Basic applied aspects of vegetable breeding & seed production

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AVRDC
The World Vegetable Center

Vegetable?

An edible part (as root, tuber, stem, leaves inflorescence, fruit or seed) that is used as human food and usually eaten cooked or raw during the principle part of a meal rather than as a dessert – contrasted with fruit

Watermelon is also a vegetable?

It is planted from seeds/seedlings, harvested, and then cleared from the field like other vegetables. Since watermelon is grown as a vegetable crop using vegetable production systems, watermelon is considered a vegetable

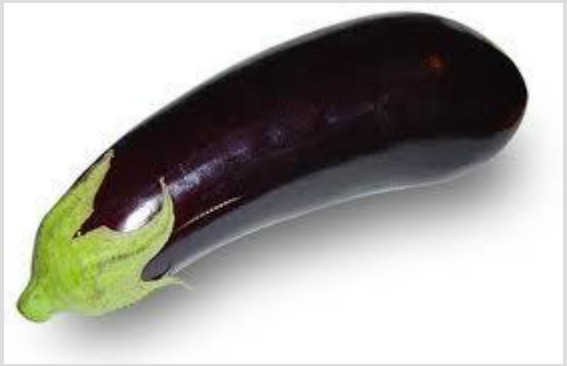
Plant breeding vs Vegetable breeding

Plant breeding is the art, science and business of improving plants for human benefit

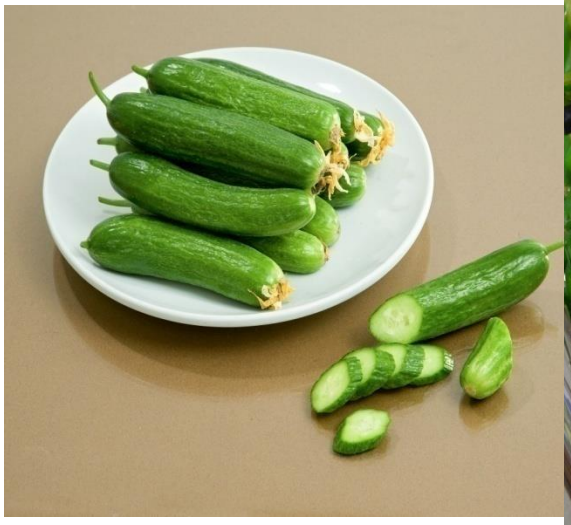
How vegetable breeding differ from crop breeding?

- Wide choice of different species
- Range of traits
- Local preferences
- Increasing breeding capacity with private-sector seed companies

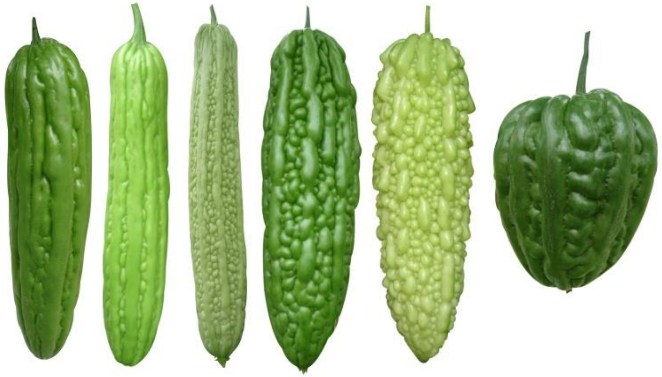
Consumer preference



Cucumber market types



Thailand type



Chinese type

Taiwan type

Okinawa type

Vietnam type

Philippine type

South Asian types

Indonesia type

Bitter gourd market types

Pumpkin market types

Pumpkins in S. Korea

Melon market types

Carrot market types

When is a breeding program needed?

A breeding program is needed if current varieties are not producing up to the capacity of the environment

Sponge gourd: Resistance to begomovirus

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Cucumber: Resistance to *Zucchini yellow mosaic virus*

Bitter gourd: Resistance to powdery mildew

Bottle gourd: Resistance to ZYMV

Chili pepper: Resistance to anthracnose

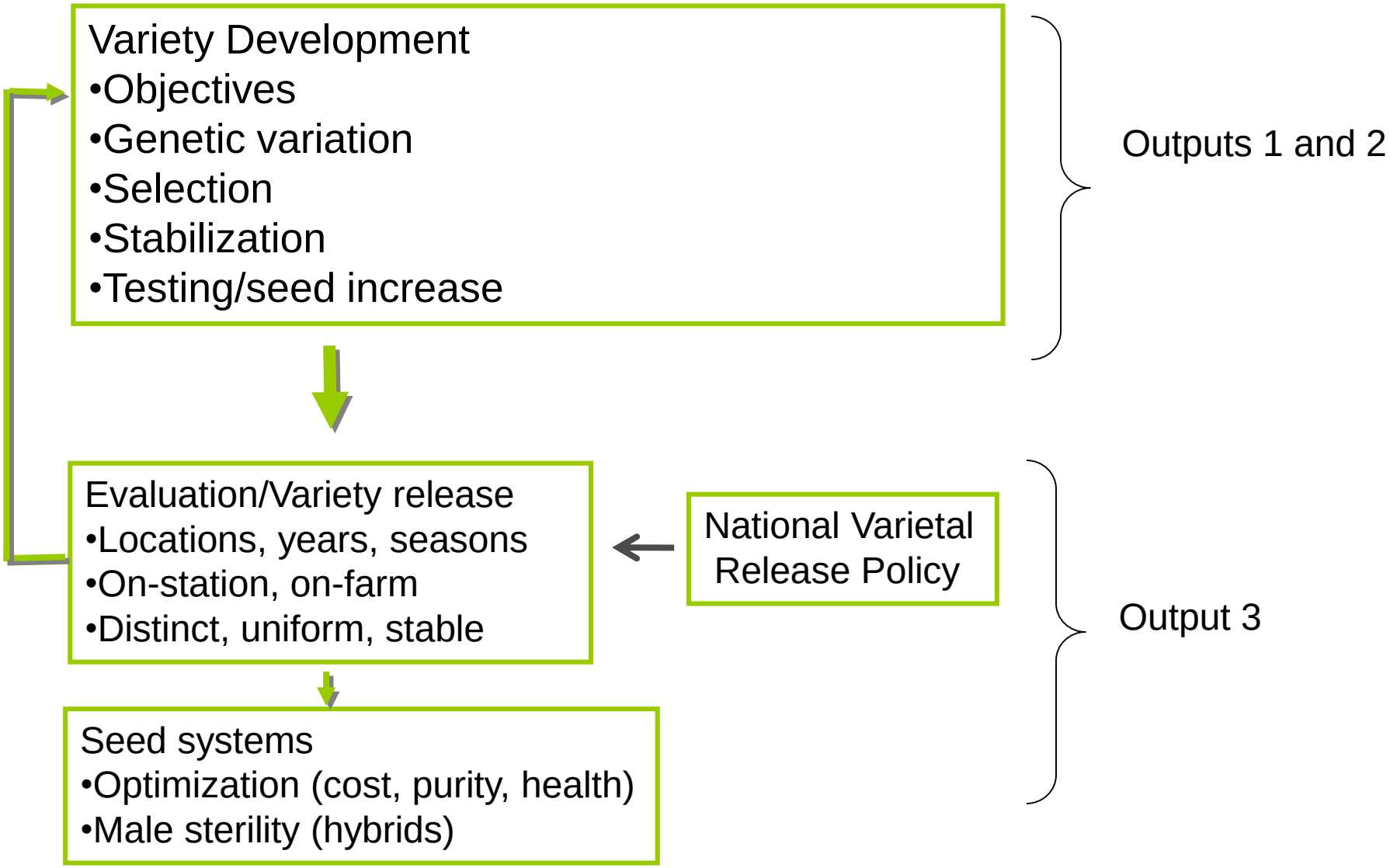
Chili pepper: Resistance to leaf curl virus

Tomato: Resistance to *Tomato yellow leaf curl virus*

Types of cultivars

- Pure line
- F_1 hybrids
- Open pollinated
- Clones

Relation of Breeding Outputs



Methods of vegetable breeding

Introduction

Line breeding

Population breeding

Hybrid breeding

Clone breeding

Note: Mode of reproduction is the deciding factor to develop suitable breeding and selection methods.

Phenotype vs. Genotype

$$P = G + E + (GxE)$$

P is called the **phenotypic value**, i.e., the measurement associated with a particular individual

G is genotypic value, the effect of the genotype (averaged across all environments)

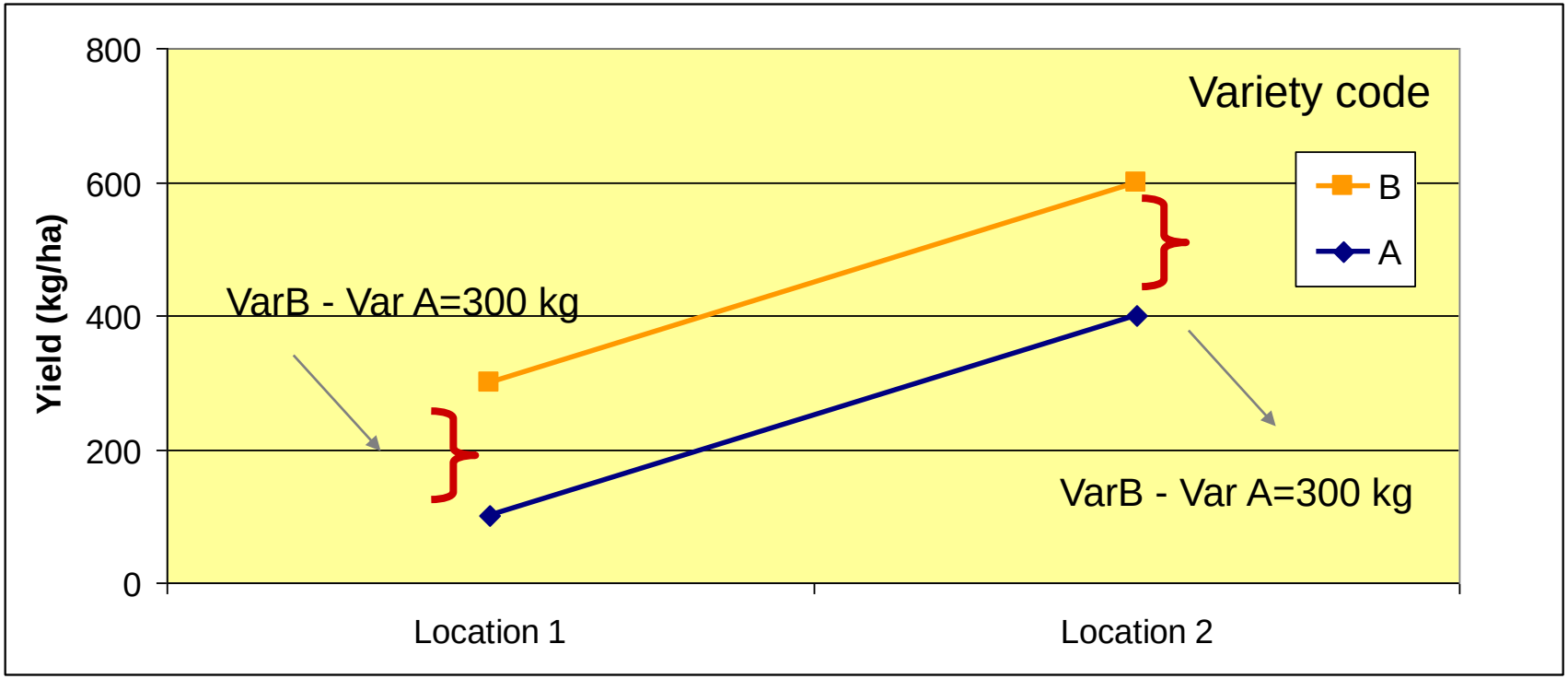
E is the effect of the environment (averaged across all genotypes)

G x E ?

