Developing Vegetables with Enhanced Levels of Beneficial Phytochemicals

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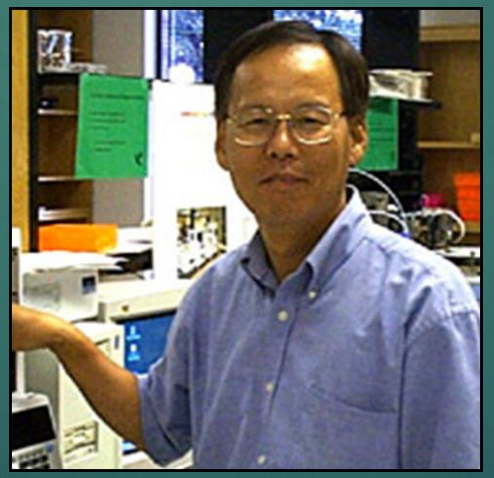
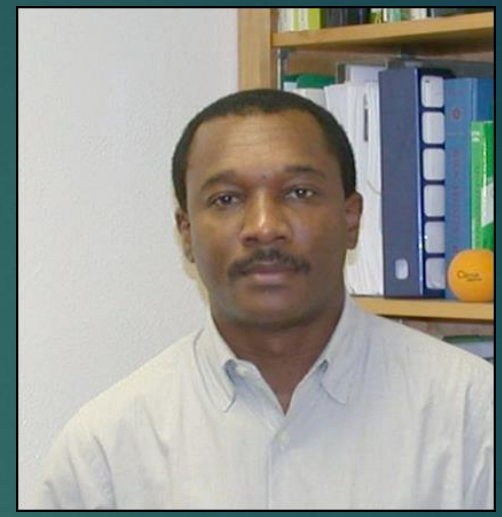
Vegetable & Fruit Improvement Center

A Team Effort

Dan Leskovar

Kil Sun Yoo





John Jifon

The Good News

- Strong correlations exist between elevated consumption of plant products and reduced disease incidence

- Key phytochemicals such as carotenoids, vitamin C, vitamin E are indispensable to human wellness

Vegetable Breeding at TAMU

- 70 years of vegetable genetic improvement
- More than 30 varieties for Texas
- Mild jalapeño, 1015 onion, β-sweet carrot, ‘Chico’ tomato, ‘Perlita’ cantaloupe
- Improved quality and health benefits


TEXAS-GROWN
FROM EDINBURG
THESE ARE THE TRIED and TRUE, OFFICIAL, UNMISTAKABLE, UNADULTERATED, TEXAS A&M-CERTIFIED REAL DEAL!
WANT TO KNOW MORE? ASK US!!
1015 ONIONS
99¢ LB
4161 ON SCALE
Little Bear
BRAND
Little Be
Sweet

Leonard Pike

Founded Vegetable Improvement Center in 1990.

Brought together industry and academia to address quality issues in vegetable crops

Dedicated 40 years to improvement of short day onions, carrots, pickles

Trained many successful plant breeders

Beta-sweet Carrot

Foods for Health Research

- Elevated levels of flavonoids, carotenoids, ascorbic acid, minerals, pectins
- Traditional plant breeding- exploitation of naturally occurring genes in germplasm
- Molecular Marker development for genomics and marker-assisted selection

Current Priorities

- Cultivar Development
  - quality
  - nutritional value
  - stress resistance
- Basic Research
  - genetic inheritance
  - genome mapping
  - tissue culture, GM

Crops of Interest

- Peppers- vitamin C, carotenoids, flavonoids, capsaicin and fiber
- Melons- beta-carotene, vitamin C, folate, potassium
- Tomatoes- lycopene, ascorbic acid, phenolics
- Onions- quercetin, pyruvates, pectins

Dietary Fiber in ½ cup Serving

Green Peppers- raw	2.4
Hot Red Peppers- raw	19.2
Hot Red Peppers- dried	57.6
Pinto Beans- cooked	9.4
Prunes- dried	3.8
Tomatoes- raw	1.4
Spinach- cooked	7.0
Squash- cooked	3.0

Pepper Projects

- Screened >1000 diverse pepper lines from around the world
- Identified several lines with exceptional levels of vitamin C, flavonoids, carotenoids, capsaicin
- Created more than 90 new families to combine important genes

High Antioxidant Parents

TAES B22 CA 377

F1 Hybrid

Molecular Markers in Pepper

- Family of high flavonoid CA377 x high vitamin C B22
- F2 progeny, F1 progeny, Parents grown in two environments for mature fruit
- Screening with RAPD markers in bulks and genotyping each F2 progeny
- Constructing genetic linkage map

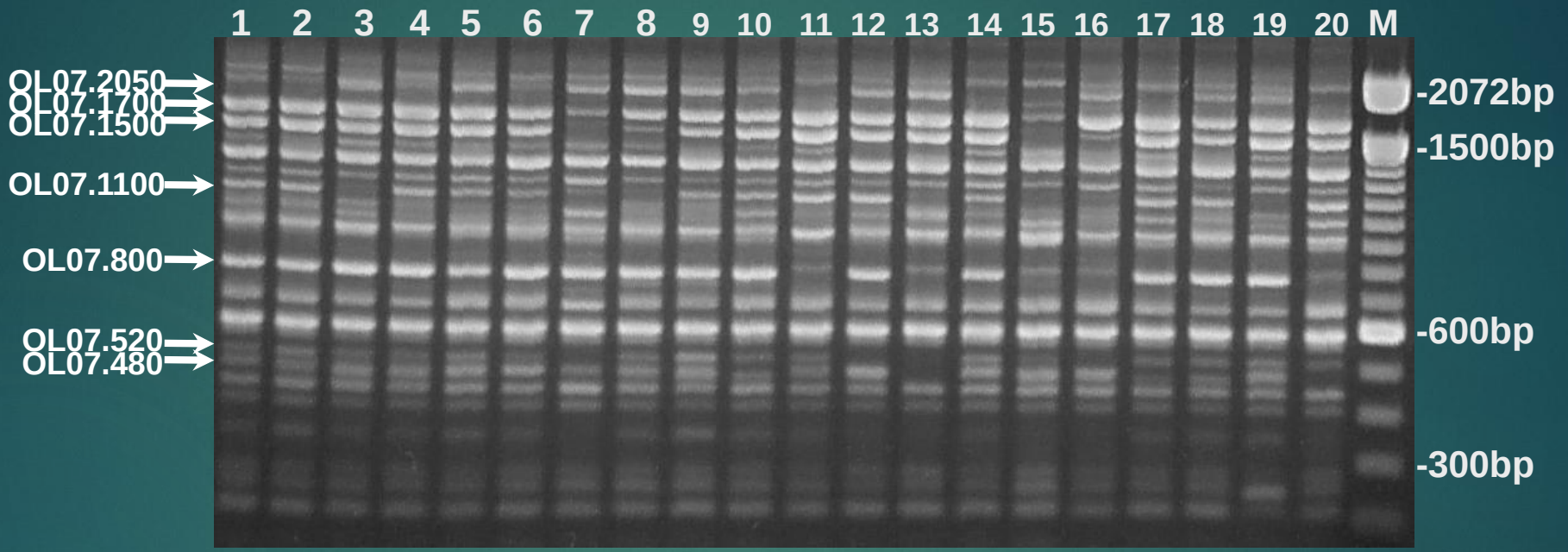


Fig. 1. Segregation of seven RAPD markers in an F2 population derived from the pepper cross of CA 377 x B 22. 1 to 20=F2 plants of the CA 377 x B 22 cross and M=a 100-bp DNA marker ladder.

