to provide a reliable commercial GI testing laboratory. Food samples are tested in healthy volunteers according to standardised methods that have been validated against overseas laboratories. Testing of foods for their glycemic index, insulin index, satiety response, and other metabolic parameters can be assessed simultaneously. Other analyses such as in vitro GI testing are available. SUGiRS has an established reputation for quality, speed and flexibility.

SUGIRS can work with your company to develop new low GI products or help lower the GI of existing ones. Foods that meet nutrition guidelines and have been GI tested can carry the GI symbol (For more go to **www.gisymbol.com/join-the-program**) or make a low GI **nutrition content claim** in Australia. Your results are strictly confidential and are your property. Data are released for publication only with your written approval.

How long does it take to measure GI values of foods?

On average, it takes approximately one week to recruit 10 healthy people to participate in a study and then one week to test each product and up to another week to complete a detailed report of the study. However, as soon as GI values are finalised, they can be emailed or faxed to clients. For larger studies and those involving the measurement of insulin values, an additional one or two weeks may be required to complete all of the biochemical analyses. However, we try to complete each project at the fastest rate possible and usually complete a study earlier than expected. Determining the GI values of foods involves the collection of blood samples from the study participants, so we have to allow time for the participants to recover from the sampling between sessions.

Road-map to

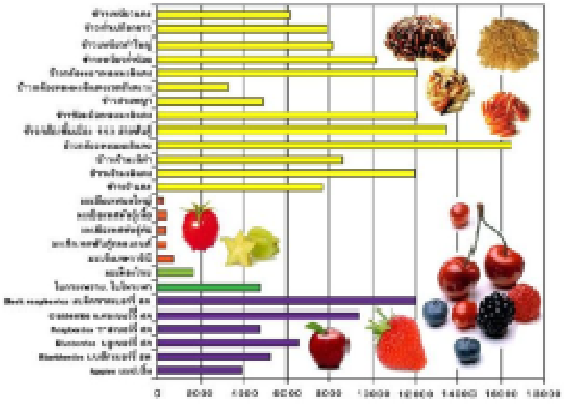
Developing Better rice for Better World

Nutrient-dense, Eco-friendly Rice

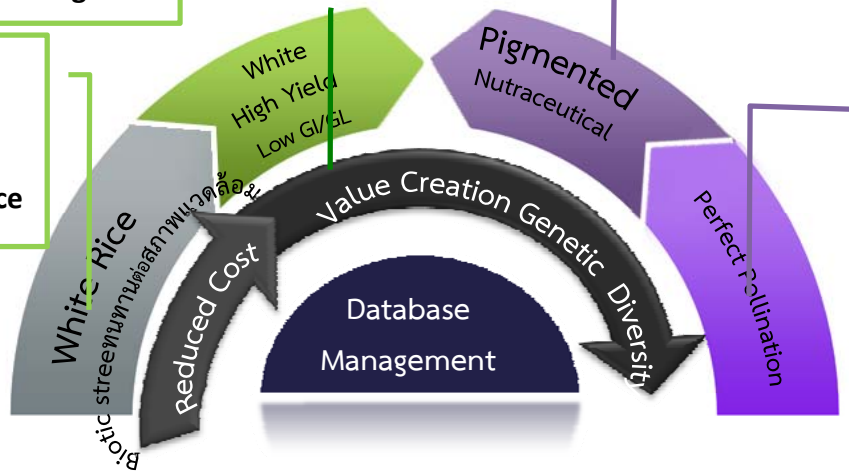
- **Jasmine+4**
- Rice blast
- BPH
- Leaf blight
- Submergence

- Low GI/GL
- **PinK+4**
- Rice blast
- BPH
- Leaf blight
- Submergence

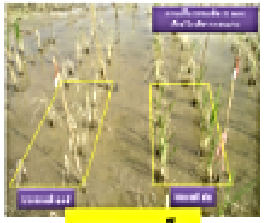
- **Riceberry**
- Antioxidant
- Vit B complex
- Carotenoid
- Hi fiber
- Hi minerals



- **RD6+4**
- Rice blast
- Leaf blight
- Submergence



- **Climate Ready**
- Heat
- Cold
- Fe toxic
- Drought
- Stomata
- Salinity



Hypothesis behind low GI Rice

Am/Ap	Amylopectin Structure	Resistant starch
Dietary fiber	Amylolysis enzyme	Polyphenolic compounds

Varietal Development of Low GI Rice

Stage 1: ScreenSinlek and Riceberry (2007)

Stage 2: Gene pyramiding breeding Pinkaset+4 (2010)

Stage 3: Screening improved RD and local cultivars (2018)

Clinical Glycemic Index 2010

Rice varieties: Riceberry and Sinlek

Rice type: Brown and Polished

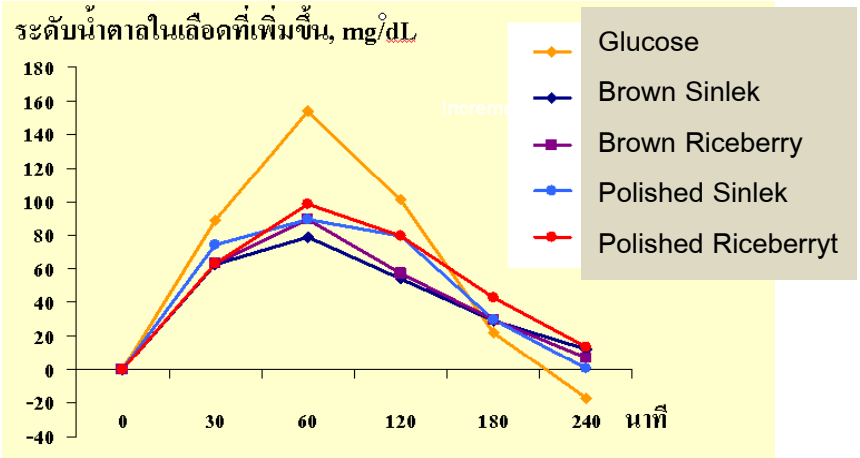
Clinical subjects: 16 Diabetes type II, Hyperchrolesterol