When different genotypes exhibit differential responses to different sets of environmental conditions, a genotype x environment (G x E) interaction is said to occur

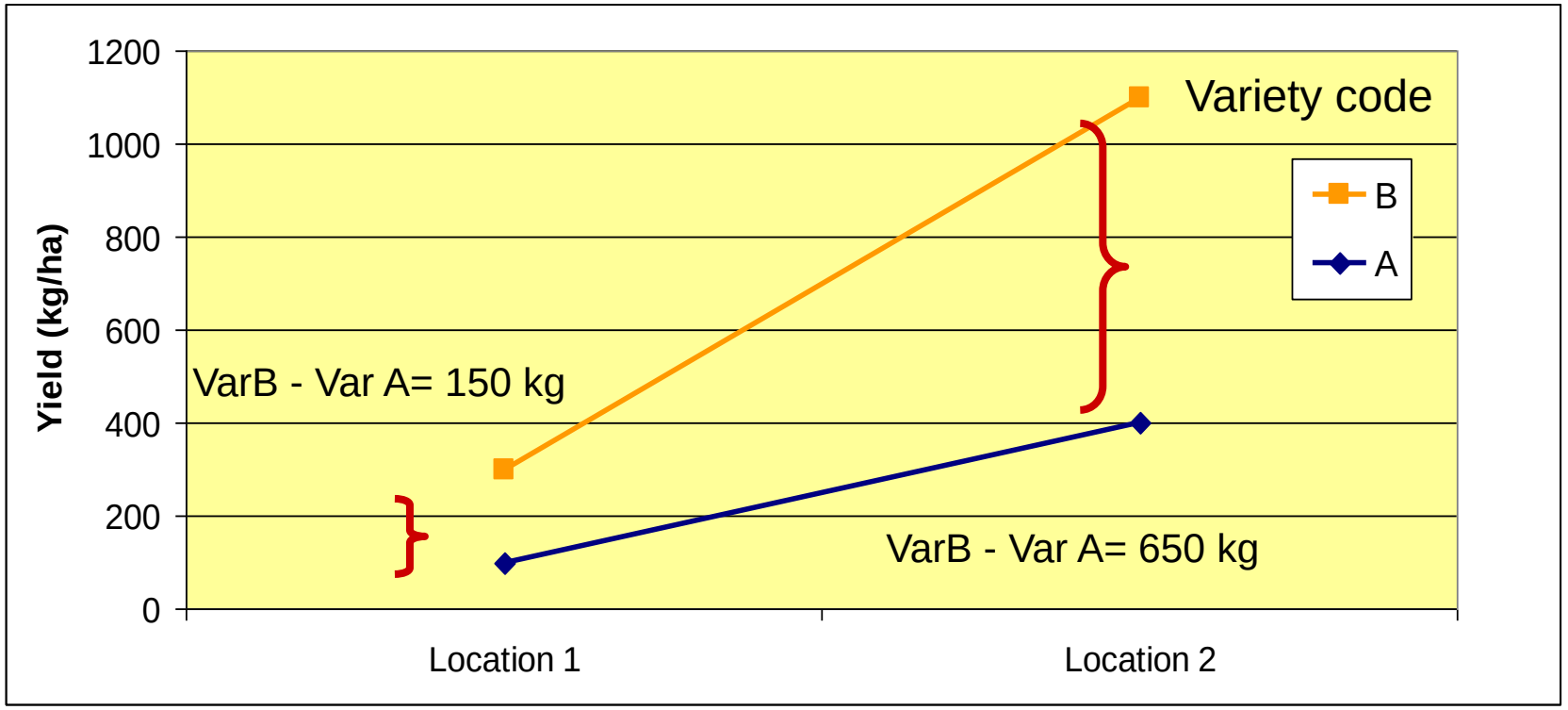
No Genotype-Environment Interaction

- Variety ranking is same between locations 1 and 2
- Size of the difference between varieties is the same at both locations



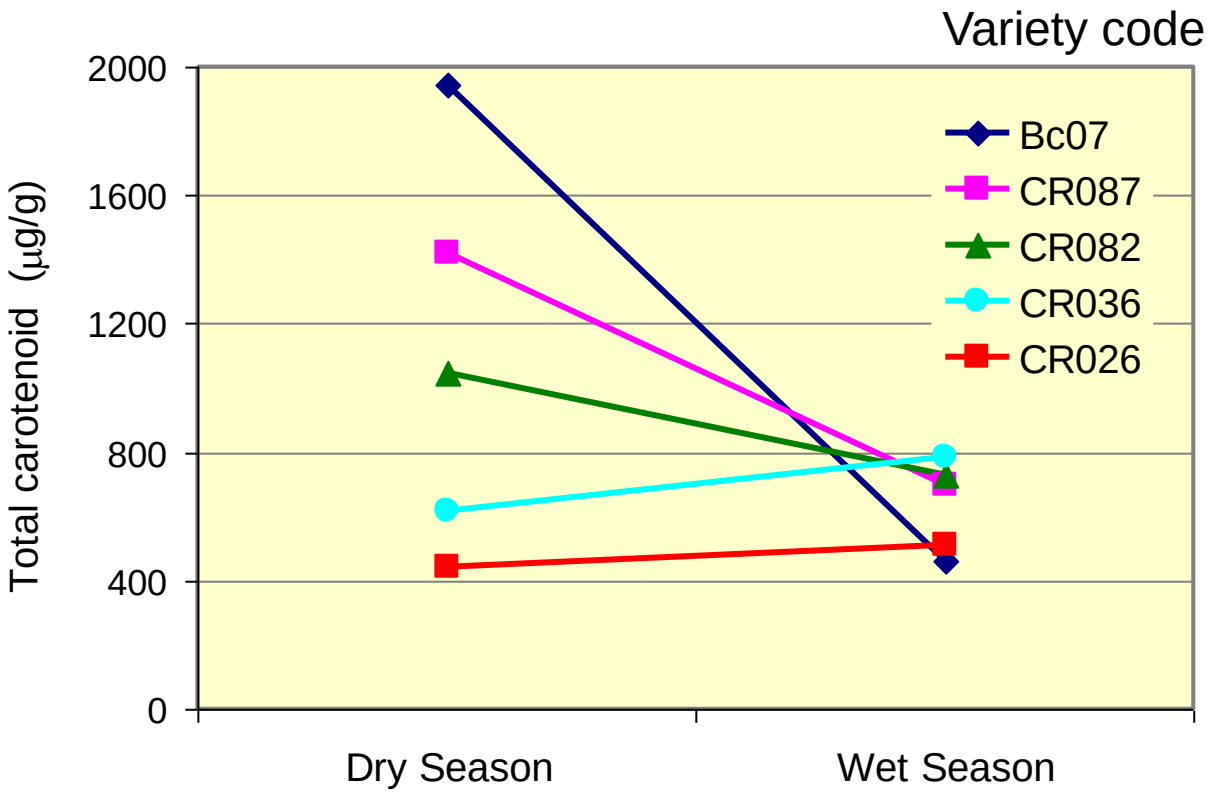
GxE Interaction Present: Case 1

- Variety ranking is same between locations 1 and 2
- Yield difference between varieties varies greatly between locations



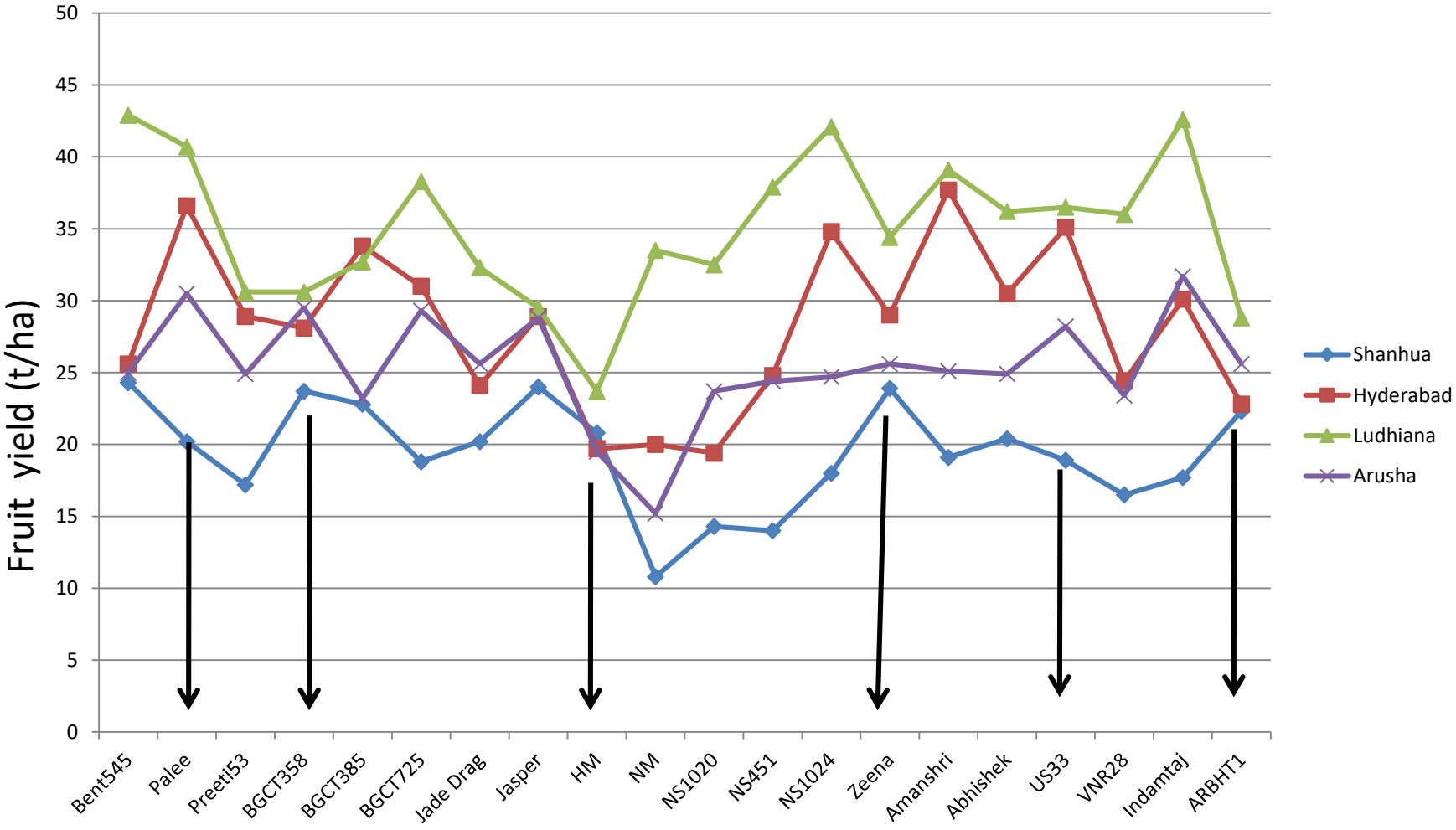
GXE Interaction Case 2:

- Total Carotenoid Contents of choysum (*Brassica rapa* cvg. *parachinensis*) in dry and wet seasons



- Variety rank changes between seasons
- Changes in the magnitude of variety differences between seasons

Bitter gourd: Multi-location trial



ANOVA

Variation	Degree of freedom	Mean square	Computed F
Year (Y)	y - 1	MS 1	MS 1/MS 2
Rep/year	y (r – 1)	MS 2	
Genotypes (G)	g - 1	MS 3	MS 3/MS 4
Y x G	(y – 1)(g – 1)	MS 4	MS 4/EMS
Pooled error	y (r – 1)(g – 1)	EMS	
Total	yrg - 1		

Conclusion for the breeder

A cultivar that is responsive to the environment may be released for a narrowly defined area of adaptation, whereas another that has a low G x E interaction may be suitable for release for use over a wider region of production

Planning Variety Trials

- “ Too little time and effort is put into the planning of experiments.”
- Cochran and Cox, 1957

Varieties

- **Varieties differ in yield potential, adaptation, disease/pest resistance, quality, nutrient content**
- **From many potential varieties we want to identify those varieties that consistently demonstrate outstanding performance and broad adaptation for traits of interest over a wide range of environments**
- **Environments can mean different locations, years, seasons**

No Variety is the Best Everywhere

Variety A-Location 1

Variety A- Location 2

- Variety performance affected by temperatures, rainfall patterns, soil types, dominant diseases and insects, crop management in different environments

Objective of Variety Trials

- Methodical and unbiased comparison of varieties versus farmer-preferred varieties (checks) in target environments
- Proper choice of experimental design, checks, protocols is critical
- Basis for science-based recommendations

Define Target Environment

- Agroecology
  - Humid lowland tropics, Semi-arid tropics, Cool highlands
  - Red versus black soil
- Open field versus protected cultivation
- Season: dry, wet, ‘regular’ versus ‘off-season’

Protected cultivation

Open-field, low input

Define Target Farmer Group

- Market gardeners, commercial growers, processors
- Access to labor and credit, willingness/ability to buy inputs (fertilizer, pesticides, irrigation, seed), access to markets

Trial Sites

- Trial sites should be representative of the target environment (major soil types, altitudes, seasons, temperatures)

- Plan trial sowing and transplanting dates appropriately for target season

Protected cultivation

Open field

Conducting the Trial

- **Choose trial sites representative of the target environment**
- **A uniform field is preferred for the trial - soil fertility, soil texture, water-holding capacity, slope**
- **If field is not uniform, identify sources of variation and choose an experimental design that can help reduce “noise” from the variation**
- **Use the simplest possible statistical design with replication and randomization**
- **Consult with statistician BEFORE the experiment about plot sizes, arrangement of blocking, sample sizes**