Results

- Vitamin C levels continuously distributed from 31-4162 µg/g, H^2= 0.88
- Total flavonoid levels continuously distributed from 19-907 µg/g, H^2= 0.85
- Fruit size skewed towards smaller sizes
- One RAPD marker associated with Luteolin (5%)

Negative Linkage

- Fruit size negatively correlated with high flavonoids
- No negative association with ascorbic acid

Mean ascorbic acid concentrations (µg g^{-1} FW) in different pepper fruits (*C. annuum*) grown in two different Texas locations in spring 2010.

Entry	Pepper Type	Ascorbic Acid Values	
		U.V.	W.E.
J-1	Jalapeño	465.07 e[π] B[Δ]	748.17 d A
J-2	Jalapeño	175.68 hij B	528.80 h A
J-3	Jalapeño	170.01 ij B	583.41 fgh A
J-4	Jalapeño	173.06 ij B	396.35 j A
J-5	Jalapeño	402.04 f B	542.05 gh A
J-6	Jalapeño	471.08 e B	676.40 de A
J-7	Jalapeño	230.13 gh B	430.80 ij A
J-8	Jalapeño	253.43 g B	575.15 fgh A
J-9	Jalapeño	500.81e B	681.46 de A
J-10	Jalapeño	405.53 f B	545.33 gh A
Dragon	Jalapeño	476.46 e A	441.46 ij A
Tormenta	Jalapeño	806.85 c A	927.83 c A
S-1	Serrano	70.57 k B	722.55 d A
S-2	Serrano	121.01 jk B	634.40 ef A
S-3	Serrano	178.53 hi B	502.64 hi A
S-4	Serrano	74.99 k B	666.35 de A
Halcon	Serrano	265.85 g B	618.36 efg A
Magnum45	Serrano	390.13 f B	683.43 de A
C-1	cayenne	1272.12 a B	2167.59 a A
C-2	cayenne	1214.74 b B	1557.65 b A
Mesilla	cayenne	610.93 d B	865.60 c A

U.V.: Uvalde location; W.E.: Weslaco location.

Π Mean separations within each location by LSD at $P \leq 0.05$. Means followed by the same lower case letters are not significantly different.

Δ Mean separations across locations by LSD $P \leq 0.05$. Means followed by the same upper case letters are not significantly different.

Mean flavonoid (μg·g^{-1})concentrations across 2 locations

Entry	Pepper Type	Quercetin Values		Luteolin Values	
		U.V.	W.E.	U.V.	W.E.
J-1	Jalapeño	7.20 cde$^{\Pi}$ A$^{\Delta}$	3.13 def B	1.25 fg A	1.20 efg A
J-2	Jalapeño	5.42 cde A	1.35 ef B	3.53 cdef A	1.69 defg B
J-3	Jalapeño	5.95 cde A	2.15 def B	2.21 efg A	0.99 efg B
J-4	Jalapeño	2.32 de A	0.96 ef B	1.05 fg A	0.99 efg A
J-5	Jalapeño	9.45 cd A	3.49 def B	5.10 c A	3.16 cd B
J-6	Jalapeño	9.80 cd A	1.48 ef B	2.14 fg A	1.58 defg A
J-7	Jalapeño	4.18 de A	1.43 ef A	1.44 fg A	2.19 cdef A
J-8	Jalapeño	6.29 cde A	2.59 def B	4.75 cd A	3.93 c A
J-9	Jalapeño	12.46 bc A	1.76 ef B	4.66 cde A	2.52 cde A
J-10	Jalapeño	0.70 e A	0.41 f B	0.88 g A	0.97 efg A
Dragon	Jalapeño	8.48 cde A	0.91 ef B	2.47 defg A	0.91 efg B
Tormenta	Jalapeño	3.73 de A	1.34 ef B	2.48 defg A	2.57 cde A
S-1	Serrano	18.64 b A	8.13 d A	2.69 cdefg A	2.33 cdef A
S-2	Serrano	9.55 cd A	2.97 def B	2.65 cdefg A	1.97 defg A
S-3	Serrano	8.14 cde A	2.27 def B	1.68 fg A	0.60 fg B
S-4	Serrano	12.91 bc A	6.64 de B	2.61 cdefg A	1.51 defg B
Halcon	Serrano	0.71 e A	0.13 f B	1.31 fg A	0.24 g B
Magnum45	Serrano	3.18 de A	0.24 f B	2.08 fg A	0.18 g B
C-1	cayenne	88.14 a A	57.92 a A	8.69 b A	10.98 a A
C-2	cayenne	80.83 a A	29.11 b B	13.16 a A	7.77 b B
Mesilla	cayenne	18.39 b A	18.95 c A	9.51 b A	8.79 b A

U.V.: Uvalde location; W.E.: Weslaco location.

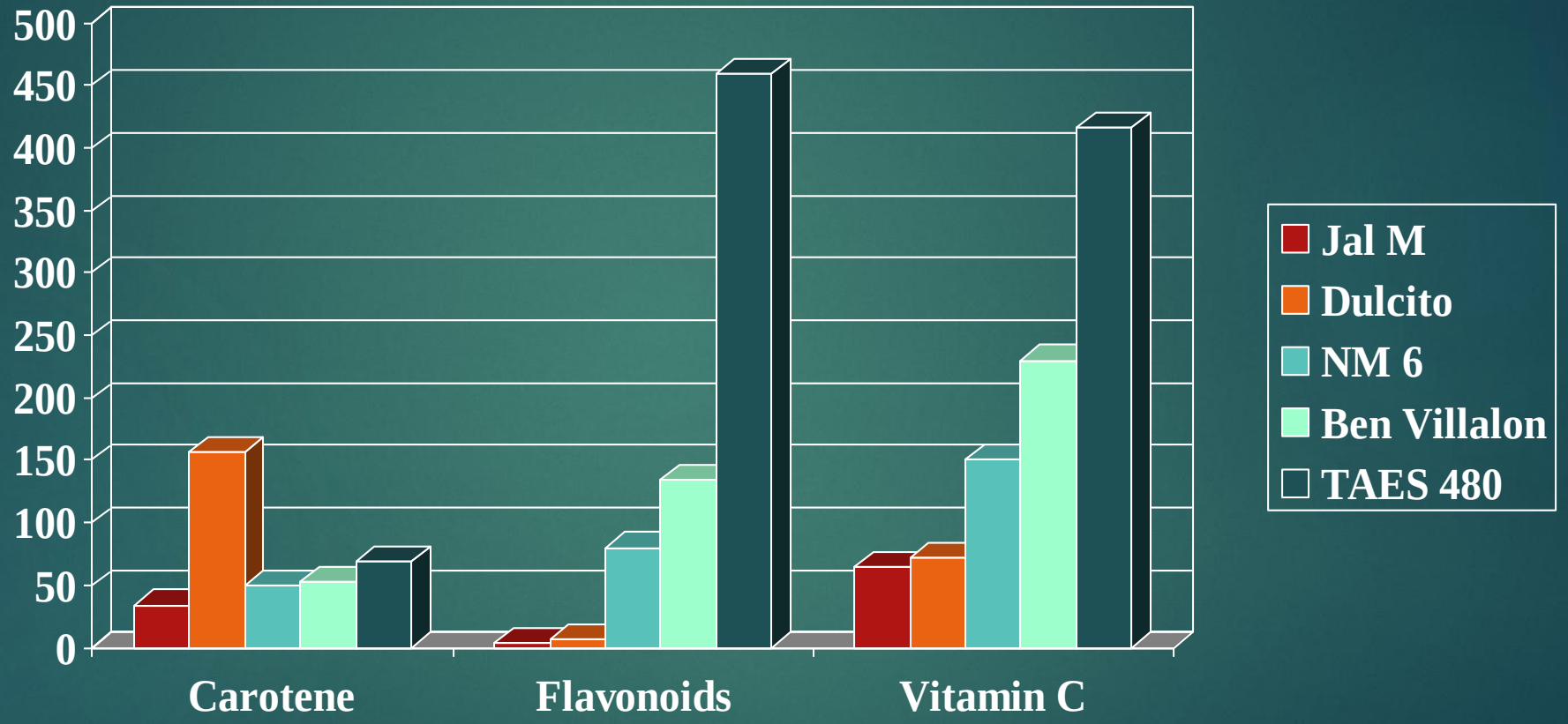
Π Mean separations within each location by LSD at $P \leq 0.05$. Means followed by the same lower case letters are not significantly different.