Meals: 50 g Carbohydrate + Hot basil minced pork

Post-prandial glucose collection: 240 min (Rama Theepbhodi Hospital)

Target trait for low GI

- **Waxy b: Low Amylose**
- **Dietary fiber**



Rice	GI
Brown Sinlek	58
Purple Riceberry	62
White Sinlek	72
White Riceberry	74

RESEARCH PUBLICATIONS RELATED TO RICEBERRY

Whole grain

- GI/GL and insulin index
- Supply chain and value chain

Riceberry Bran

- Chemopreventive properties
- Phytosterol and triterpenoids: anti-cancer properties
- Functional properties of protein hydrolysate
- Attenuation of gentamicin-induced hepatotoxicity by reducing oxidative, Inflammation, apotosis in rat
- Preventing renal dysfunction in gentamicin-induced nephrotoxicity in rat
- Gramisterol: anti-tumor and immune enhancer in acute myelogenosis leukemia

Hiberry Bran Supplement

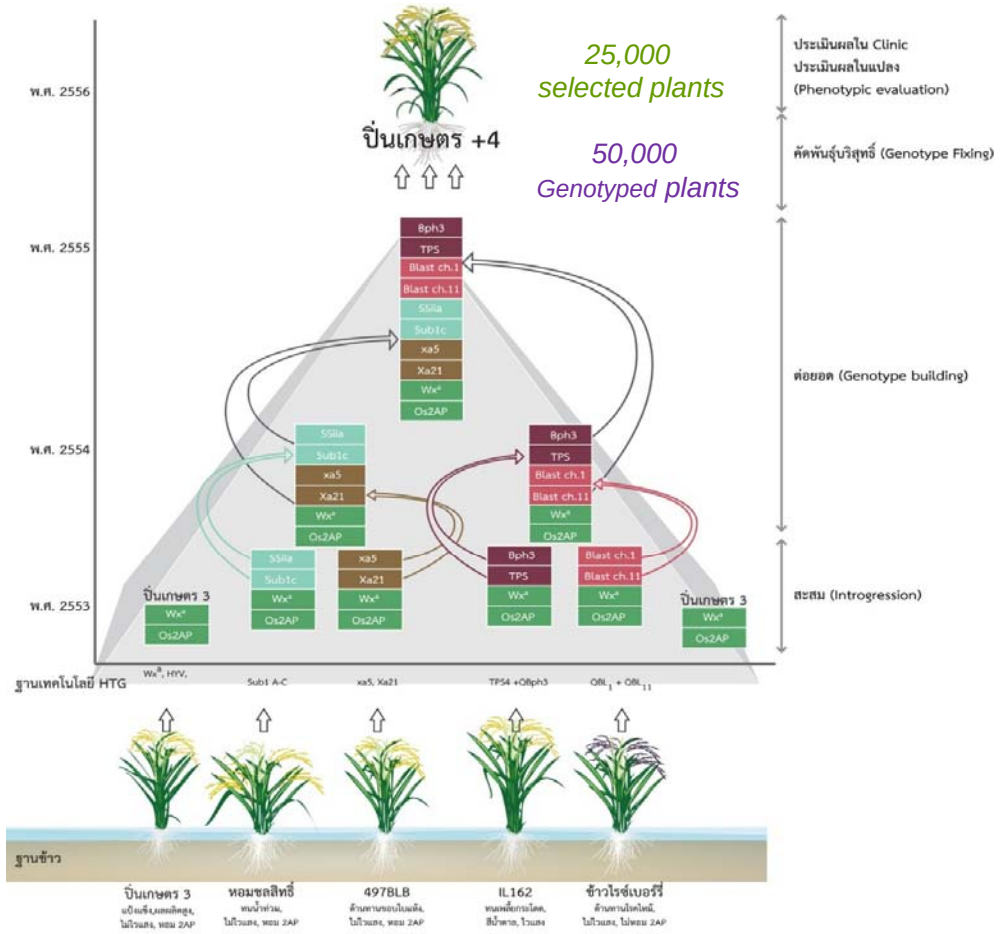
- Longterm supplementation for amelioration of oxidative stress in diabetic rats
- Hyperglycemia and change in lipid profile in diabetic rats

Polished rice

- GI/GL and insulin index

Second Generation MAS: Pyramiding

Gene Pyramiding Project



Target Genes for low GI

- **Waxy a: high Amylose**
- **SSII a: ACL =**
  Amylopectin Chain Length Distribution

Pinkaset+4
GI 55-52

Supanburi 1/DF=107 วัน

PPT 1/DF=111 วัน

PinK 3/DF=121 วัน

Pinkaset+4#66B09
Early/ DF=102 วัน

Pinkaset+44#16C03
Early/ DF=105 วัน

Pinkaset+4#46E05
Late/ DF=127 วัน

Pinkaset+4

ปิ่นเกษตร+4 #1E06

ความสูง(ถึงปลายใบธง)	115 ซม.	ต้นข้าวต่อไร่	630 ก.ก.
อายุเก็บเกี่ยว	140 วัน	ความยาวเมล็ดข้าวขัด	7.8 ม.ม.
ผลผลิตต่อไร่ (14% ความชื้น)	1244 ก.ก.	ลักษณะเด่น	ต้านทานเพลี้ยกระโดดสีน้ำตาลดีมาก

ปิ่นเกษตร+4 #20A09

ความสูง(ถึงปลายใบธง)	114 ซม.	ต้นข้าวต่อไร่	630 ก.ก.
อายุเก็บเกี่ยว	140 วัน	ความยาวเมล็ดข้าวขัด	7.8 ม.ม.
ผลผลิตต่อไร่ (14% ความชื้น)	1181 ก.ก.	ลักษณะเด่น	ต้านทานเพลี้ยกระโดดสีน้ำตาลดีมาก