Crop Management

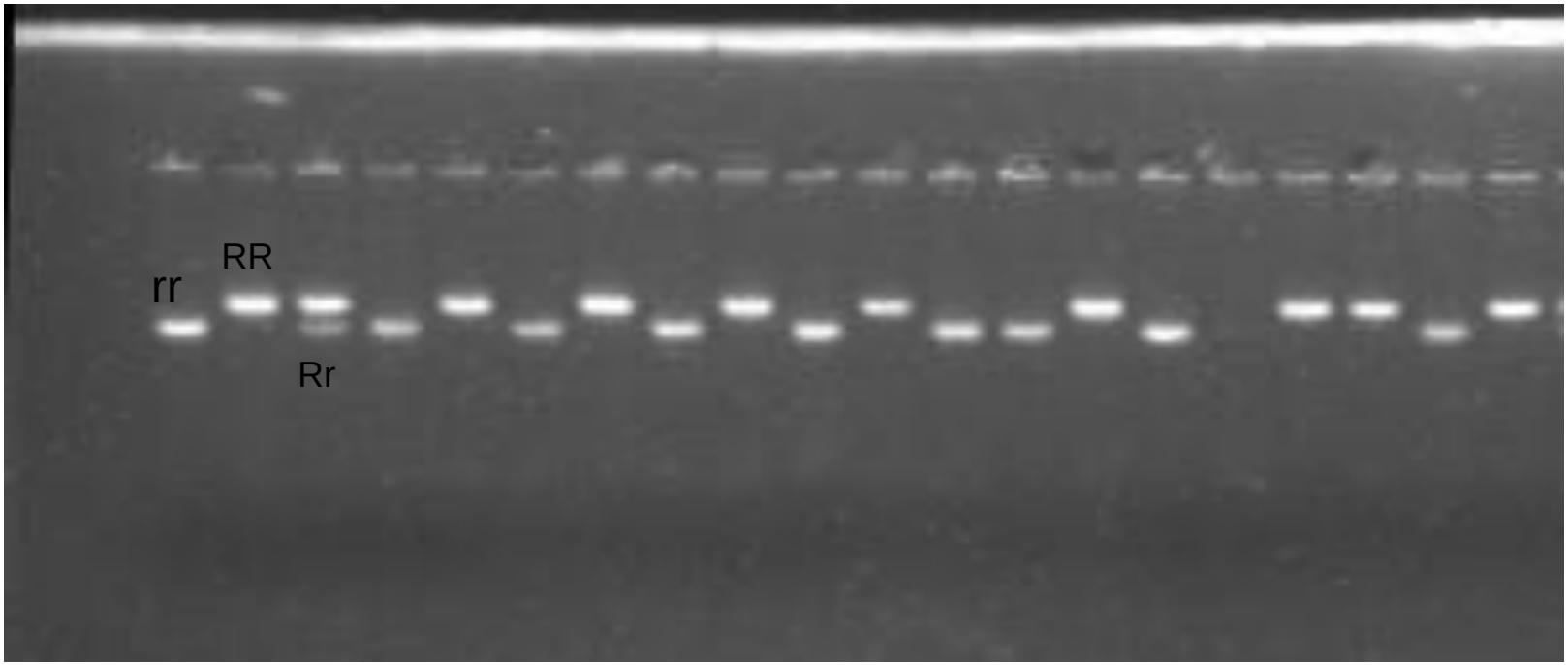
- Rule of thumb is to use practices of local progressive farmers
  - Fertilizer types, amounts, timing of application
  - Irrigated or rain-fed
  - Trellising
  - Disease, insect control methods

Major issues of vegetable breeding

1. Productivity
2. Shelf life
3. Quality traits
4. Disease and pest resistance
5. Wide adaptations
6. Tolerance to abiotic stresses

Molecular marker

genotype visualization



By migrating DNA in an electric field,we can now ‘see’ genotypes

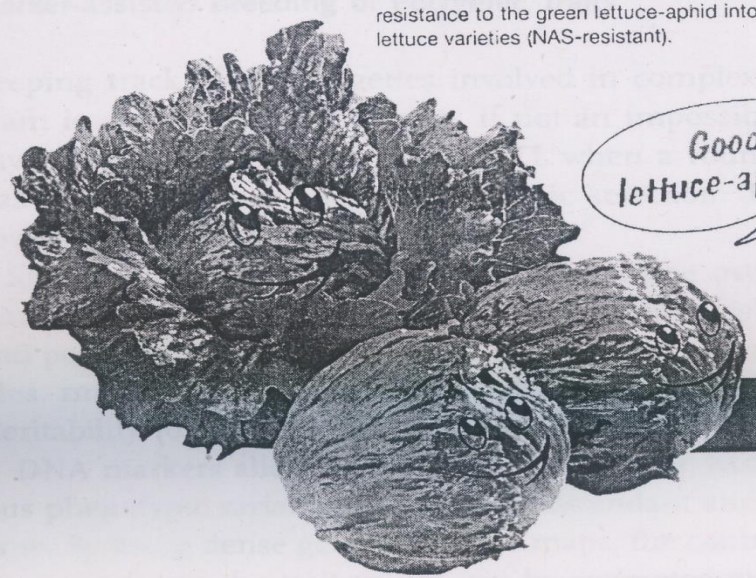
Molecular breeding – classical example

RZ-Info

THE FIRST NASANOVIA APHID-FREE ICEBERG LETTUCE!

This is good news, not just for true vegetarians but for all consumers, as the annoying aphid problem in iceberg lettuce has been solved once and for all.

RIJK ZWAAN is the first seed breeding company in the world to have succeeded in crossbreeding resistance to the green lettuce-aphid into iceberg lettuce varieties (NAS-resistant).



WORLD-FIRST

ICEBERG LETTUCE FORTUNAS RZ

Principles of vegetable seed production

Seed?

Reasons for the deterioration of variety

1. Developmental variations
2. Mechanical mixtures
3. Mutations
4. Natural crossing
5. Minor genetic variation
6. Selective influence of diseases
7. Technique of the breeder

Maintenance of genetic purity of seed

1. Control of seed source

    i. Breeder's seed

    ii. Foundation seed

    iii Registered seed

    iv Certified seed

2. Crop rotation

3. Isolation

    i. Isolation by time

    ii. Isolation by distance

Maintenance of genetic purity – contd.

3. Roguing of seed crop

    i. Vegetative stage

    ii. Flowering stage

    iii. Maturity stage

4. Seed certification

5. Grow-out tests

Minimum isolation distance requirements for vegetable seed crop

Vegetable	Isolation distance (m)		Vegetable	Isolation distance (m)	
	BS/FS	CS		BS/FS	CS
French bean	10	5	Peppers	400	200
Cow pea	10	5	Okra	400	200
Garden pea	10	5	Brinjal	200	100
Lettuce	50	25	Tomato	50	25
Cole crops, Chinese cabbage	1600	1000	Spinach	1600	1000
Carrot	1000	800	Potato	5	5
Radish/Turnip	1600	1000	Garlic	5	5
Onion	1000	500			
Cucurbits	1000	500			

Agronomic principles for seed production

1. Selection of suitable areas for seed production
2. Selection of variety
3. Source of seed
4. Seed treatment
5. Better agronomic management
6. Supplementary pollination
7. Harvesting, drying & storage

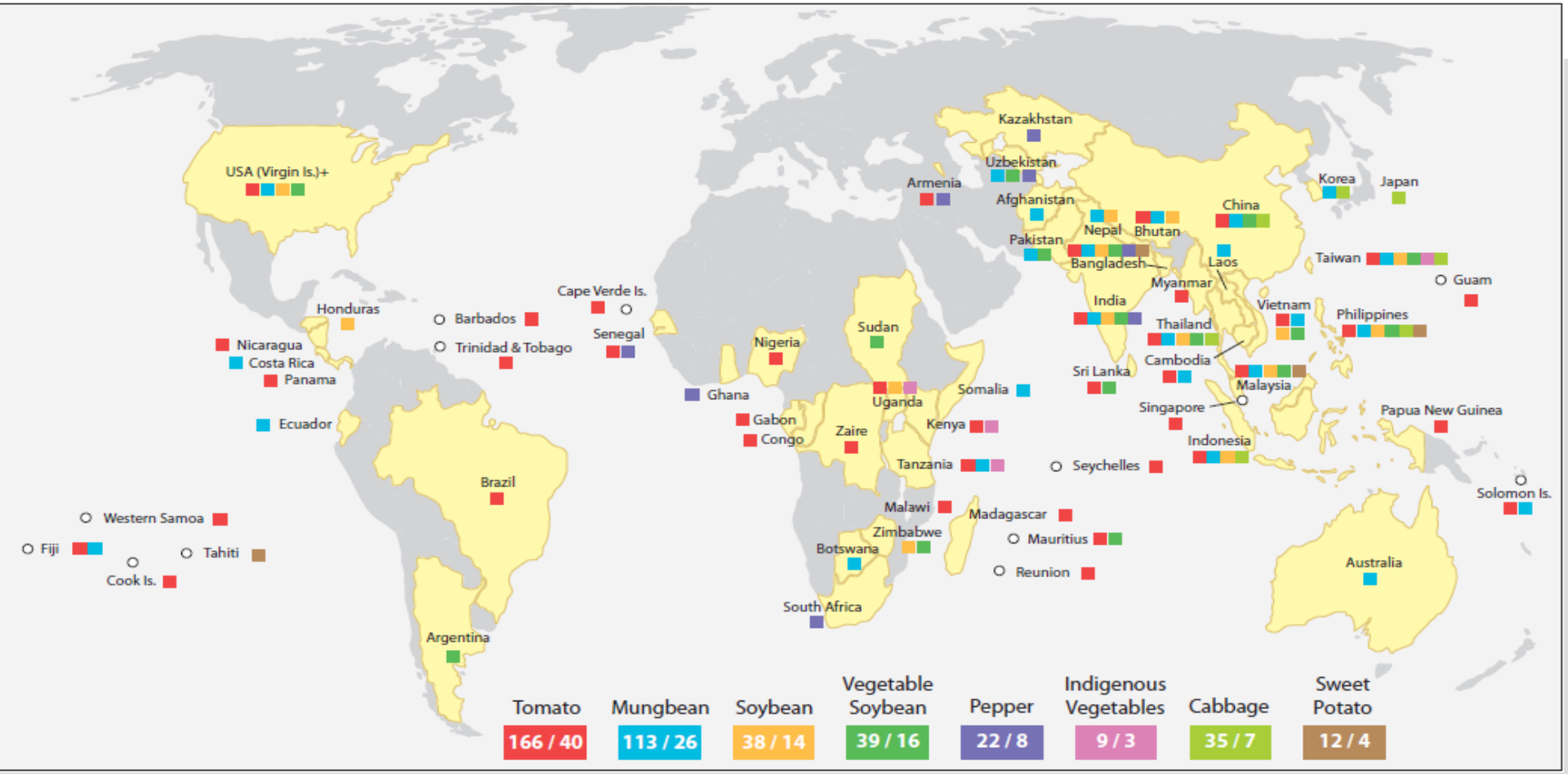
Labels for various seed classes

AVRDC's genebank

61,494 accessions from

- 439 species
- 172 genera
- 155 countries

Cultivars released since 1978



434 improved vegetable varieties benefit farmers around the world

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Current fresh market tomato releases of AVRDC

AVTO9601

AVTO9601

AVTO9803

AVTO1001

AVTO1002

AVTO1130

Current high beta carotene tomato lines of AVRDC

AVTO1017

AVTO1016

AVTO1015

AVTO1020

AVTO1019

AVTO0102

Current hot pepper releases of AVRDC

AVPP9905

AVPP0506

AVPP9813

AVPP0105

AVPP0206

AVPP0514

Current sweet pepper releases of AVRDC

AVPP0504

AVPP0402

AVPP9807

AVPP0408

AVPP0701

Soybean releases of AVRDC

AVSB0301

VI060637

VI060636

AVSB0805

AVSB0803

AVSB0304

Mungbean breeding program of AVRDC

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Cucurbit breeding at the World vegetable Center






World annual production of cucurbits

Vegetable	World production per annum (million)	
	Tonnes	Hectares
Watermelon	105.37	3.472
Cucumbers, Gherkins	65.13	2.109
Melon, cantaloupe	31.92	1.339
Pumpkins, squash, gourds	24.61	1.778
Total cucurbits	**227.03**	**8.698**
Tomatoes	161.79	4.803
Chilies, peppers, green	31.17	1.914