Δ Mean separations across locations by LSD $P \leq 0.05$. Means followed by the same upper case letters are not significantly different.

Mid-parent heterosis (%) estimates for each fruit characteristic and phytochemical group.

Gen.	FW	FL	FD	WT	AA	Cap.	Total DHC	Cap.	Quercetin	Luteolin	Total Flav.
Pap2	2.51	13.96	-4.43	-8.00	52.38	0.00	-100.00	-100.00	-54.54	-46.69	-52.37
Pap4	64.42	17.88	32.49	13.68	58.55	0.00	-100.00	-100.00	-38.57	-42.36	-39.09
Pap5	-31.42	6.41	-16.04	-21.57	37.37	0.00	-100.00	-100.00	5.75	19.56	9.29
S11	11.54	-2.47	3.45	-9.86	8.63	33.98	26.39	29.73	4.82	83.61	31.22
S12	-4.38	-1.71	2.15	-7.25	55.87	-52.98	-43.85	-48.25	241.35	233.49	238.39
S14	8.81	-3.02	6.67	18.31	45.91	84.42	78.29	81.25	135.19	124.68	131.37
S27	-6.23	21.74	-16.53	-18.64	12.07	1289.23	417.14	838.07	-10.11	4.26	-8.10
S28	16.24	18.88	0.27	5.08	58.38	357.53	119.44	250.70	-16.68	44.76	-8.83
S30	12.63	1.24	3.03	4.92	-23.70	329.66	92.87	220.96	-30.16	-26.15	-29.46
S32	-17.32	8.20	-15.41	0.00	30.46	1121.93	560.61	902.32	-16.89	-6.04	-15.69
S36	-0.58	24.43	-20.69	-35.59	25.45	170.65	113.72	141.22	42.56	5.13	34.36
S37	-4.96	6.15	-7.51	-11.86	-12.17	204.65	101.98	157.54	22.73	2.05	18.34
S38	31.99	11.38	4.93	13.79	-6.16	45.58	40.96	44.07	8.07	-16.31	0.94
S40	9.65	13.82	-6.43	-1.64	17.06	188.19	91.00	142.74	4.98	-22.89	-2.35
S41	-1.67	25.05	-12.87	1.59	11.32	125.13	62.65	99.52	85.14	25.15	70.18
S43	76.46	36.20	9.14	20.00	9.27	-34.13	-49.38	-42.22	-74.93	-55.92	-71.49
S46	8.37	18.88	-6.62	-4.35	18.76	90.20	15.84	61.23	33.91	-16.60	20.04
S47	4.92	1.43	3.32	-8.33	-10.01	93.78	28.69	68.32	-22.38	0.65	-19.84
S48	-15.00	17.42	-21.49	-28.30	18.42	38.58	-11.02	17.29	79.68	8.43	59.09
S56	22.07	6.69	11.46	9.09	40.92	102.30	63.10	81.21	34.30	63.86	46.05
S60	-18.92	-0.32	-11.29	-12.12	-51.31	37.91	73.62	53.78	-43.50	-39.46	-41.70
S68	-15.48	9.88	-13.64	-1.64	28.54	63.45	89.22	73.59	10.93	21.67	15.87
S70	-12.33	9.43	-10.78	-25.42	28.26	48.38	101.62	69.62	-25.25	-37.17	-30.32
S74	5.33	27.60	-10.85	-21.57	21.73	19.85	21.27	20.42	7.96	-22.38	-5.58
S90	-22.24	-14.41	-3.41	3.57	104.93	-0.12	51.39	23.19	40.41	59.11	45.34
S91	-15.11	9.82	-10.49	-24.53	24.90	221.76	74.51	147.57	62.20	63.72	62.47
S95	-2.12	15.62	-8.02	-5.66	53.38	31.79	-31.64	-0.17	52.58	44.72	51.19
S107	3.29	12.38	-4.22	-1.89	24.97	490.85	337.25	429.80	32.48	78.66	40.38
S108	-1.20	4.41	-6.52	-16.98	-12.44	697.50	437.30	609.46	-0.33	122.40	10.91

Pepper Beneficial Phytochemicals



High Flavonoid Wild Pepper

Cultivar: Fidel
Vitamin C: 1897 ppm
Flavonoid: 277 ppm

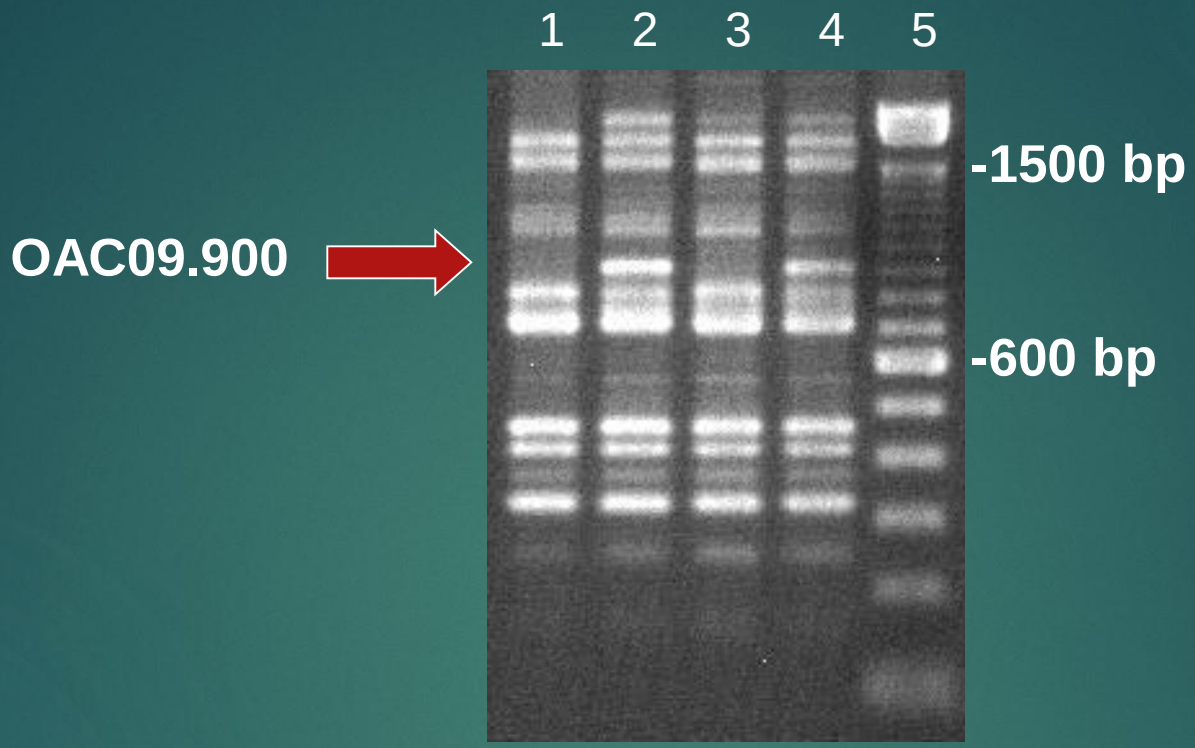
New *P. capsici* Resistant, high flavonoid, high capsaicinoid Orange Serrano