ปิ่นเกษตร+4 #66B09

ความสูง(ถึงปลายใบธง)	127 ซม.	ต้นข้าวต่อไร่	527 ก.ก.
อายุเก็บเกี่ยว	126 วัน	ความยาวเมล็ดข้าวขัด	7.0 ม.ม.
ผลผลิตต่อไร่ (14% ความชื้น)	988 ก.ก.	ลักษณะเด่น	อายุเก็บเกี่ยวสั้น

ปิ่นเกษตร3 (สายพันธุ์รับที่ใช้ในการปรับปรุงพันธุ์)

ความสูง(ถึงปลายใบธง)	123 ซม.	ต้นข้าวต่อไร่	576 ก.ก.
อายุเก็บเกี่ยว	141 วัน	ความยาวเมล็ดข้าวขัด	7.4 ม.ม.
ผลผลิตต่อไร่ (14% ความชื้น)	1329 ก.ก.	(ปิ่นเกษตร3 เป็นสายพันธุ์รับที่ใช้ในการปรับปรุงพันธุ์)	

Nounmusing et al. 2018. The effect of low and high glycemic index based rice varieties in test meals on postprandial blood glucose, insulin and incretin hormones response in prediabetic subjects. IFRJ 25(2): 835-841.

Table 3. Maximum increase in plasma glucose (MIPG), glycemic response (GR) and glycemic index (GI)) for each test rice.

Test foods	MIPG	GR	GI	
	(mmol/L)	(mmol.min/L)	(%)	classification
Reference food (glucose)	3.1±1.7	178±41^{a}	100	High
Jasmine rice	2.4±0.6	164±28^{a}	90.7±12.0^{a}	High
Basmati rice variety	2.6±1.0	111±21^{ab}	66.2±8.0^{ab}	Medium
PK+4#1_E06	2.1±0.8	100±21^{b}	54.6±6.5^{b}	Low
PK+4#20A09	2.1±1.1	92±24^{b}	48.1±6.2^{b}	Low
PK+4#66B09	2.1±1.0	136±31^{ab}	66.1±11.0^{ab}	Medium
PK+4#117A08	2.0±0.9	114±25^{ab}	63.8±12.5^{ab}	Medium

GI of polished and brown rice of 15 Thai rice varieties.

Clinical Subjects for GI

1/ Using
2/ Using 12 pre-diabetic senior subjects
3/ Using 12 young healthy subjects

Rice Varieties	GI Polished	GI Brown	AC (%)	Gel Temp
Riceberry (Purple)[1/]	**74**	**62**	**15.6**	**low**
Sinlek	**72**	**58**	**16.5**	**low**
Sunkyod (Red)[2/]	75	63	15± 2.0	low
RD 15 (Waxy)	69	62	14-17	low
Surin Black Jasmin (Purple)	64	61	13.2	low
KDML 105	63	57	16.23±1.0	low
Pathumtani	63	57	14-18	low
RD 6 (Waxy)	62	61	5-7	low
Tubtim Chumpae (Red)	60	59	12.3	low-medium
Phitsanulok	59	65	17.3	low
RD43	58	57	18.82	low
Kradunkgha (Red)	57	54	22.7	low
Khoatahang	54	49	25	low
Luengpratiw 123	50	41	29-32	low-medium
Jekcheua	49	49	27.1	low
Pinkaset+4_1E_06	**55**	**<50**	**29.1±1.5**	**low**
Pinkaset+4 _20A09	**52**	**<50**	**27.2±2.2**	**low**

%AC = Amylose content
Gel temp = Gelatinization temp

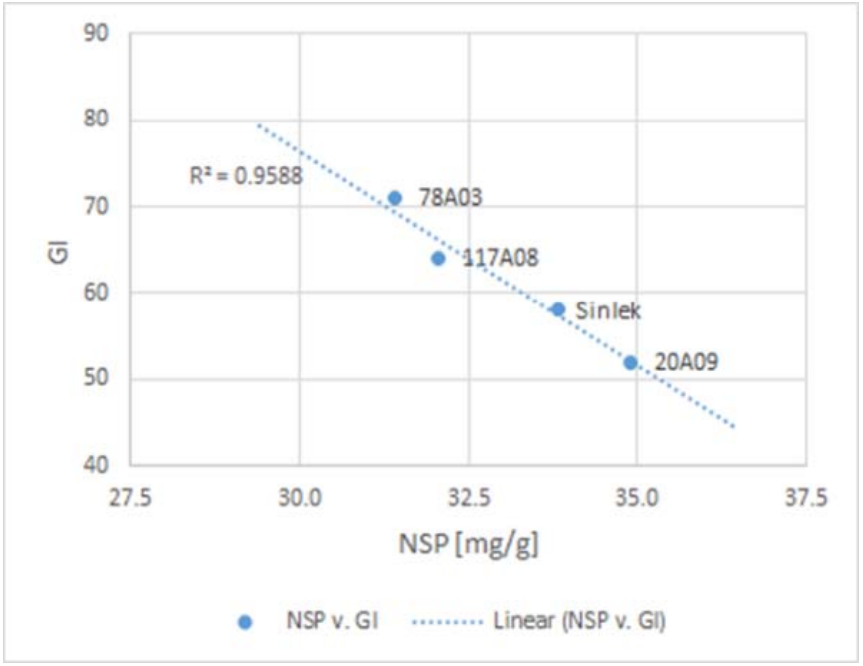
Comparison between GI studies on Pink+4 vs RD43

Comparison	Pink_4	RD43
Amylose content	Amylose 30%	Amylose 18-19%
Methodology	Young, healthy subjects	Elderly, DBII
GI of polished rice	48.1 – 54	57
GI of reference KDML105	90.7	63
RD6	-	62
Publication	Nounmusing et al., 2018	None

Can we really breed for soft-tender rice but low GI ?