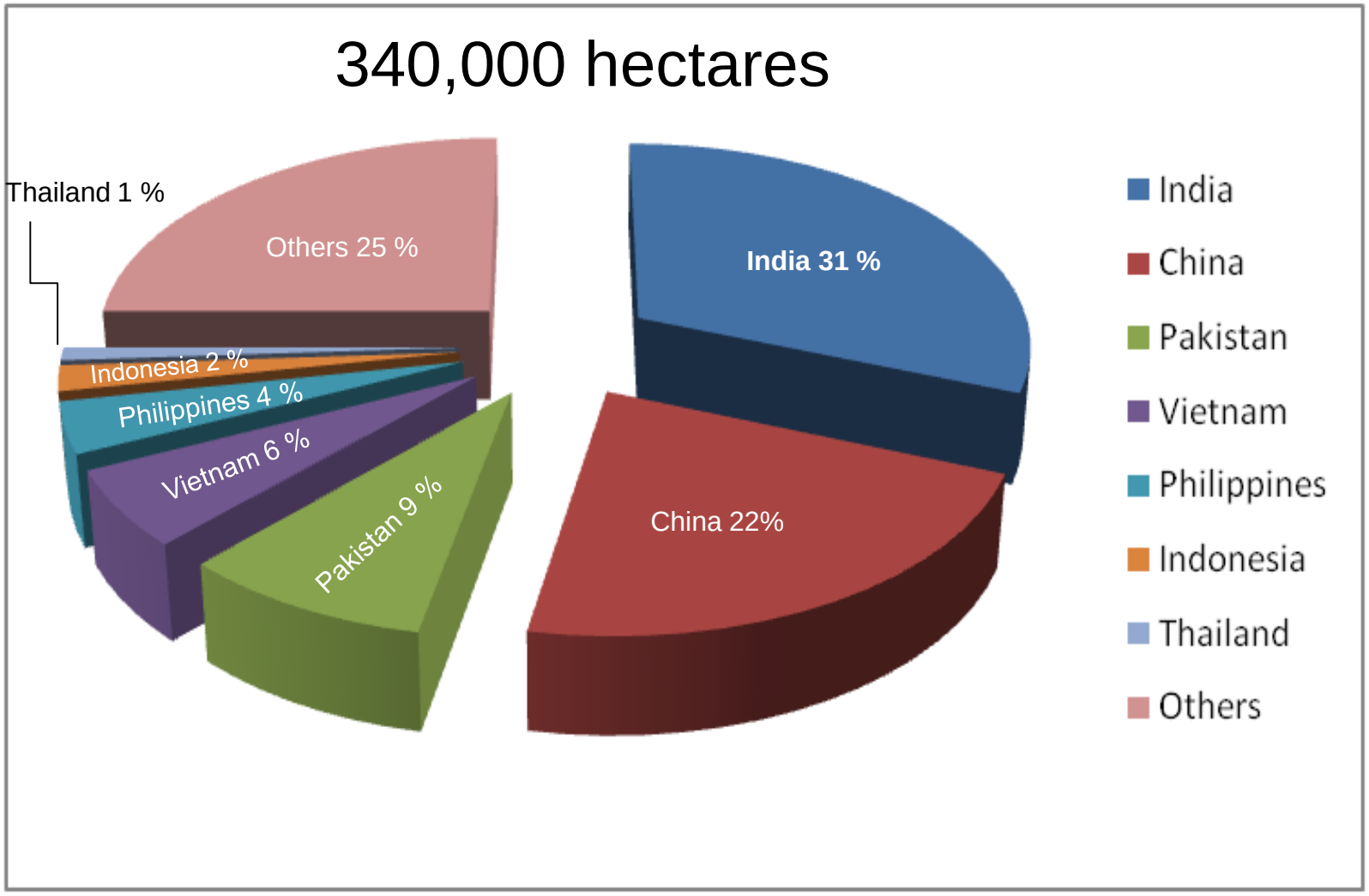
Source: FAOSTAT 2015

Crop focus

Major emphasis

- **Bitter gourd** (*Momordica charantia* L.)
- **Pumpkin** (*Cucurbita moschata* L.)

Bitter gourd cultivation in Asia



Source: EWS

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Bitter gourd breeding at AVRDC

Human Trial in India

Results after 8 weeks of intervention:

- **Reduction in body weight, BMI, and waist circumference** ($P<0.01$)
- **Reduced mean fasting blood glucose** from 110.66 mg/dl to 99.86 mg/dl ($P<0.01$)
- **Reduced HbA1C levels** from 6.37 % to 5.53 % ($p<0.01$)
- **Increased insulin level** from 9.5 to 10.57µU/dl in those treated with bitter gourd compared to a reduction of 0.33 µU/dl in the placebo treatment
- **Reduced triglyceride** ($P<0.05$), **total cholesterol** ($p<0.01$) and **LDL cholesterol** ($p<0.01$)
- No change in blood pressure

Anti-Hyperglycemic Substances in Bitter Gourd

Proteins

- p-Insulin-like
- v-Insulin-like
- Lectin
- Mormodin I, II….
- ……

Water fraction

Alkaloids

- Vicine
- …

Steroid and triterpenoid saponins

- Momordicine I, II, III
- Momordicosides Q, R, S, T
- Momordicosides A, D, E, F2, I, K, L
- Kuguacin A,…, S
- Charantin
- Cucurbitan B, K…
- ……

Saponin fraction

PKG3

PKG2

Lipids

- c9, t11 conjugated linoleic acid
- c9, t11, t13 conjugated linolenic acid
- . α- Eleostearic acid
- -……

Lipid fraction

PKG1

66

Bitter gourd statistics and importance

- Nearly 340, 000 ha are devoted to bitter gourd cultivation annually in Asia
- Fruit often used in folk medicine to treat type 2 diabetes
- 60% production area is under OPV in India, Bangladesh, Sri Lanka
- Hybrids yield 20-30 t/ha whereas OPVs yield 8-10 t/ha

Hypothesis

OP lines comparable to hybrids

in yield, earliness and fruit quality

Answer: Yes

Evidence?

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Genotype	Fruit					
	No./plant	Weight (g)	Yield (t/ha)	Bitterness	Color	Skin
Medium fruit length segment (South Asian type)						
AVBG1304	41	173	35	L	DG	Spiny
AVBG1310	23	266	33	L	G	Spiny
BARI 1 (Check)	22	178	23	H	DG	Spiny
Palee (Check)	30	194	36	L	G	Spiny
Long fruit length segment (Southeast Asian/Chinese type)						
AVBG1313	19	374	41	M	LG	Ribbed
AVBG1314	21	350	41	L	LG	Ribbed
Benteng (Check)	19	407	40	L	LG	Ribbed
Small fruit length segment (South Asian type)						
AVBG1323	47	138	32	M	G	Spiny
AVBG1324	59	115	38	M	G	Spiny
Noor (Check)	48	134	36	L	MG	Spiny
LSD (*P* = 0.05)	9	70	7			