Melon Projects

- Screened >500 diverse melon lines
- Identified many important fruit quality and disease resistance traits
- Developed > 1200 new families; began selection and development of advanced inbreds with elevated phytochemicals

Molecular Marker Development

- Melon family- 120 F_2 progeny
- Segregation for ascorbic acid, sugar, beta-carotene concentrations
- High (Uvalde), low (Sunrise) β-carotene parents
- Bulked segregant analysis and RAPD markers



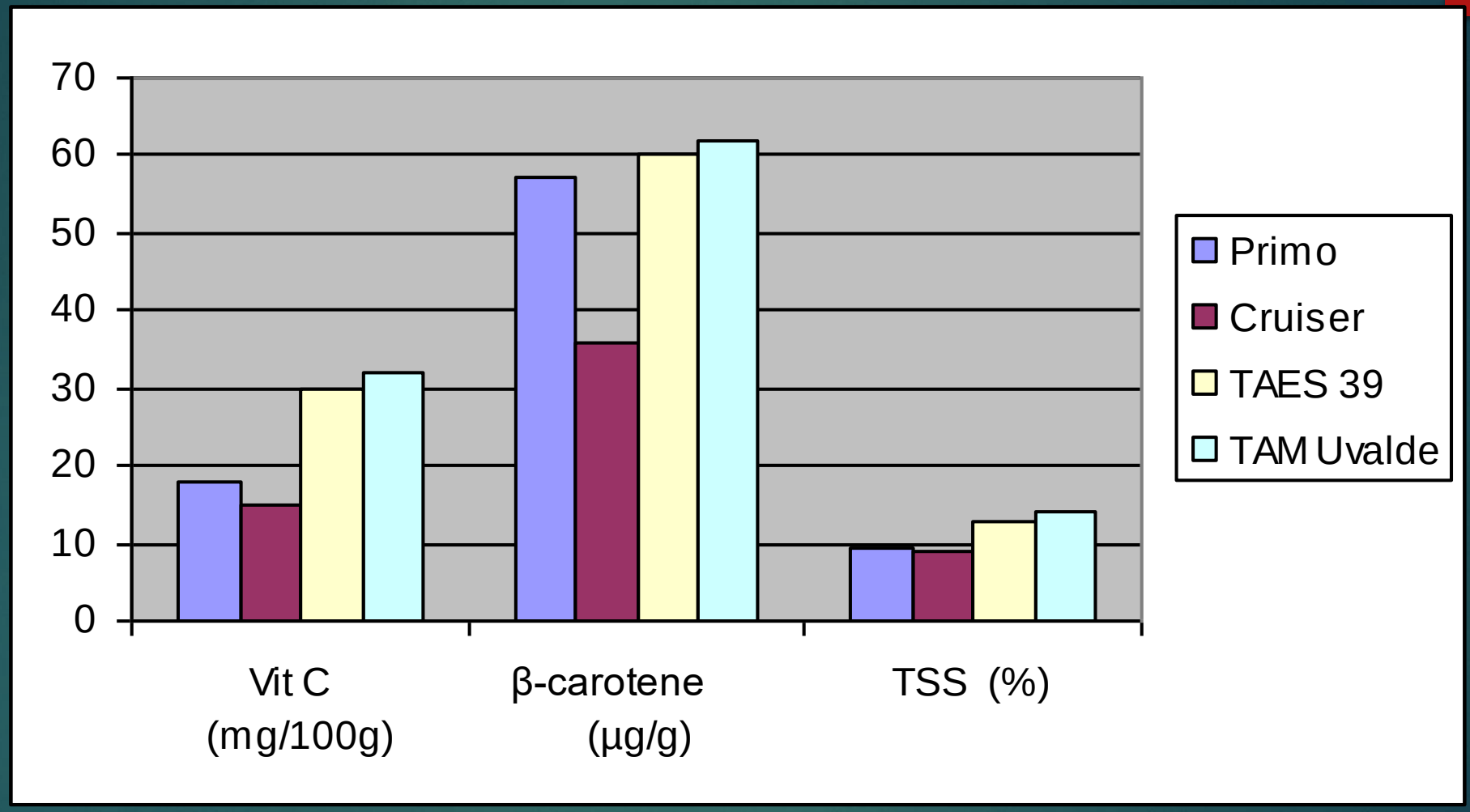
RAPD marker OAC09.900 expressing polymorphism between two DNA bulks from high and low beta-carotene F2 plants. 1 = Sunrise (low parent), 2 = TAM Uvalde (high parent), 3 = DNA bulk from low beta-carotene F2 plants, 4 = DNA bulk from high beta-carotene F2 plants, and 5 = a 100-bp DNA marker ladder.

Results

- Identified several reproducible RAPD markers polymorphic between parents and bulks
- Continued screening with more primers and AFLP's to identify tightly linked markers
- No success with more than 500 primers
- Released 2 new melon cultivars

High β-carotene ‘Chujuc’