- **Dietary Fiber Hypothesis**

Relationship between GI and dietary fibre (total non-starch polysaccharides determined as monosaccharides) in 4 Thai rice lines (Polished rice).



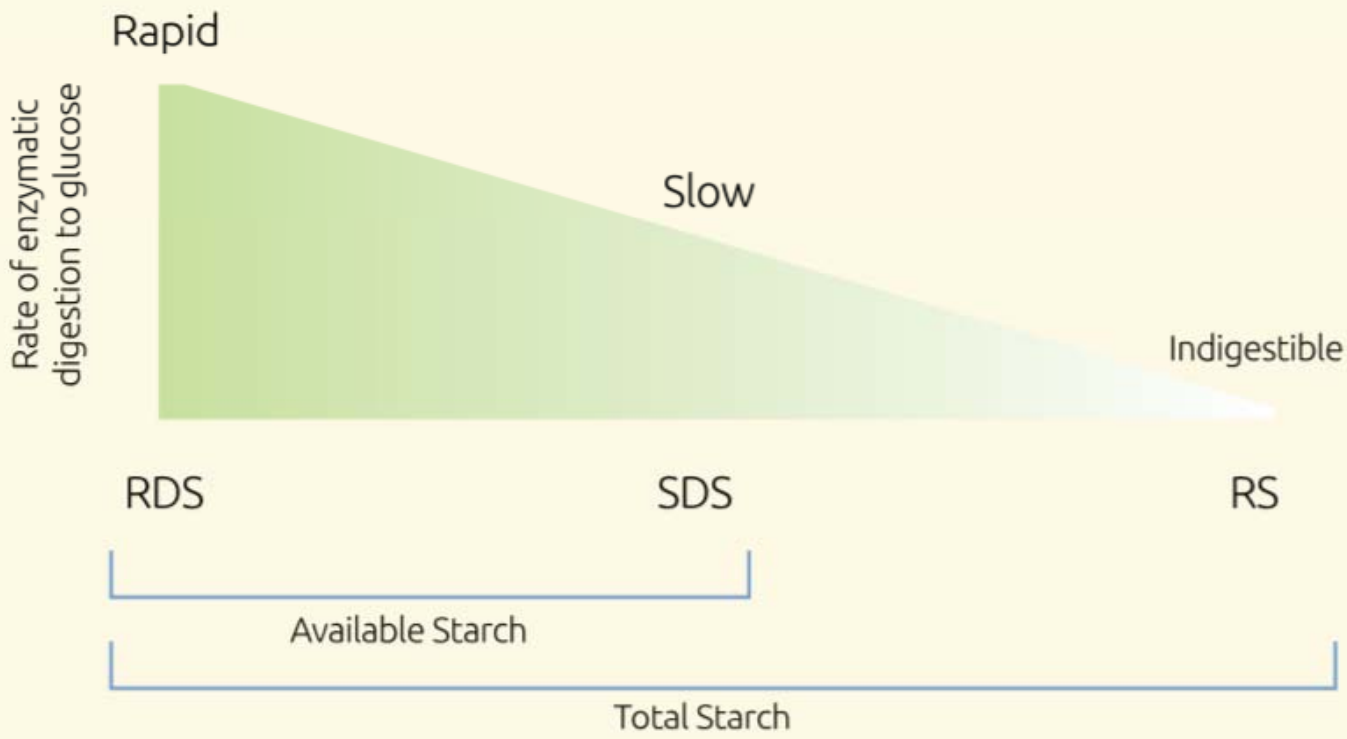
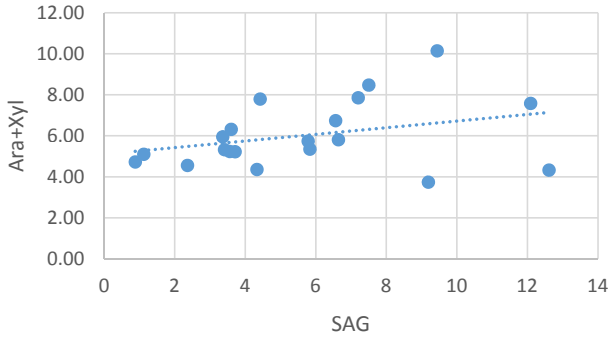


Figure 1:
Classification of starch by the rate of enzymatic digestion to glucose in the small intestine. RDS, rapidly digestible starch; SDS, slowly digestible starch; RS, resistant starch.