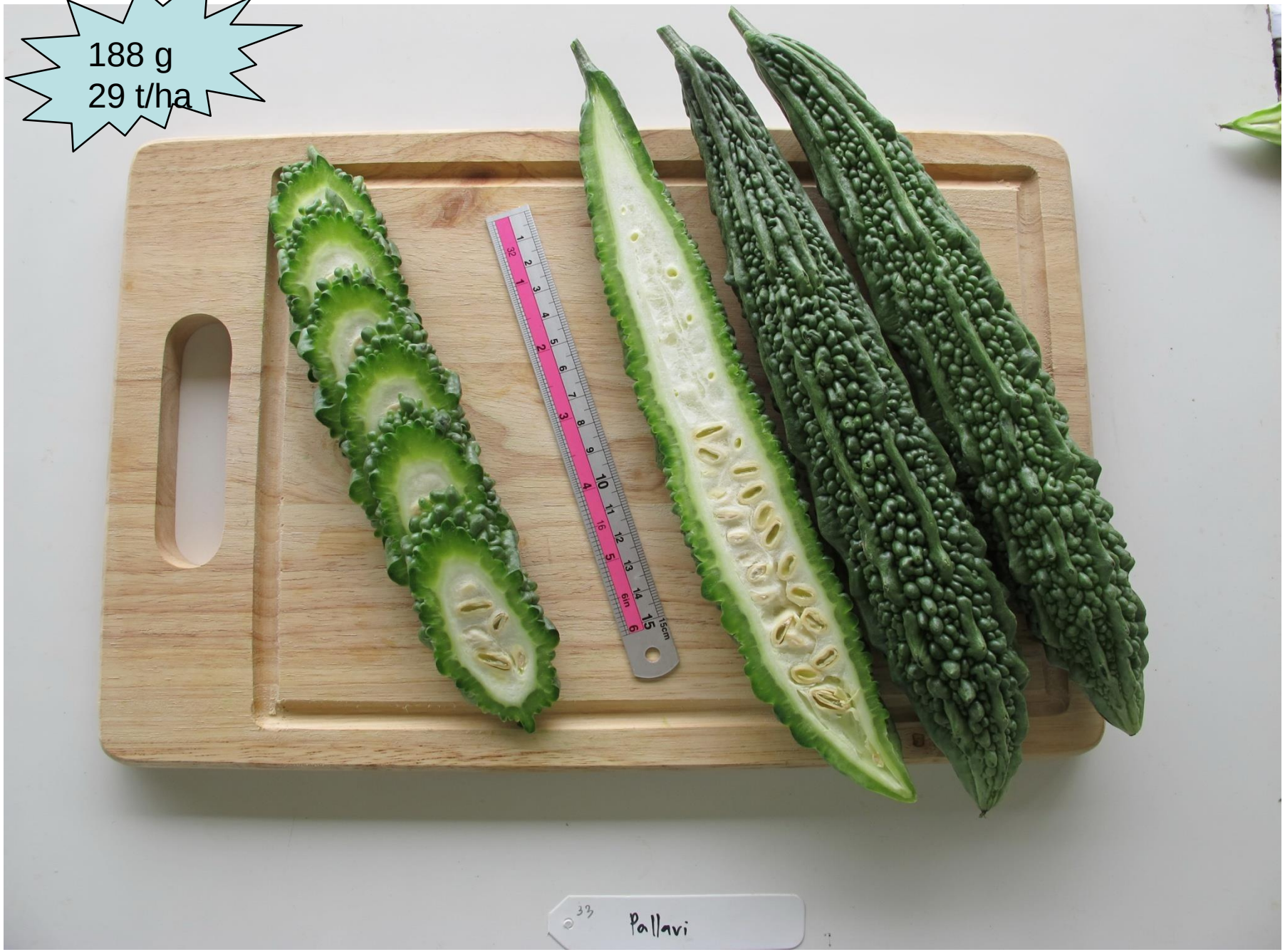
Bitter gourd: Trial in Kamphaeng Saen

Bitter gourd breeding block – Aerial view


174 g
32 t/ha
4) 12THBG2-06A6-20

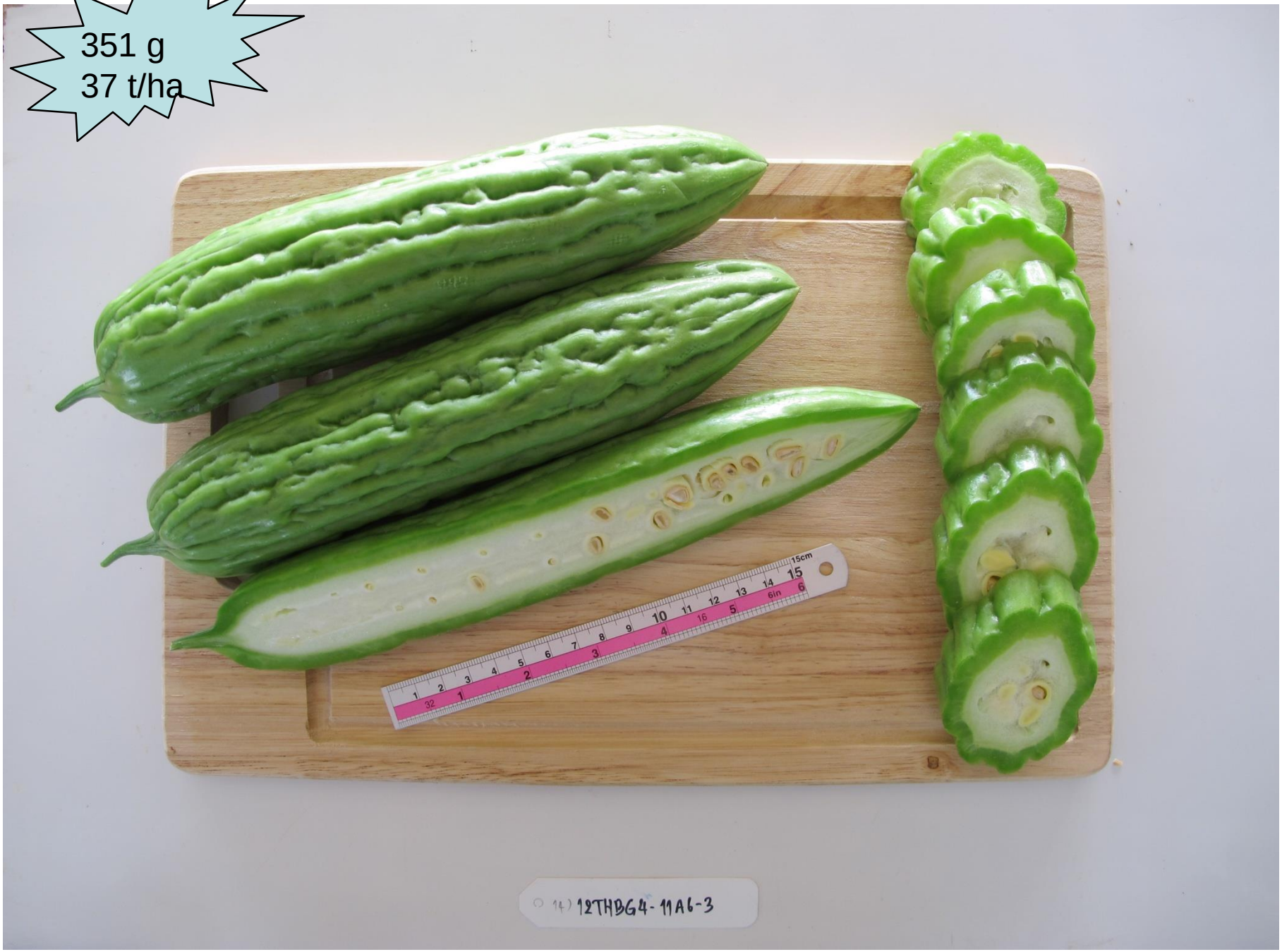

266 g
33 t/ha
12THBG3-09A6-20

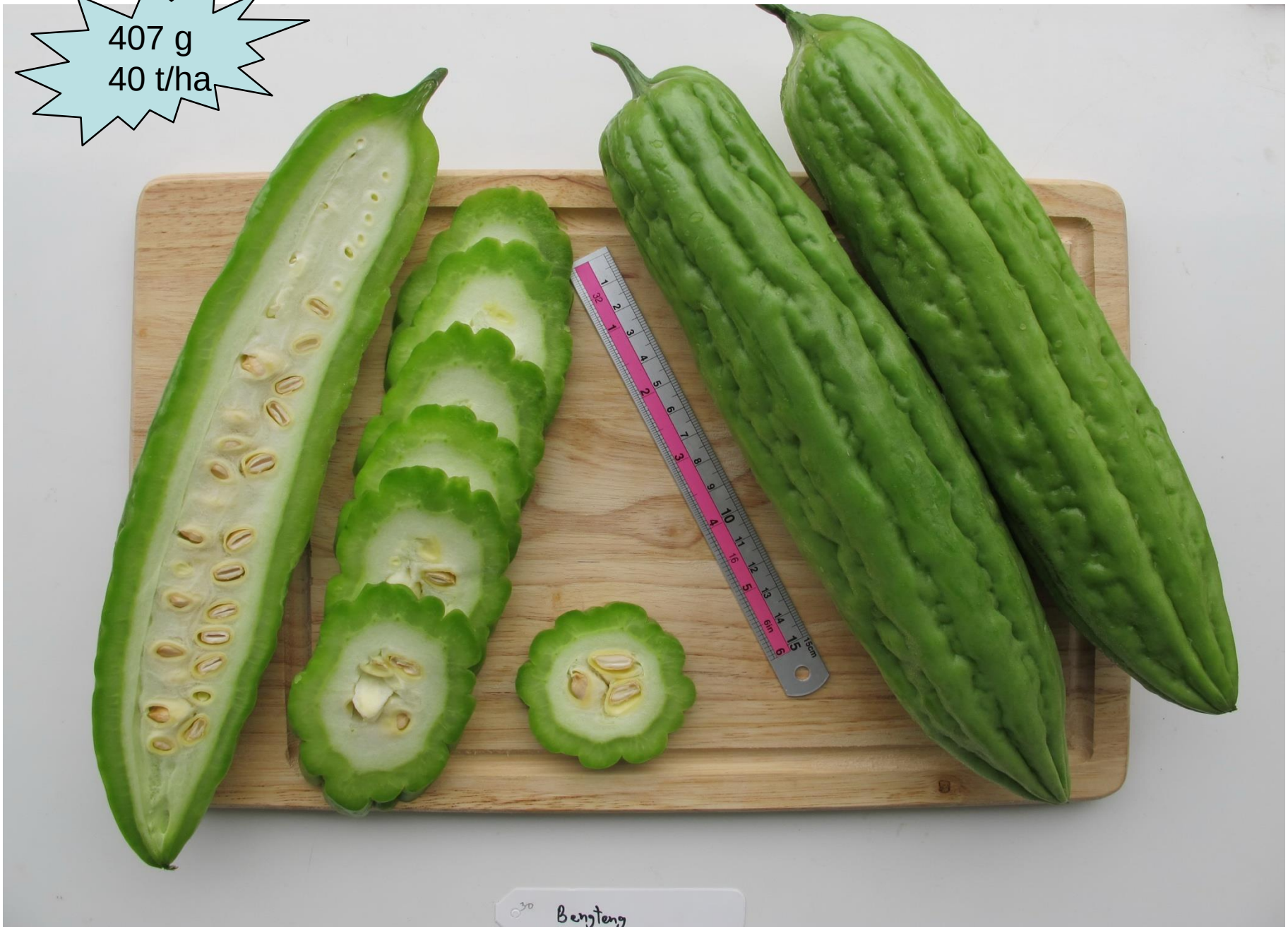

195 g
35 t/ha

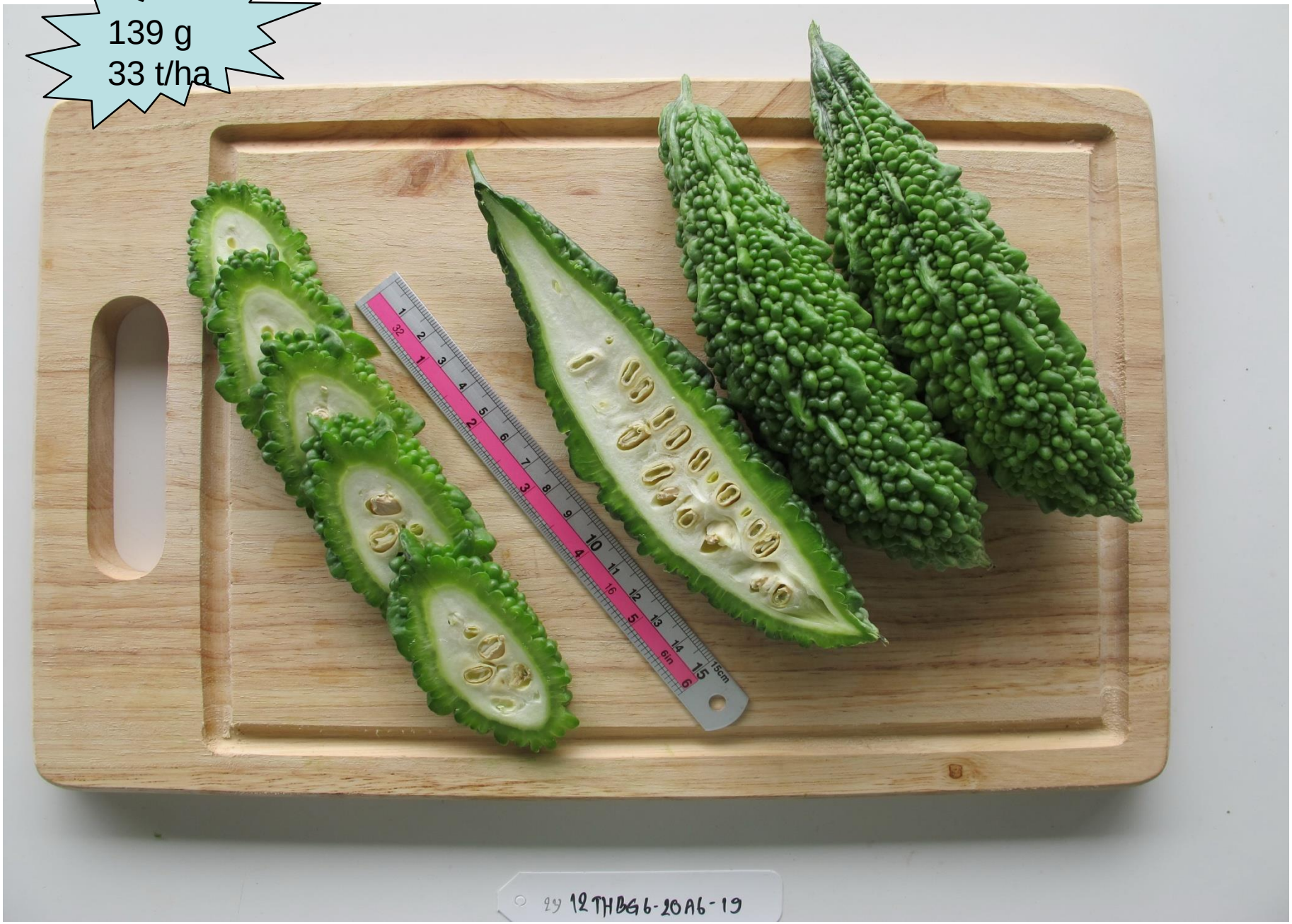

188 g
29 t/ha
Pallavi

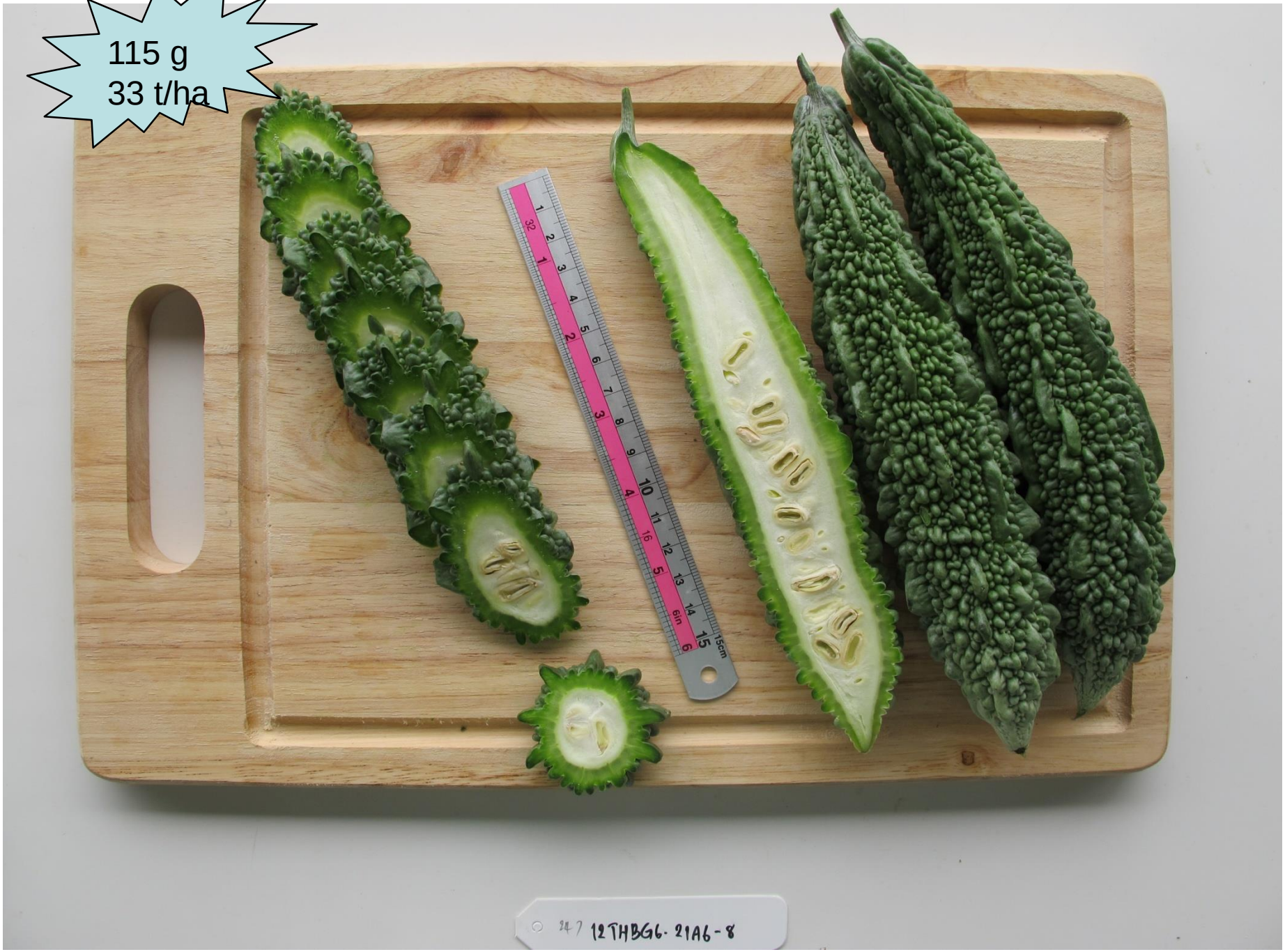

178 g
22 t/ha
BARI-1


351 g
37 t/ha
15cm
6in
14) 12THBG4-11A6-3


351 g
37 t/ha
12THBG4-11A6-3


407 g
40 t/ha
Bengteng


139 g
33 t/ha
12THBG6-20A6-19


115 g
33 t/ha
247 12THBG6-21A6-8


129 g
30 t/ha


110 g
29 t/ha
12THBG6-17A1-2


125 g
29 t/ha
12THBG1-03A6-13

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Bitter Gourd

Momordica charantia L.