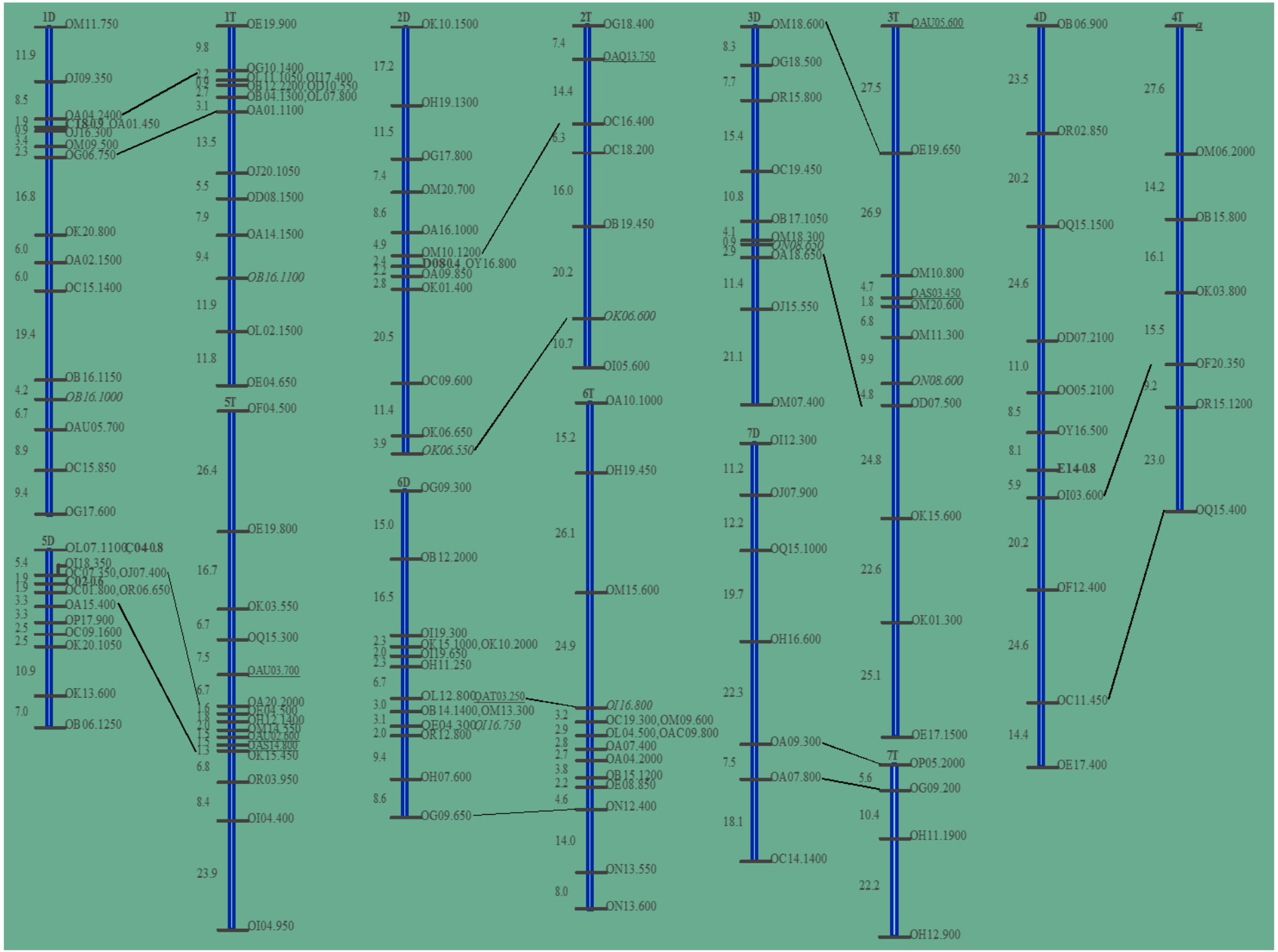
Melon Beneficial Phytochemicals



Genetic Linkage Map

- ‘Deltex’ x ‘TGR 1551’ family
- 120 F2 progeny screened with RAPD markers- PCR
- Assembled linkage groups with Mapmaker software; anchored with known genes

F_2 Progeny of 'TGR x Deltex'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OA02.1500
6.0
OC15.1400
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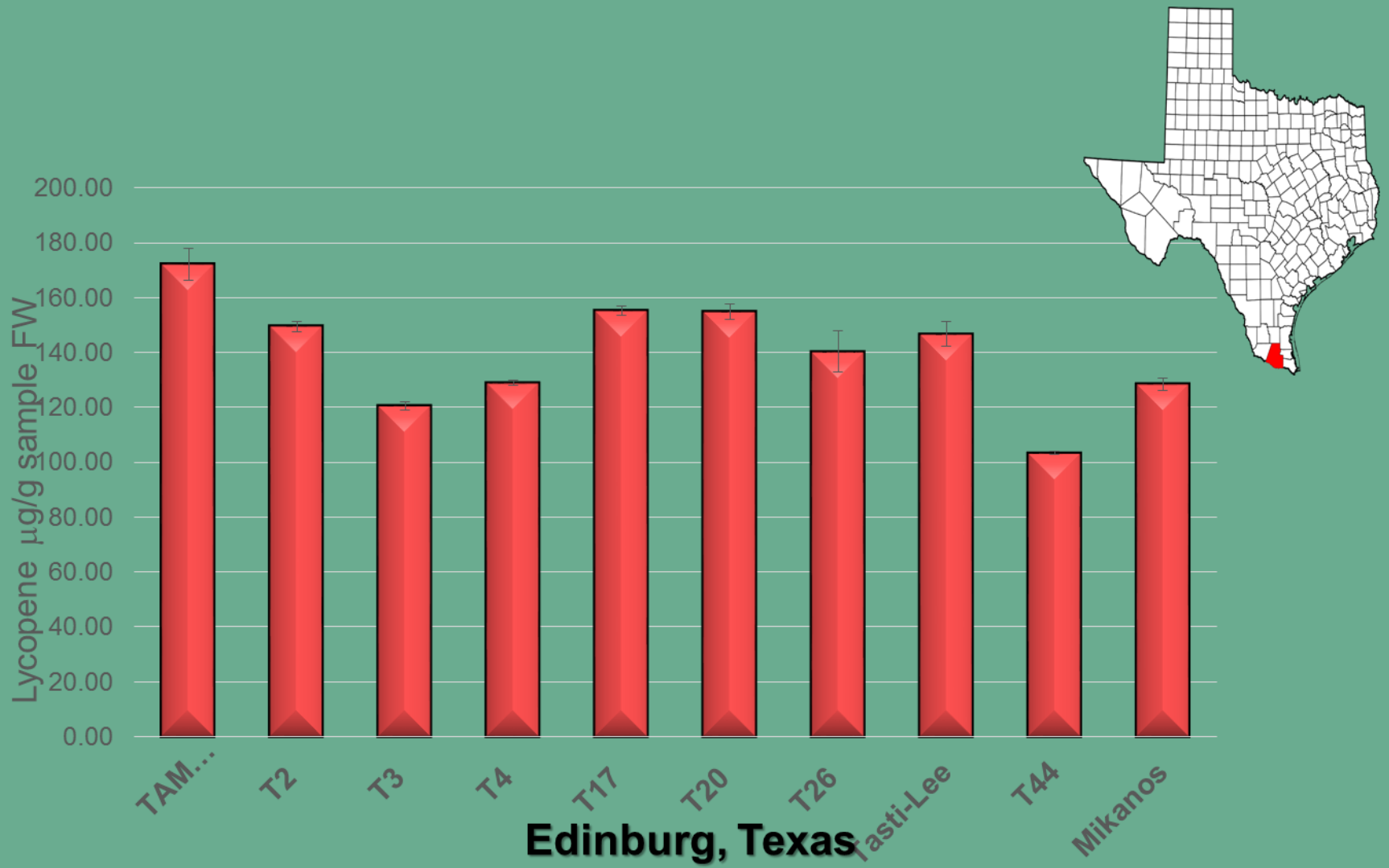
16.1
OK03.800
15.5
OF20.350
9.2
OR15.1200
23.0
OQ15.400

TAMU Tomato Breeding

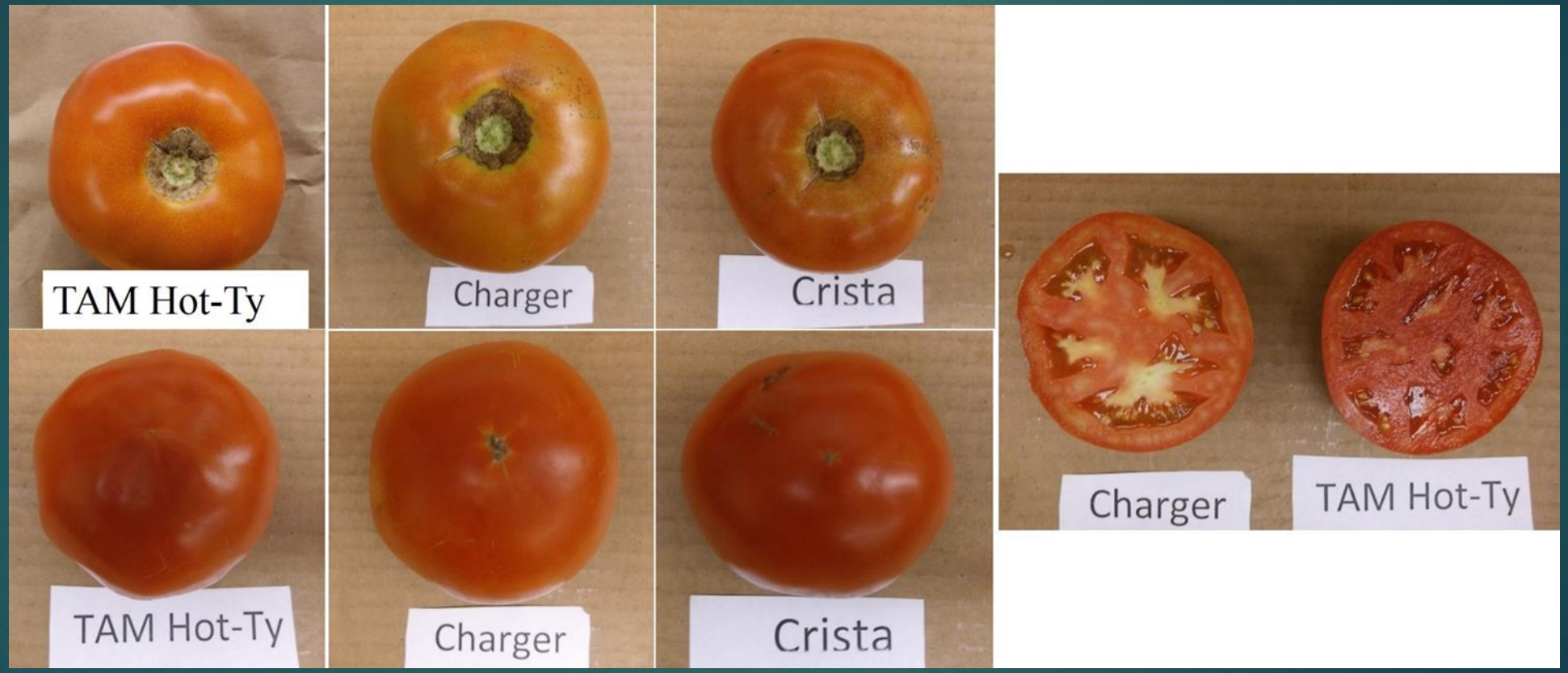
- Efforts to breed improved, heat tolerant processors began in 1940's- Paul Leeper
- Historically focused on canning industry
- Currently more focus on fresh market and specialty types