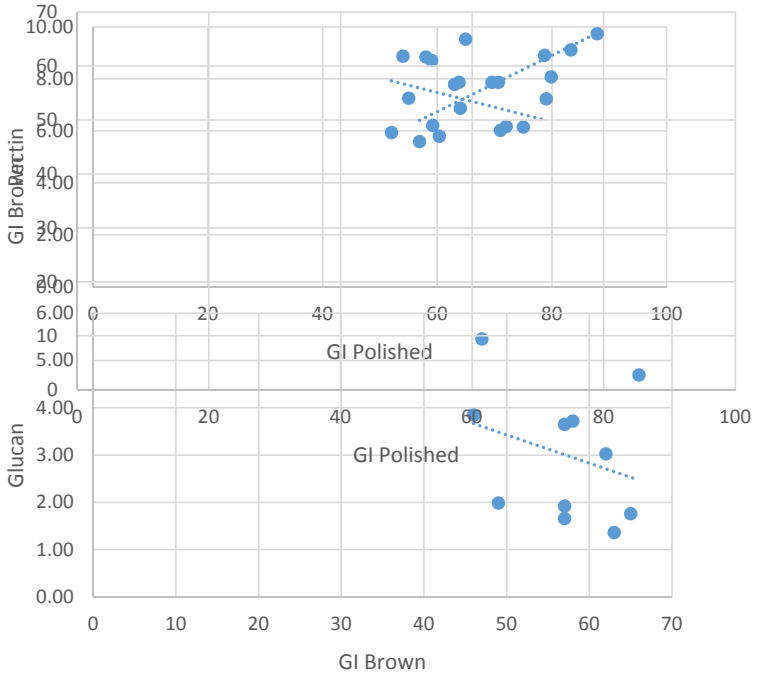
No.	Variety	RAG	SAG	Ara+Xyl	Pectin	Glucans
1	Pinkaset plus IV (117A08)	18.88	4.33	4.36	6.87	3.65
2	Pinkaset plus IV (1E06)	18.75	3.41	5.33	7.26	5.46
3	Pinkaset plus IV (20A09)	14.54	9.19	3.74	5.94	3.85
4	Pinkaset plus IV (66B09)	22.72	3.56	5.24	7.23	4.69
5	Pinkaset plus IV (78A03)	14.77	12.61	4.33	6.02	3.03
6	Pinkaset 3 (PK3)	20.75	12.09	7.59	8.85	1.78
7	Pinkaset plus IV_104A03	16.18	3.36	5.96	7.51	3
8	Pinkaset plus IV_105G03	20.22	3.71	5.23	6.74	1.68
9	Pinkaset plus IV_16C03	18.75	6.56	6.74	7.45	3.05
10	Pinkaset plus IV_19G11	13.84	5.83	5.35	6.69	2.81
11	Pinkaset plus IV_30A10	15.7	3.6	6.32	7.56	3.21
12	Pinkaset plus IV_33H04	16.57	9.44	10.15	10.11	2.54
13	Pinkaset plus IV_35A10	16.73	5.78	5.75	7.76	2.66
14	Pinkaset plus IV_37C11	23.41	7.5	8.48	8.85	1.59
15	Pinkaset plus IV_38C02	15.71	4.42	7.8	7.5	3.05
16	Pinkaset plus IV_46E_05	18.72	2.36	4.56	8.05	1.49
17	Pinkaset plus IV_60E_03	23.66	0.88	4.73	7.81	2.84
18	Pinkaset plus IV_83C02	15.8	1.12	5.1	6.66	3.5
19	Pinkaset plus IV_90A08	14.36	7.2	7.86	9.56	1.39
20	Sinlek	22.29	6.64	5.81	6.17	3.72

	RAG	*SAG*	*Ara+Xyl*	*Pectin*	*Glucans*
RAG	1				
SAG	-0.19609	1			
Ara+Xyl	0.027426	0.324907*	1		
Pectin	0.108452	0.152922	0.815192**	1	
Glucans	0.011796	-0.22989	-0.39858	-0.53199*	1



No.	Variety	GI Polished	GI Brown	Ara+Xyl	Pectin	Glucans
1	RD43	58	57	6.34	8.85	1.66
2	KDML105	63	57	9.34	7.79	1.92
3	Khoatahang	54	49	6.43	8.88	1.99
4	Pinkaset plus IV (117A08)	64	57	4.36	6.87	3.65
5	Pinkaset plus IV (1E06)	55	47	5.33	7.26	5.46
6	Pinkaset plus IV (20A09)	52	46	3.74	5.94	3.85
7	Pinkaset plus IV (66B09)	79	66	5.24	7.23	4.69
8	Pinkaset plus IV (78A03)	71	62	4.33	6.02	3.03
9	Phitsanulok 80	59	65	7.27	8.71	1.76
10	Sunkyod	75	63	3.91	6.15	1.36
11	Sinlek	72	58	5.81	6.17	3.72

	GI Polished	*GI Brown*	*Ara+Xyl*	*Pectin*	*Glucans*
GI Polished	1				
GI Brown	0.78446964**	1			
Ara+Xyl	-0.203931384	0.088962	1		
Pectin	-0.447557998*	-0.01158	0.680478	1	
Glucans	0.051850413	-0.30609*	-0.38273	-0.41361	1



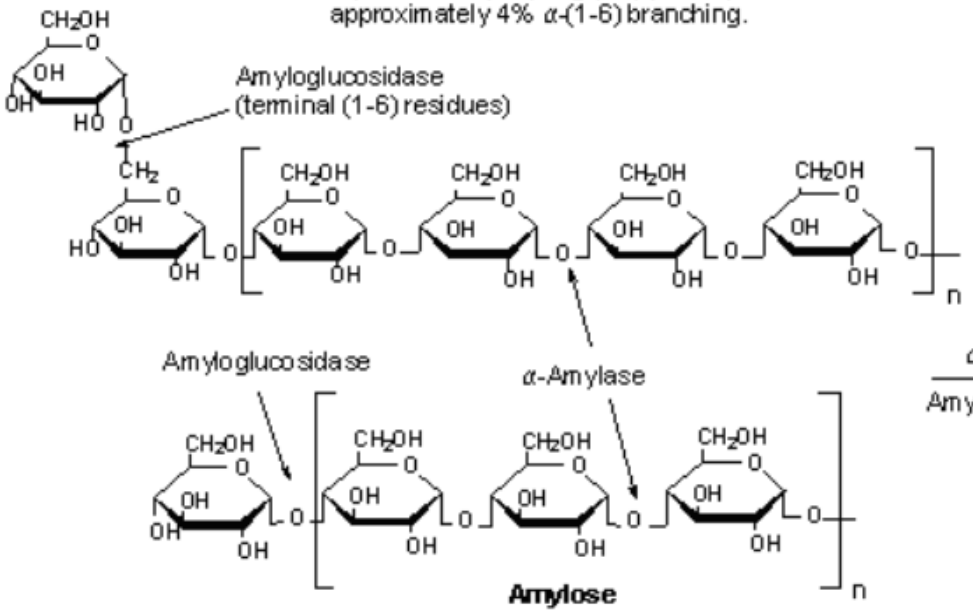
Can we really breed for soft-tender rice but low GI ?