Lines developed at AVRDC – The World Vegetable Center

AVBG1301

Pedigree: 12THBG1-03A6-13
Parentage: Vivek
Adaptation: Hot dry and hot-wet open field
Remarks: Vines vigorous, fruit medium size and green, spindle and spiny

To order seed, please email:

seedrequest@worldveg.org

A handling fee will be charged.

For seed distribution policies, please visit the AVRDC website: **www.avrdc.org**

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Box 42
Shanhua, Tainan 74199
TAIWAN

Descriptors	
Bitterness*	M
Fruit color**	MG
Fruit surface	Spiny
Fruit shape	Spindle

*S=strong, M=medium, L=low
**G=green, LG=light green, MG=medium green, DG=dark green

Mean quantitative data*	
Number of 1st female flower node	25
Days after flowering to harvesting	14
Number of fruit/plant	45
Fruit length (cm)	18.8
Fruit width (cm)	4.6
Fruit weight (g)	125
Yield (t/ha)	35
Maturity (DAS)**	59
Shelf life (day)***	2.3

*Average quantitative data were measured in August 2013 at AVRDC, East and Southeast Asia, Kamphaeng Saen, Thailand
**DAS: days after sowing to commercial harvest stage
*** Shelf life: days before the fruit becomes soft under the shade in the field conditions (day/night= 32-34/26-28 °C, RH=80-85%)

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Bitter Gourd

Momordica charantia L.

Lines developed at AVRDC – The World Vegetable Center

AVBG1313

Pedigree: 12THBG4-10A6-19
Parentage: Benteng 545
Adaptation: Hot dry and hot-wet open field
Remarks: Medium fruit size, cylindrical and light green, blunt blossom end and high yielding

Descriptors	
Bitterness*	L
Fruit color**	LG
Fruit surface	Ribbed
Fruit shape	Cylindrical

*S=strong, M=medium, L=low
**G=green, LG=light green, MG=medium green, DG=dark green

Mean quantitative data*	
Number of 1st female flower node	24
Days after flowering to harvesting	13
Number of fruit/plant	19
Fruit length (cm)	20.5
Fruit width (cm)	5.6
Fruit weight (g)	375
Yield (t/ha)	41.3
Maturity (DAS)**	59
Shelf life (day)***	2.1

*Average quantitative data were measured in August 2013 at AVRDC, East and Southeast Asia, Kamphaeng Saen, Thailand
**DAS: days after sowing to commercial harvest stage
*** Shelf life: days before the fruit becomes soft under the shade in the field conditions (day/night= 32-34/26-28 °C, RH=80-85%)

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Powdery mildew resistance evaluation in bitter gourd

Looking for potential sources for resistance to

powdery mildew (*Podosphaera xanthii*) in genebank

accessions of bitter gourd and subsequent fixing

through Inbreeding and testing against various races

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Powdery mildew infection in bitter gourd field in China

Photo: Fu Jiqin, Enza Zaden

AVRDC
The World Vegetable Center

"The Chinese growers will spray a lot to control the powdery mildew in bitter gourd fields, every 3 or 4 days. In the normal time, its every 7 days. The cost is about 75 RMB – 300 RMB (USD 12-50) for one hectare for one spray"

Fu Jiqin, Enza Zaden

"Farmers in Chhattisgarh have to spray fungicide against powdery mildew which is a costly affair as they have to spend 7 to 10% of the total outflow of the crop in fungicides only which interms of INR is costing them 10,000 to 30,000 (USD 160 – 480) per ha depending on the management practices and duration of the crop"

Vimal Chawda, VNR Seeds

Powdery mildew attack in glasshouse bitter gourds in S. Korea

Bitter gourd Pm resistance evaluation across Asia

Pm evaluation sites in Asia

Countries: 6
Locations: 12

Disease rating of seven bitter gourd lines to local isolates of *P. xanthii* at 12 locations in 6 countries

Country	Location	Bitter gourd breeding line						
		THMC 113	THMC 143	THMC 144	THMC 153	THMC 167	THMC 170	THMC 177
Thailand	Kamphaeng Saen	R	R	S	R	R	S	R
	Chiang Mai	R	R	S	R	R	R	R
Vietnam	Hanoi	R	R	S	R	R	R	S
	Ho Chi Minh City	R	R	S	R	R	R	R
Taiwan	Shanhua	S	R	S	R	R	S	R
Philippines	Bukidnon	R	R	S	R	R	R	S
India	Bangalore	R	R	S	R	R	R	S
	Pune	R	R	S	R	R	S	S
	Hyderabad	R	R	S	R	R	R	R
	Raipur	S	R	S	R	R	S	R
	Jagdalpur	S	R	S	R	R	R	S
China	Guangzhou	R	S	S	R	S	R	R

Scale 0-5

0=0%, 1=1-10%, 2=11-25%, 3=26-50%, 4=51-75%, 5=75-100%,

Resistant = 0-1

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Pm screening in Bangalore, Rasi seeds

Pm screening in Philippines, East-West Seeds

World Vegetable Center

Bitter Gourd Open Field Days

21 August - 6 September 2016

East and Southeast Asia/Oceania
Research and Training Station, Kasetsart University, Kamphaeng Saen Campus
Kamphaeng Saen, Nakhon Pathom, Kingdom of Thailand

Bitter gourd open field days

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Breeding line trial


Breeding line trial

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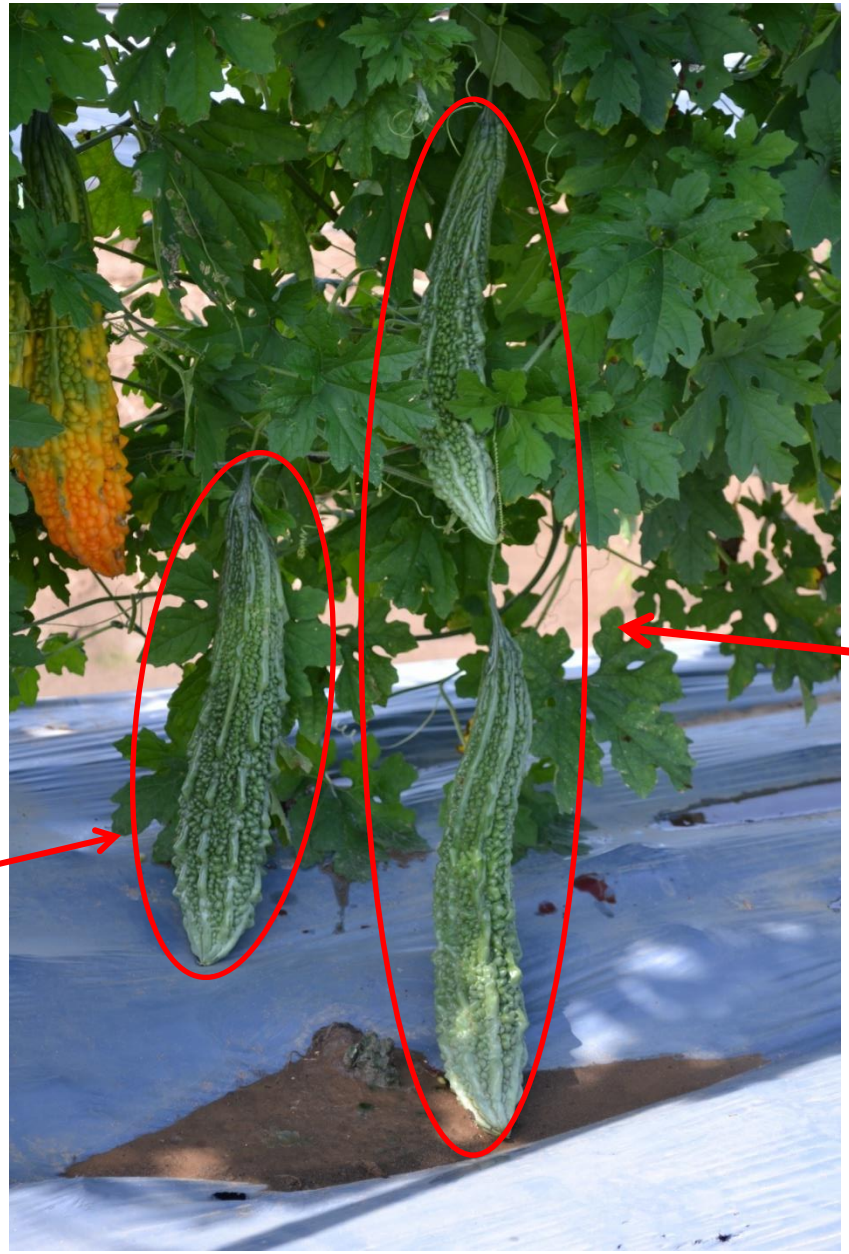
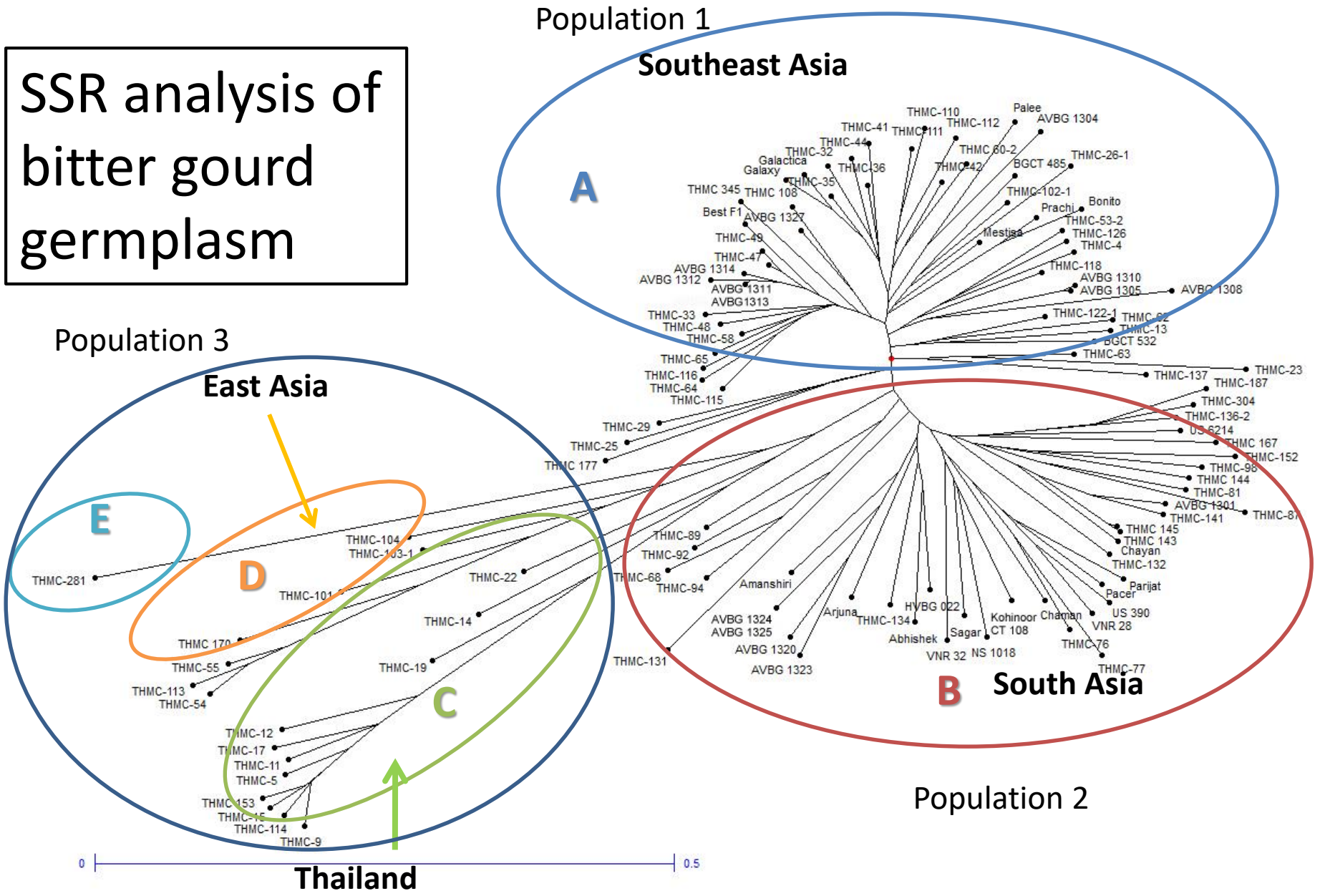
Market innovations