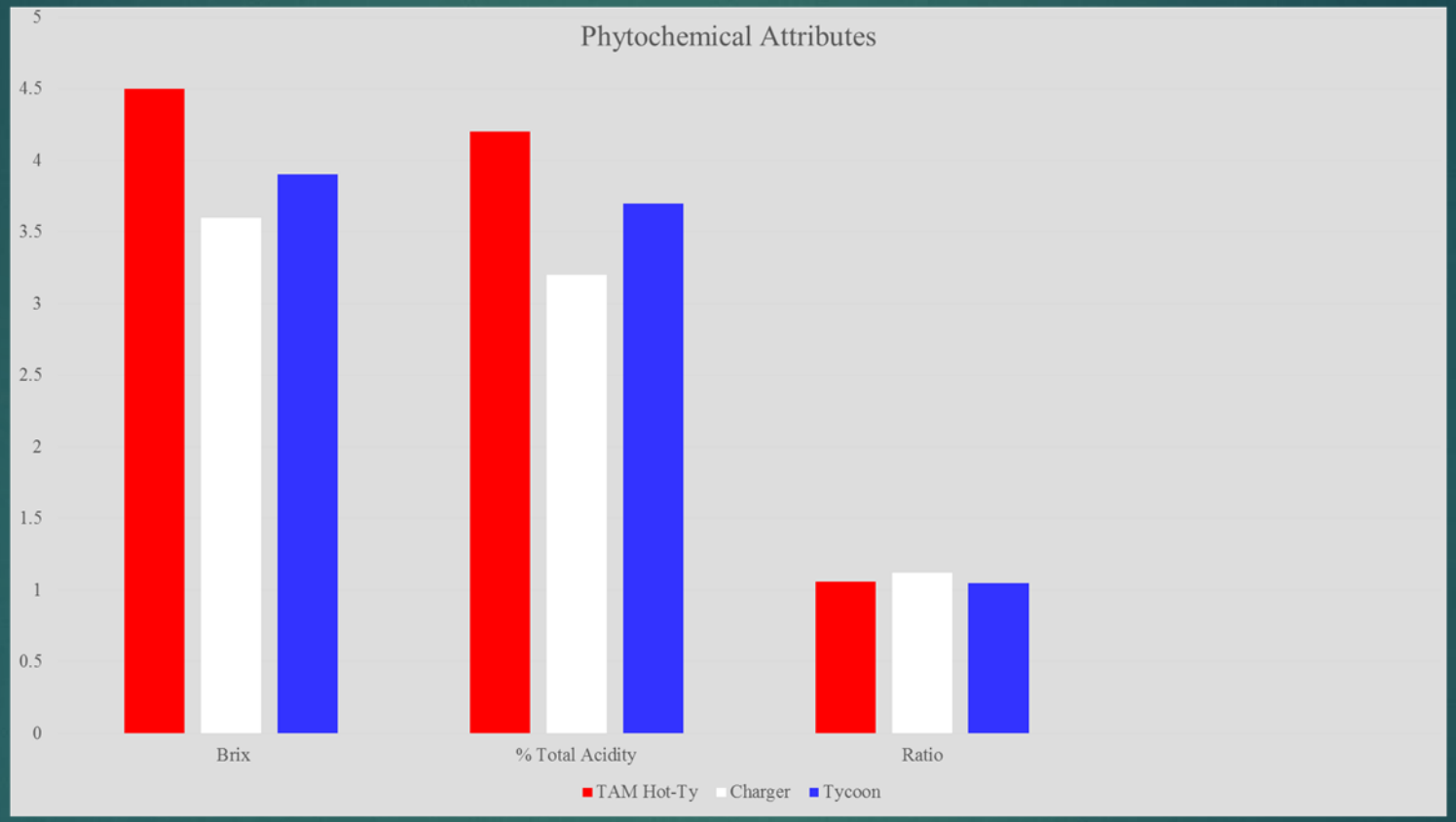
Levels of Lycopene in tomato varieties



TAM Hot-Ty

Attributes: very heat tolerant, TY and F1-2 resistance, hi yield, determinate, 180 g +, first early, 7-10 days before Tycoon and Charger




Phytochemical Attributes
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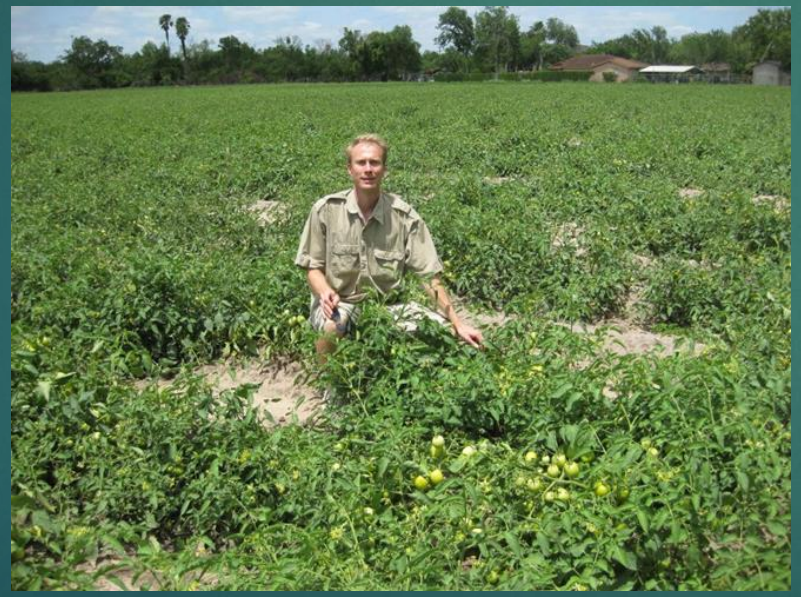
4.5
4
3.5
3
2.5
2
1.5
1
0.5
0
Brix
% Total Acidity
Ratio
TAM Hot-Ty
Charger
Tycoon

Processing Tomato

T5- high lycopene and acid; resists heat, TY, FCR, St



Onion Breeding Priorities

- Bulb Quality- size, shape, color, single centers
- Pink root resistance- improved root systems
- Stress tolerance- drought, heat, cold, bolting
- Yield and early maturity

Pink Root (*Phoma terrestris*)