- **Endogenous Amylase Inhibitor Hypothesis**

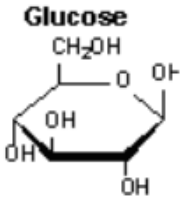
Starch

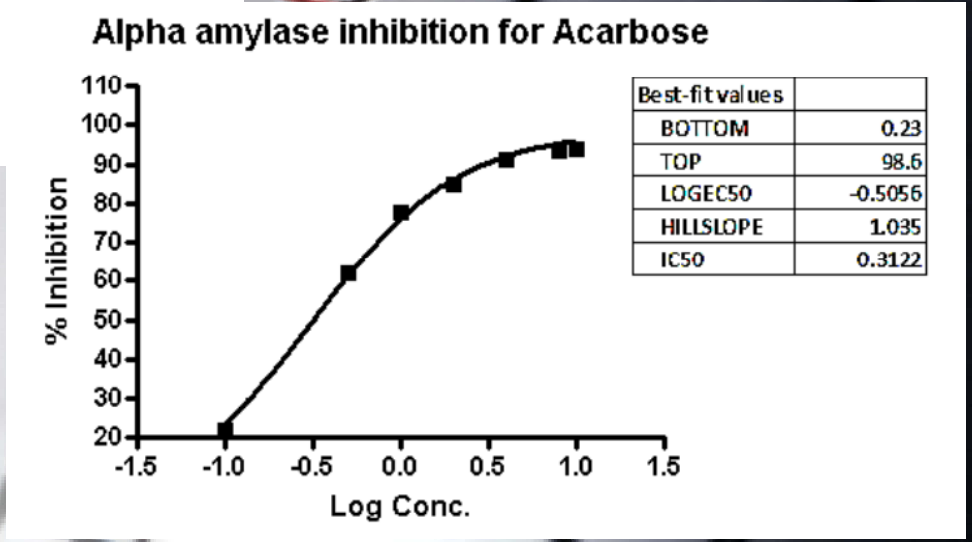
Amylopectin

Polymers of α-(1-4)-D-glycopyranosyl units with approximately 4% α-(1-6) branching.



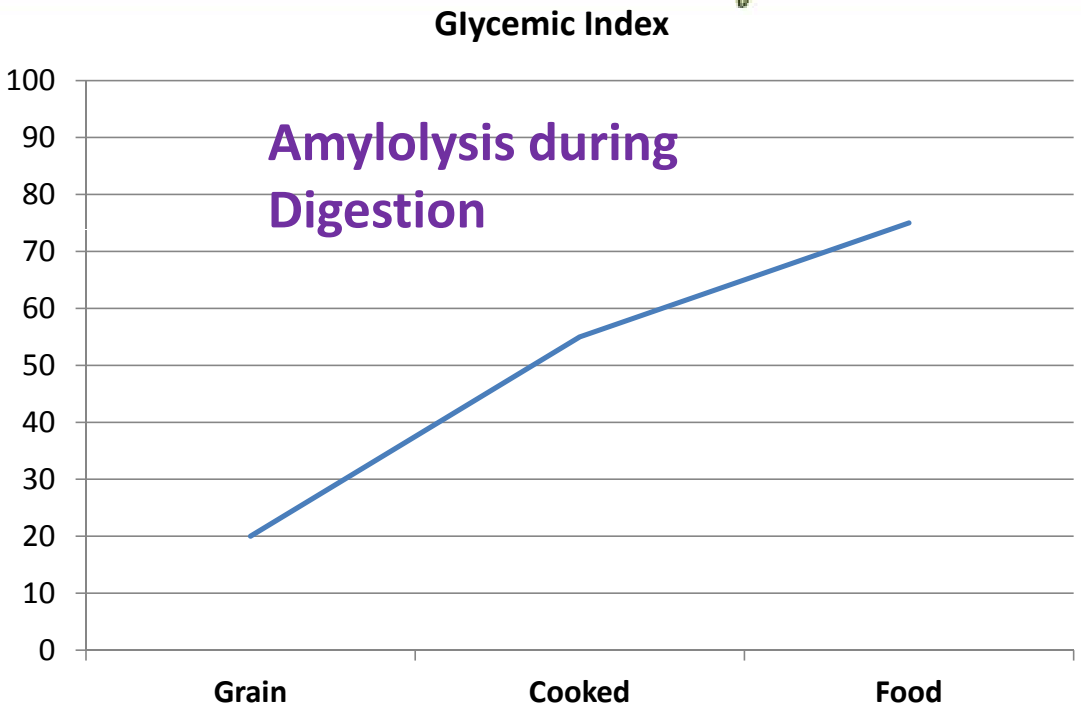
α-Amylase
Amyloglucosidase





Best-fit values	
BOTTOM	0.23
TOP	98.6
LOGEC50	-0.5056
HILLSLOPE	1.035
IC50	0.3122