Unique traits to capture the bitter gourd market in South Asia


178 g
22 t/ha
BARI-1


AVBG
Hybrid
Palee

SSR analysis of bitter gourd germplasm



Dhillon et al. 2016

Pumpkin breeding

Statistics: Pumpkin, squash, gourd

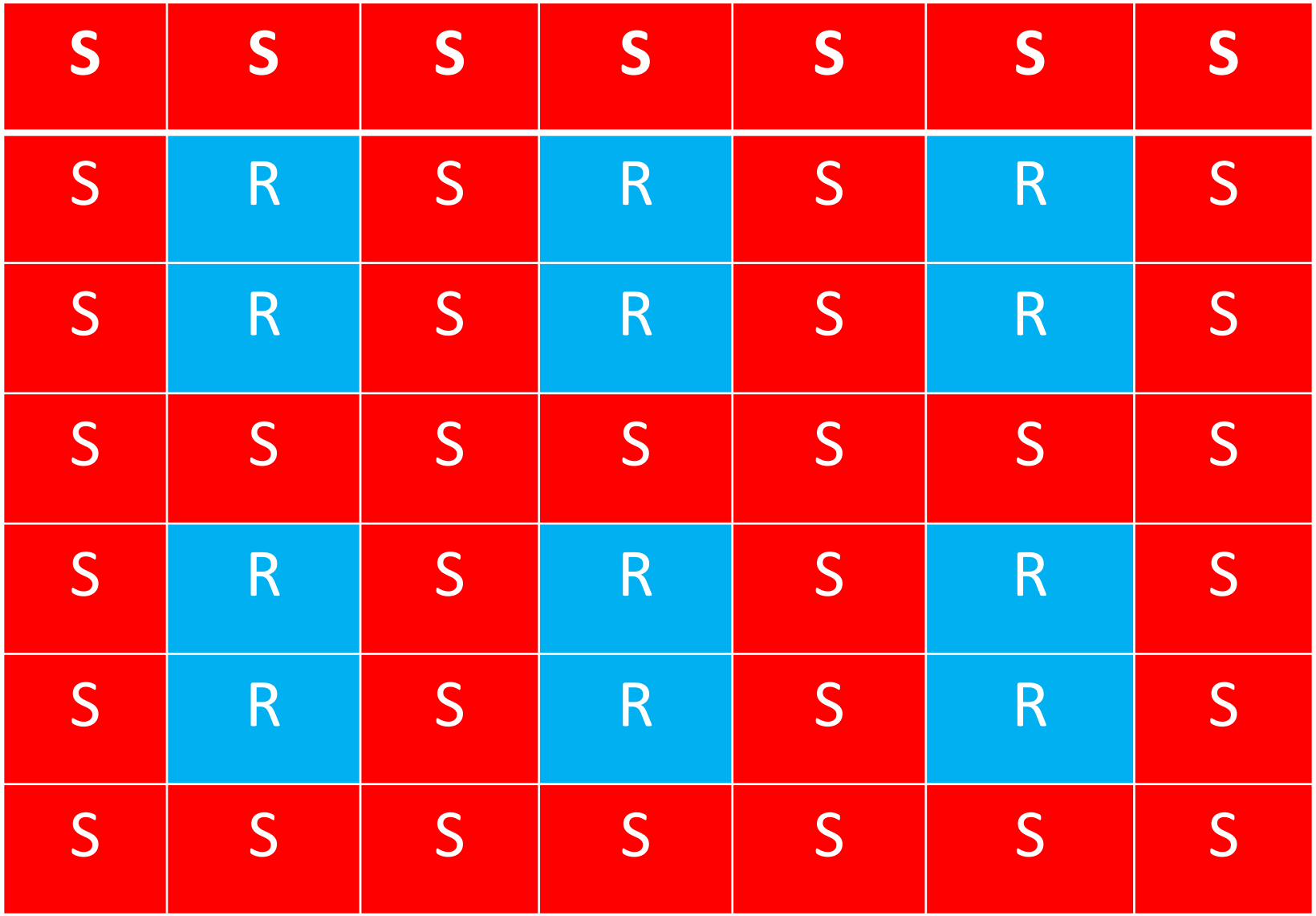
Continent	Area (ha)	Production (t/ha)	Average yield (t/ha)	Remarks
Africa	268,889	1,939255	7.2	Poor fruit quality, virus susceptibility, nutritionally poor
Asia	1,158942	15,951786	13.7	Poor fruit quality, virus susceptibility, nutritionally poor
N. America	42,144	903,285	21.4	
World	1,774554	24,256767	13.6	

Source: FAOSTAT 2014

Pumpkin virus resistance breeding in AVRDC

Farmer’s pumpkin field in Cagyan de Oro

Field design for multiple virus resistance evaluation



Field screening for multiple virus resistance

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Field screening for multiple virus resistance

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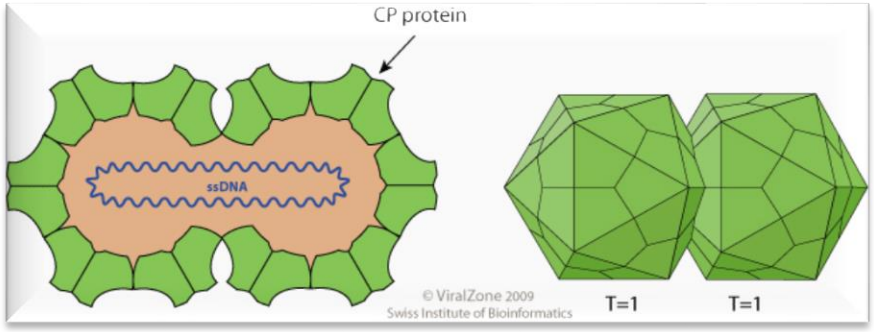
Artificial inoculation with potyviruses

Artificial inoculation

THMC 122-1-6-8-7-5			
Virus	Total plants	R	S
CMV	30	30	0
PRSV	30	29	1
ZYMV	30	25	5
THMC 120-1-3-2-9-7			
CMV	30	30	0
PRSV	30	29	1
ZYMV	30	26	4
Sus Check	30	0	30

Screening pumpkin lines for resistance to begomovirus



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Screening pumpkin lines against SqLCV

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Screening pumpkin lines against SqLCV

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Screening pumpkin lines against SqLCV

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Screening pumpkin lines against SqLCV

WorldVeg code	No. of tested plant	No. of infected plant				
		Week 3	Week 5	Week 7	Week 9	Week 11
THCM 114-2-2-2	30	1	2	3	4	6
THCM 119-2-3-4-10	30	2	2	2	2	3
AVPU 1501	30	13	14	14	15	15
AVPU 1502	30	26	28	28	28	28
AVPU 1504	30	0	2	2	2	2
THCM 102-3	30	2	2	3	3	3
Nigerian local	27	13	18	19	24	25
THCM 122-1-6-8-7	30	0	1	1	2	3
THCM 120-1-3-3	30	0	0	0	0	3
THCM 114-2-2-6	30	0	0	0	0	5
THCM 119-2-3-4	**30**	**0**	**0**	**0**	**0**	**0**
Suscep. Check-Walthum B	**10**	**10**	**10**	**10**	**10**	**10**

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Board of Directors & AVRDC staff in pumpkin breeding block

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Board of Directors & AVRDC staff in pumpkin breeding block

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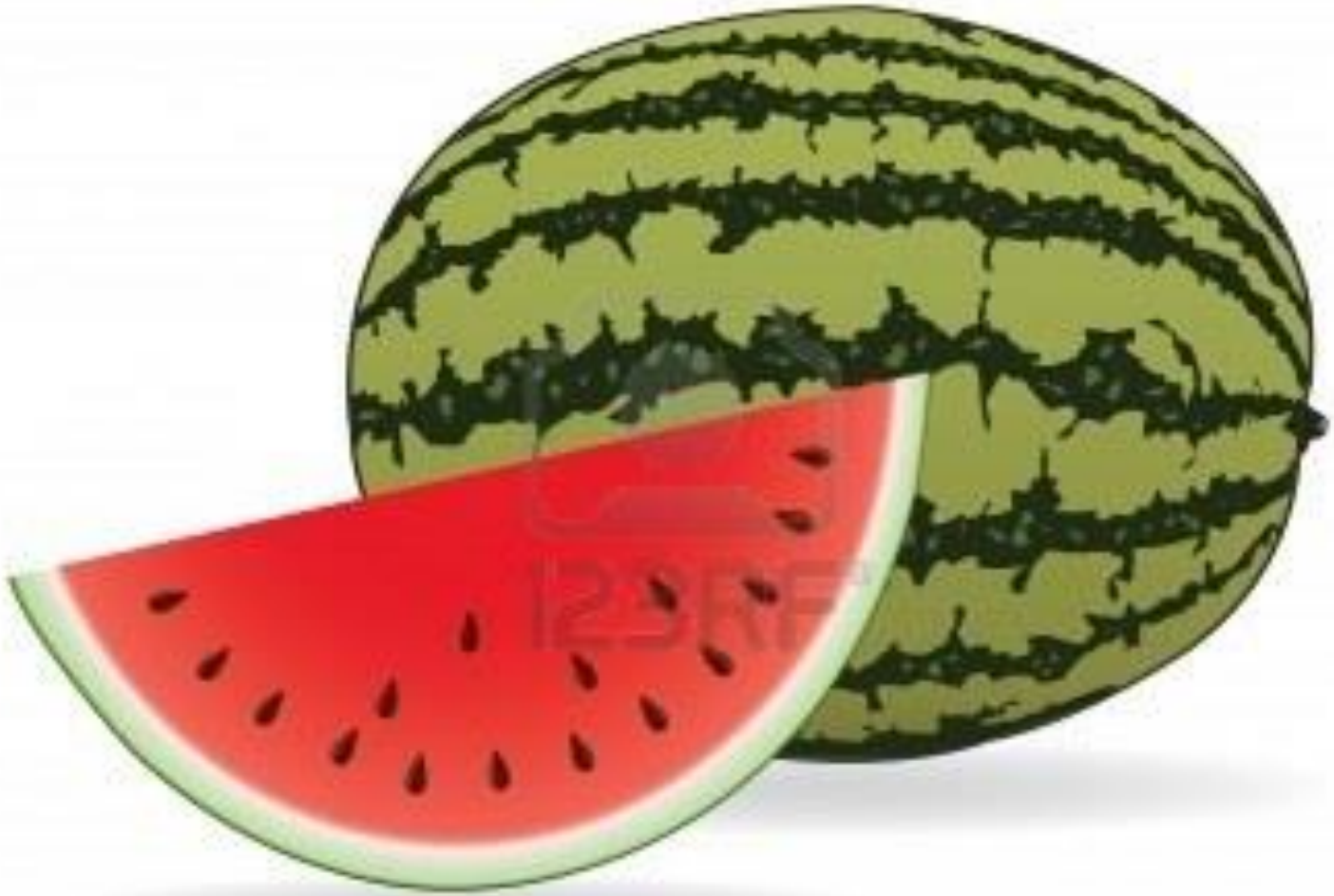
Lemon melon