Susceptible

Resistant

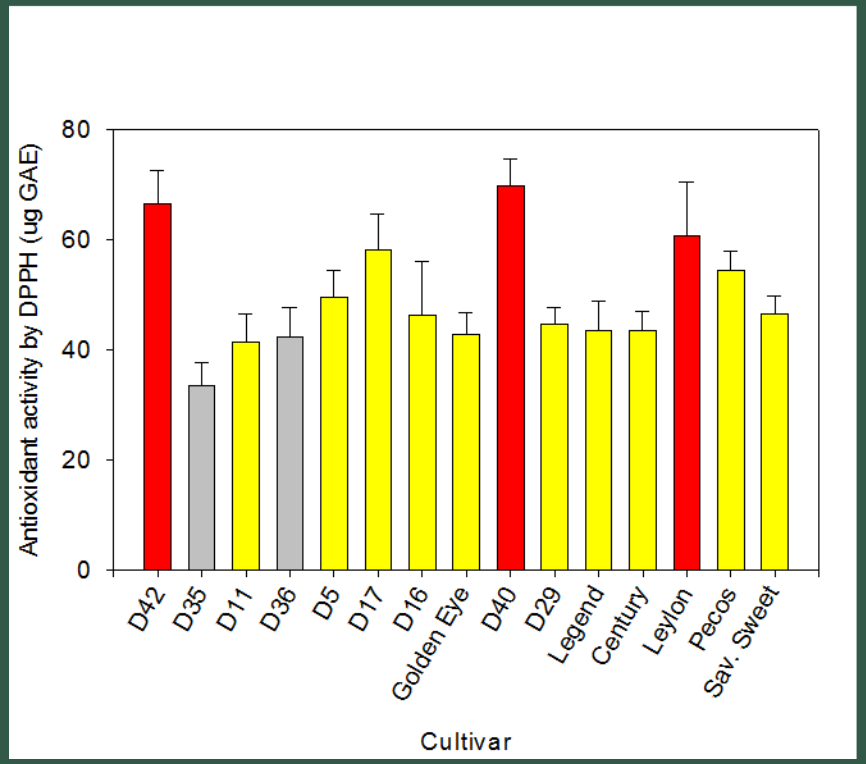
Less Damage and Larger Root System

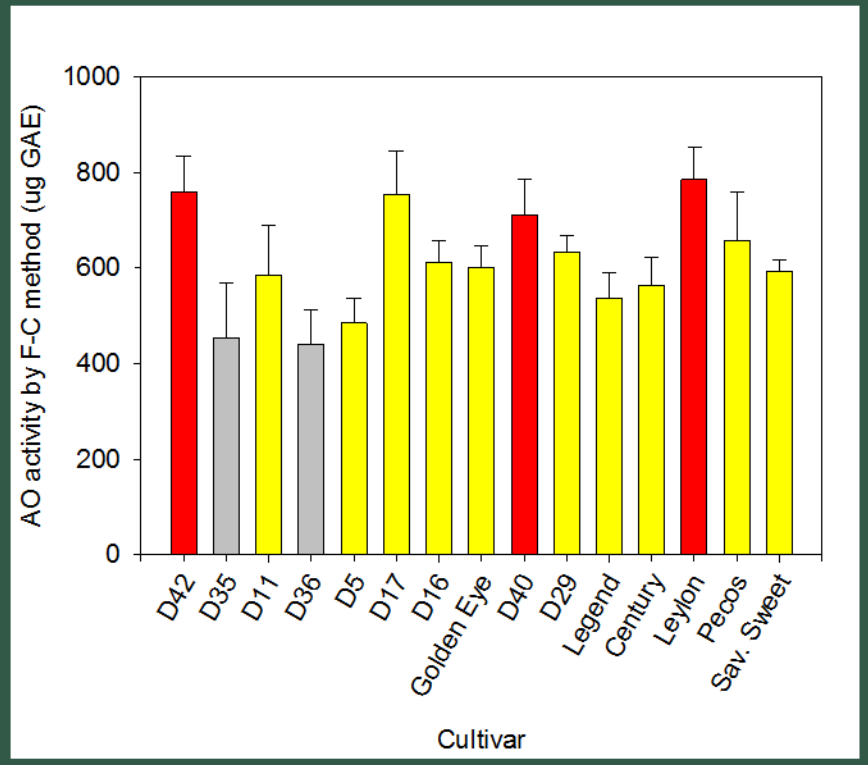
Yield on Drip at Weslaco

Entry	Yield (lbs/ac)	Maturity	Pyruvates
Legend	22256	Early	1.8
Lancelot	35658	Mid	3.3
TX 1015y	26050	Mid	4.2
BL 5015 y	43508	Early	1.4
TEW	33958	Late	5.7
EMW 55107	40254	Early	5.2
BL 5002 w	32661	Mid	2.8
BL 5009 w	32118	Late	2.4
BL 5001 r	32373	Early	5.7

Antioxidant activities of selected shortday onions with yellow, red, or white colors

Red onions tend to have higher AO, while white onions had lower AO.
Thus, red onions are considered healthier than yellow or white onions.



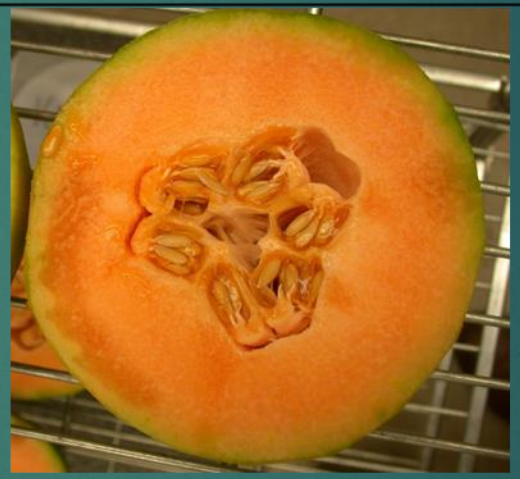


Genotype x Environment

- Cultural practices and environment will impact levels of beneficial phytochemicals
- Levels of carotenoids were higher in peppers grown at Uvalde (Lee et al., 2005)
- Fertilizers (K) and irrigation regimes impact levels

Treatment	°Brix	Ascorbic Acid	β-Carotene	Sucrose	Total sugars
		mg·100g^{-1}fwt	µg·g^{-1}dwt	mg·g^{-1}dwt	mg·g^{-1}dwt
Control	8.8b	29.8 b	302.6 c	317 b	594 b
Suppl. K	9.8a	33.6 a	348.3 a	426 a	850 a

+Suppl. K



Control

Cultivar Development

- Ultimately, new cultivars must be released to deliver the health benefits to the public
- These must have improved quality and good yield, disease resistance, etc.

Habanero β-carotene

TAM Mild- 760 µg/100g Yucatan- 20 µg/100g

High Capsaicin!