Relationship between GI and cooked rice and food processing



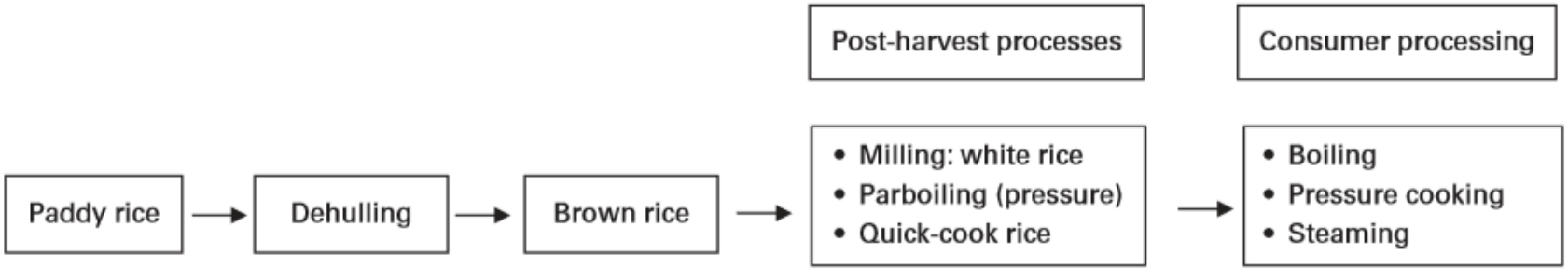


Table 4. Correlation among GI, RS, AAC, AAIA and IR parameters

Pearson Correlation	GI	RS	AAC	AAIA	IR
GI	1	-0.279	-0.796*	-0.057	-0.789*
RS		1	-0.071	0.836**	-0.274
AAC			1	-0.005	0.787*
AAIA				1	-0.284
IR					1

SHOZIB, H.B.[1*], HOSSAIN, M.M.[2], JAHAN, S.[3], ALAM, M.S.[4], DAS, S.C.[4], ALAM, S.[4], AMIN, R.B.[4], HASAN, M.M.[4], MALO, R.[6], ISLAM, M.R.[5], SHEKHAR, H.U.[2] and SIDDIQUEE, M.A.[1]

GI=Glycemic index
RS=Resistance starch
AAC=Apparent amylose content
AAIA=Alpha amylase inhibitory activities
IR=Imbibition ratio

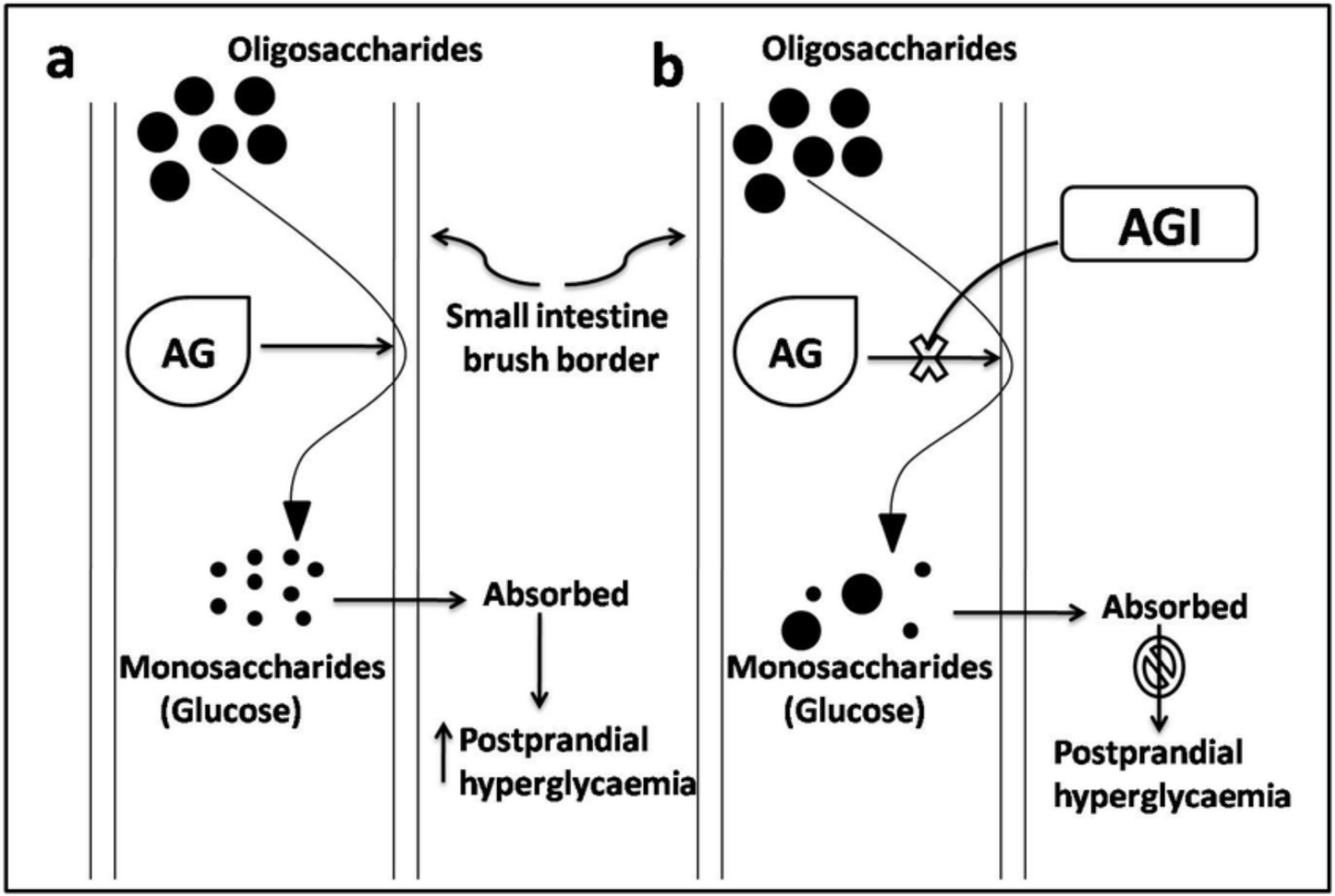
Variety	Glycemic Index (GI)	Alpha Amylase inhibitory activity (µg/g AE)	Apparent amylose content (AAC%)	Resistant starch (%)
BR16	52.4^{f}	122.0^{a}	27.0^{ab}	4.68^{a}
BRRI Dhan28	70.8^{bc}	11.3^{g}	28.0^{a}	1.87^{b}
BRRI Dhan33	66.0^{e}	58.0^{d}	25.0^{c}	2.49^{b}
BRRI Dhan47	67.0^{d}	75.0^{c}	26.0^{bc}	2.82^{b}
BRRI Dhan49	71.5^{b}	15.7^{f}	25.0^{c}	1.69^{b}
BRRI Dhan50	69.3^{c}	92.6^{b}	27.0^{ab}	2.27^{b}
BRRI Dhan53	77.3^{a}	28.7^{e}	21.0^{d}	2.39^{b}
BRRI Dhan62	63.1^{e}	92.0^{b}	19.0^{e}	3.27^{ab}

Rice samples are in clean rice condition. Any two means having common letter (s) are not statistically different at $P< 0.05$, as measured by the Duncan Multiple Range Test (DMRT).