700/800 mg of citric acid/100 g of fruit flesh

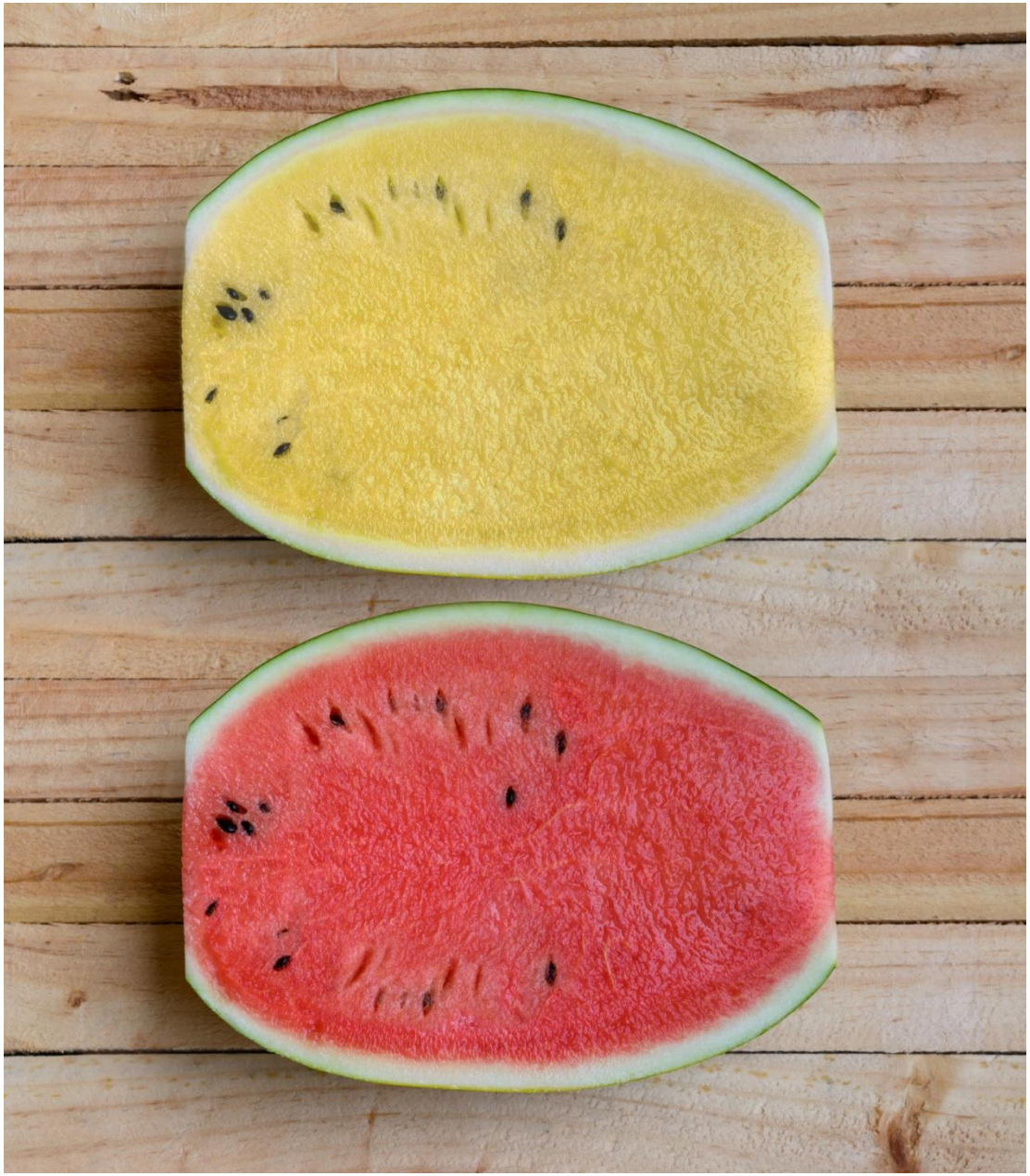
pH 4.5 compared with 6.5 for a standard melon

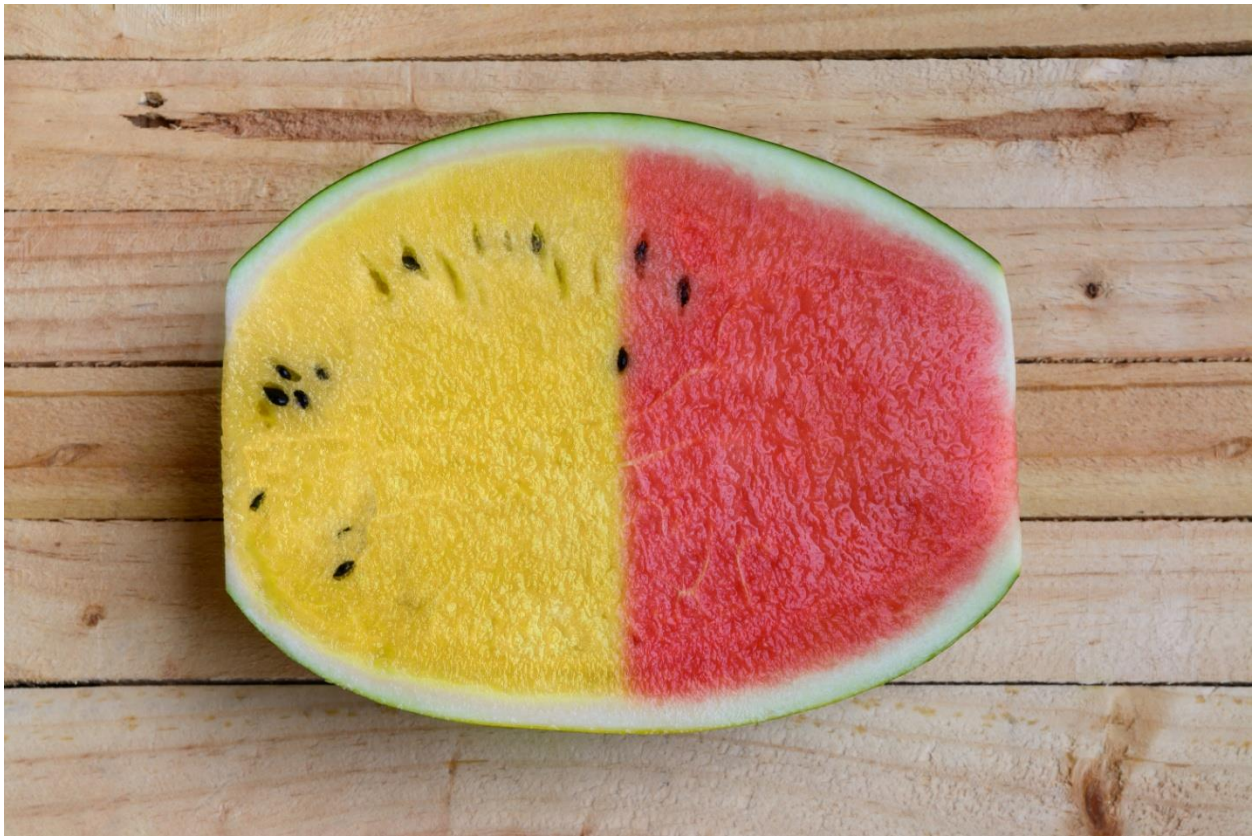
Cosmetic breeding !!!

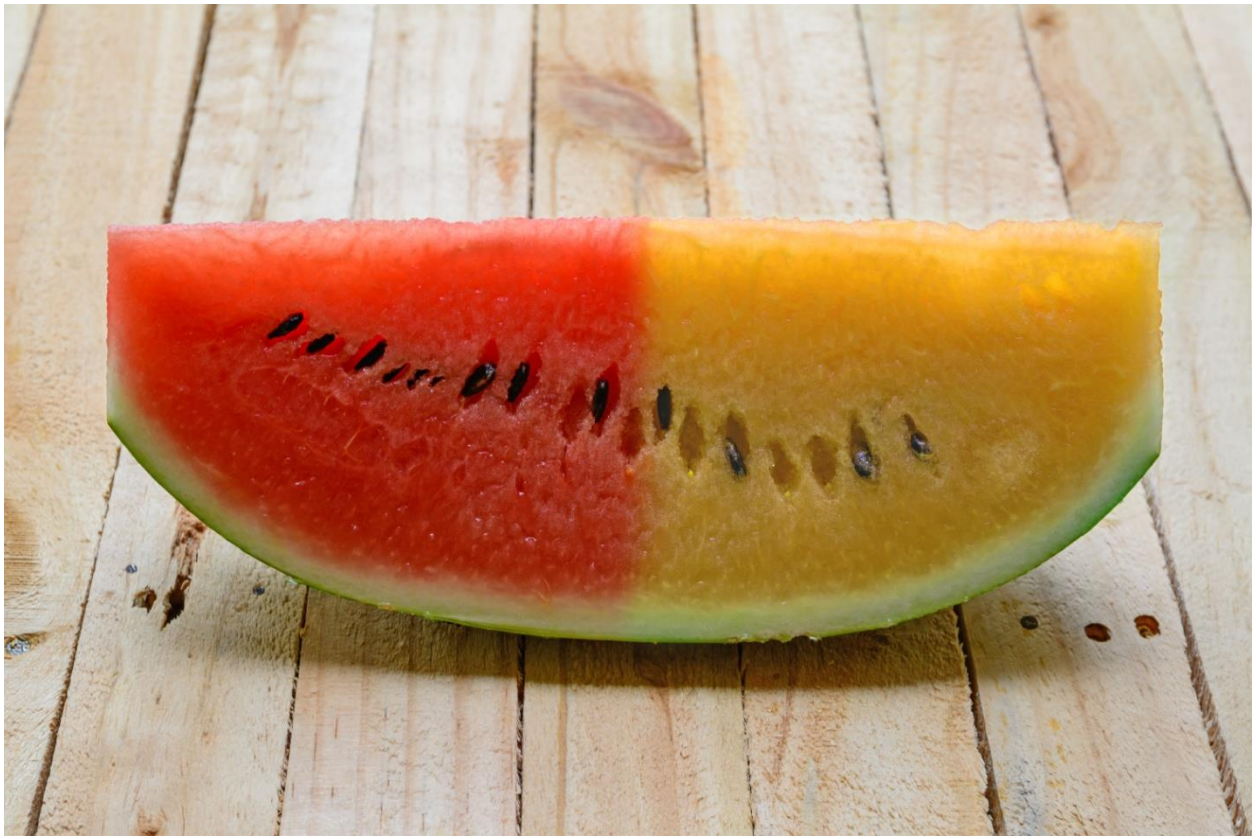
Future melons - Cosmetic breeding !



Future Watermelons – Cosmetic breeding !



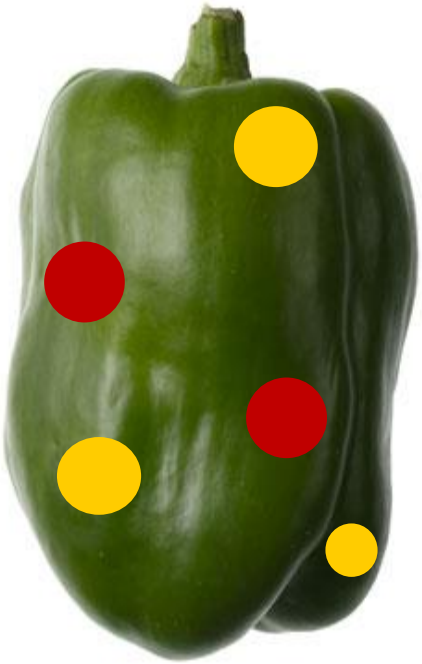


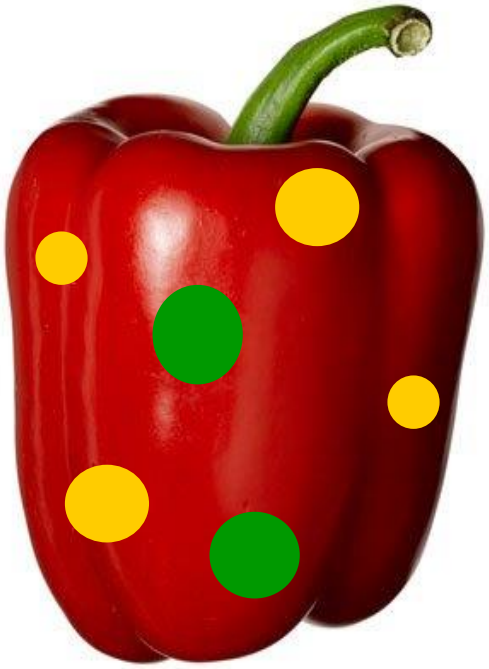


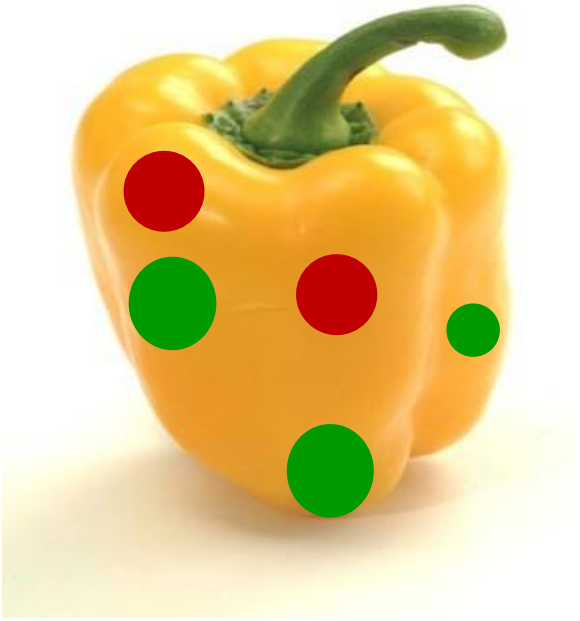
Future cabbages!

Future cabbages!

Future Sweet peppers – Cosmetic breeding!







COSMETIC

BREEDING

COSMETIC

BREEDING

Cucurbit Crew of World Vegetable Center

Have a good day!