‘Caro-Tex 312’ F1 Hybrid

‘Helios’ F1 Hybrid

‘TAM Pacal’ orange casaba

- Higher in beta-carotene than standard casaba
- Higher in super oxide dismutase than other inodorus melons
- Resistant to powdery mildew

TAM Pacal

Caroline

More New Cultivars

TAM Dulcito

Pearl

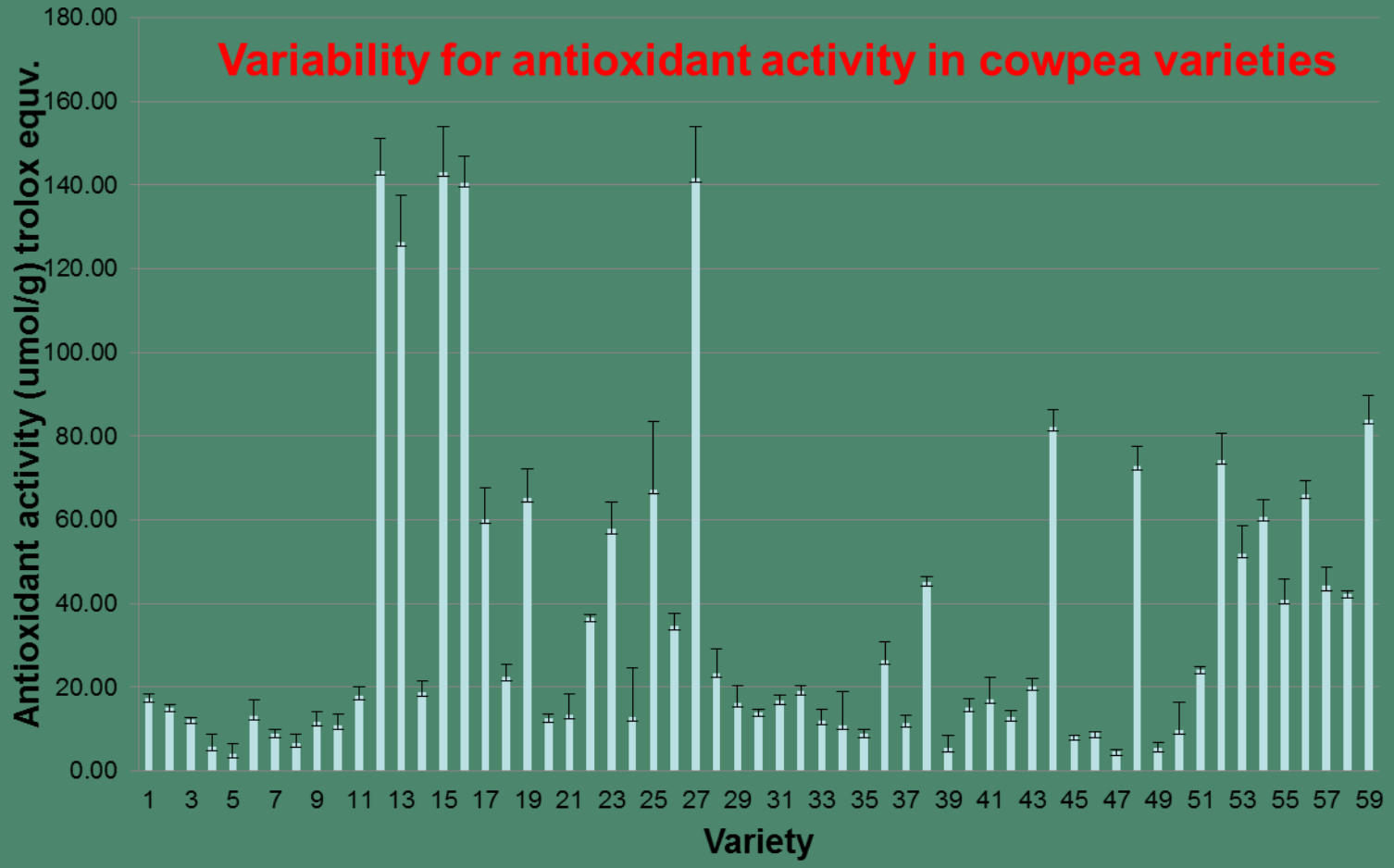
TAM Ben Villalon

TAM Mild Habanero

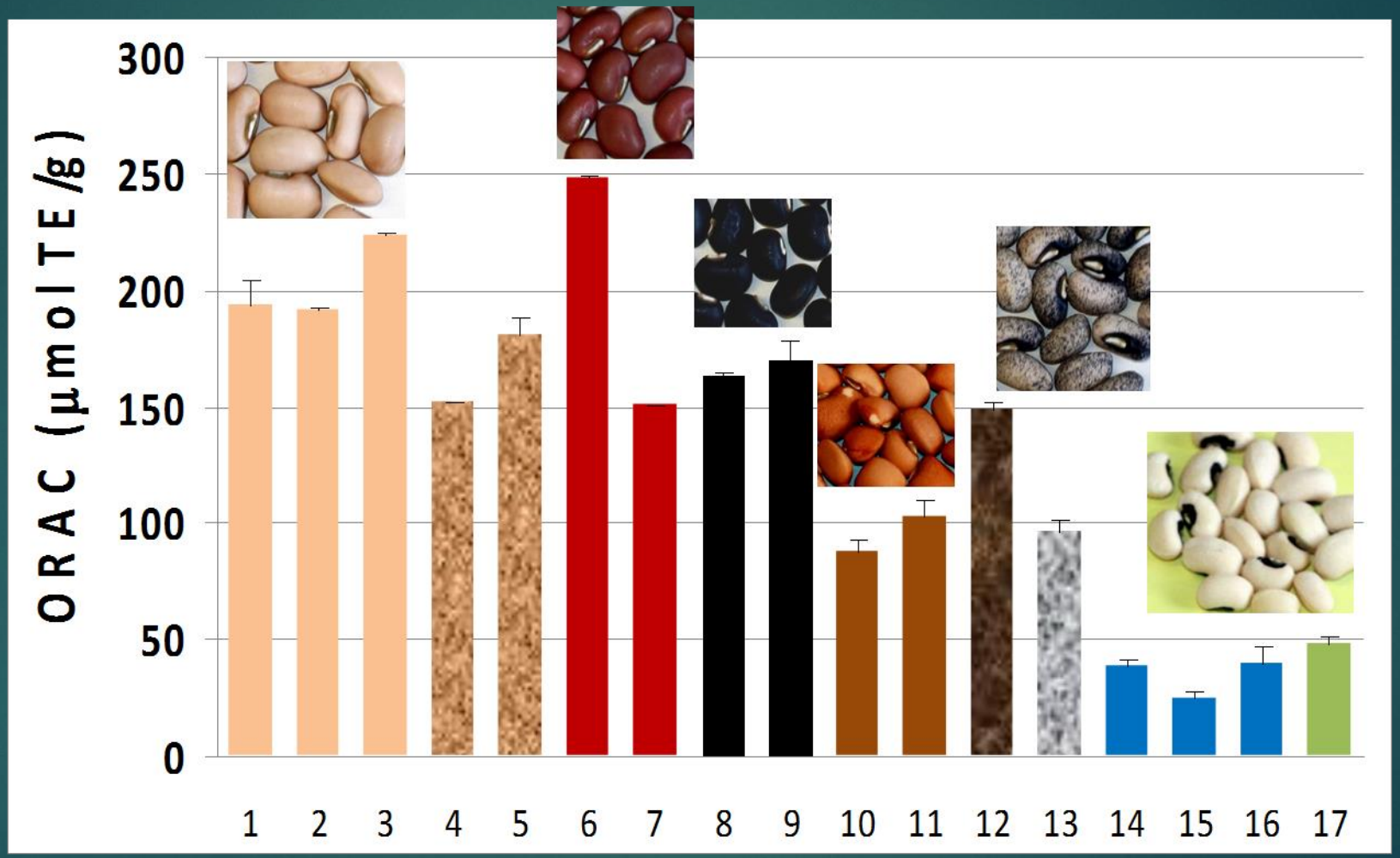
Breeding High Yielding Cowpea Varieties with Improved Seed Quality and Enhanced Nutritional and Health Factors.

B.B. Singh (bsingh@ag.tamu.edu), Visiting Professor, Texas A&M University and G.B. Pant University.

Breeding for Health-related Compounds



Association between seed color and antioxidant activity



Cowpea as poor man's meat and rich man's health food

New cowpea varieties have up to 30% protein, and rich in calcium, iron, zinc, complex carbohydrates, soluble fibers and full of antioxidants:

good for health and heart