Amylolysis during Digestion

AG=alpha glucosidase

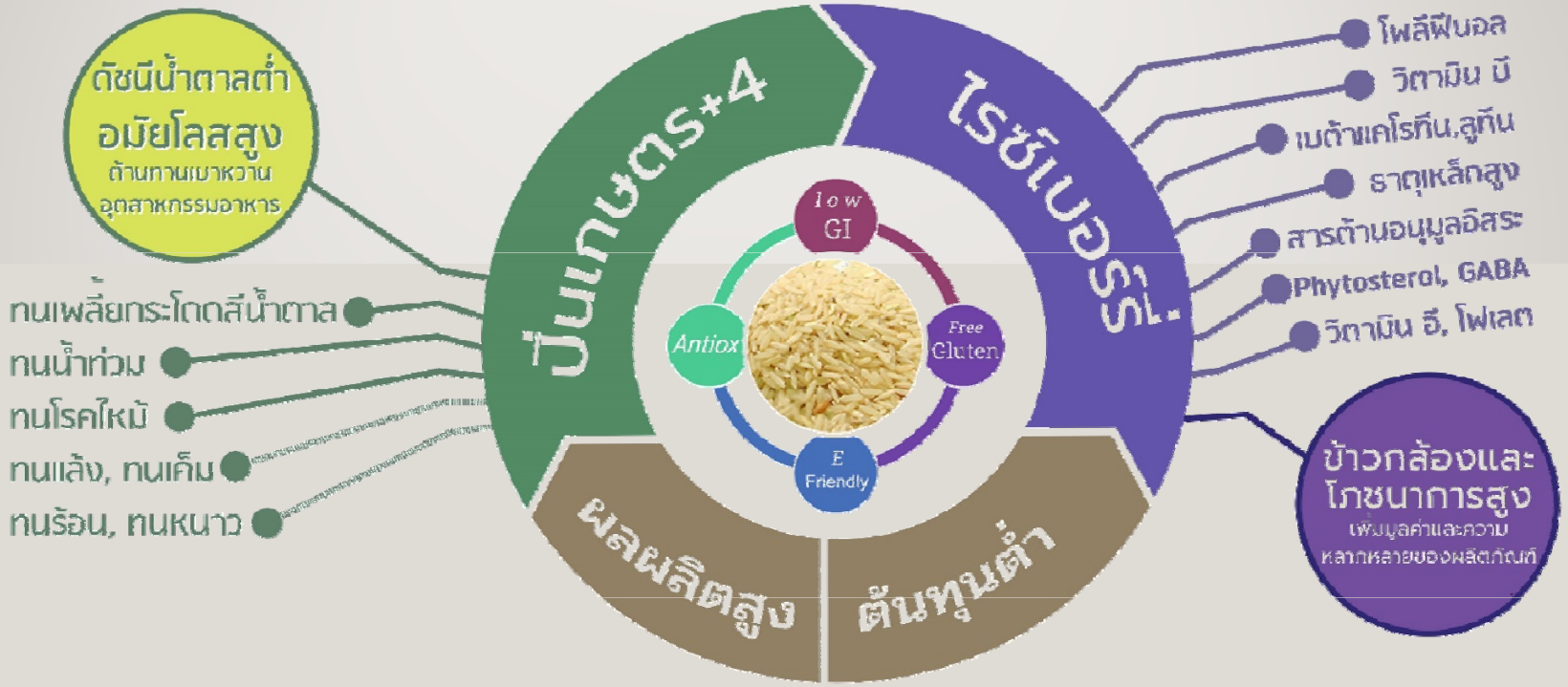
AGI=AG inhibitor



High Temperature-Induced Expression of Rice α-Amylases in Developing Endosperm Produces Chalky Grains

Nutritional Factors Contributing to GI/GL and Insulinotropic Effects

Nutrition Factors	Polished	Whole Grain
Alpha Amylase activities	+	++
Polyphenol/Tannin	-	++
Glycemic index	+	++
Dietary fiber	+	++
Resistance starch	+	++
Protein/Peptide	+	++
Fat	-	++
Flavonoids	-	++
Gelatinization temperature	+	++
Prebiotic	-	++


ดัชนีน้ำตาลต่ำ
อมัยโลสสูง
ต้านทานเบาหวาน
อุตสาหกรรมอาหาร
ทนเพลี้ยกระโดดสีน้ำตาล
ทนน้ำท่วม
ทนโรคไหม้
ทนแล้ง, ทนเค็ม
ทนร้อน, ทนหนาว
ปีนเกษตร*4
ไรซ์เบอร์รี่
low GI
Antiox
Free Gluten
E Friendly
ผลผลิตสูง
ต้นทุนต่ำ
โพลีฟีนอล
วิตามิน บี
เบต้าแคโรทีน,ลูทีน
ธาตุเหล็กสูง
สารต้านอนุมูลอิสระ
Phytosterol, GABA
วิตามิน อี, โฟเลต
ข้าวกล้องและ
โภชนาการสูง
เพิ่มมูลค่าและความ
หลากหลายของผลิตภัณฑ์


Riceberry