

# P. Chaires, Industry Introduction: The Situation

- HLB Endemic Environment
- Short Supply to feed processing infrastructure
- Blending challenges for NFC
- Strong need for high quality early maturing selections
- Recognition that perhaps the best opportunity to accelerate development of varieties with processing potential is through hybrids
- What we know as sweet orange is itself a hybrid.
- The juice of some hybrids is indistinguishable from orange – and have quality characteristics that may improve NFC
- Some promising hybrids are showing useful levels of HLB tolerance, but can't be used in significant quantities under the current standards.

# Initiating the Conversation

April 18, 2019

- A small group gathered at the FDOC, Bartow
  - Florida Citrus Processors Assoc., NVDMC, FL Citrus Mutual, FDOC staff, CRDF, UF-IFAS and USDA breeding teams, two processing brands.
- Initiated a conversation:
  - The Genetics. What is a sweet orange?
  - How might hybrids help restore supply and improve processed products
  - Advantages and disadvantages/risks to changing the Standards of Identity
  - How might changes be made if the industry elects to do so?
  - International implications
- Amending the standard is one option for discussion in connection with developing HLB resistant trees-- the ramifications should be fully investigated (i.e. economic impact etc.) before any industry decision to move forward
- Its now time to take this conversation to industry.

# **HLB-Tolerant Hybrids: update & potential for supplementing Florida OJ production?**

**A collaborative effort of the Florida citrus industry  
and research community**

**Fred Gmitter, Ed Stover, Liz Baldwin, Jude Grosser,  
Jinhe Bai, Yu Wang, Peter Chaires, Juan Carlos  
Motamayor**



# US Citrus is near monoculture

NASS 2017 (2016-17 data)

US production: 66% sweet orange, 13% mandarin, 11% lemon, 9% grapefruit (some of the most important mandarins and all grapefruit have sweet orange in pedigree)

FL citrus    -88% oranges  
                  -10% grapefruit

When a new pathogen enters a monoculture of a susceptible genotype, strong potential for a severe epidemic

# Citrus Breeding and HLB

- Initially the wisdom was that HLB would kill ALL citrus. Tolerant selections have become obvious over time, as HLB spread throughout Florida's citrus breeding populations
- There is useful tolerance to HLB in conventional citrus and even stronger resistance in citrus relatives
- Caution: HLB is a difficult disease to study and what we "know" changes. At first, grapefruit seemed less sensitive than sweet orange, now we know grapefruit is among the most sensitive
- Breeding programs have been selecting more tolerant types from pre-existing populations, and making new crosses specifically targeting HLB tolerance for future selection

# HLB Tolerant Citrus

- There is good news here for growers of some mandarin-types!
- But Florida citrus is founded on Orange Juice (and grapefruit)
- First we will provide some evidence for HLB-tolerance
- We will then address the issue of using HLB-tolerant citrus to supplement the OJ stream
  - What are current legal implications?
  - How might federal regulations be changed to provide opportunities to use new hybrids in OJ?
  - What do we need to know?





**Sugar Belle® near Vero Beach, HLB+ >8 years !**

# But what does tolerance mean?

- It should be noted that earlier studies reported blotchy mottle symptoms and CLas titer
- It is now apparent that some citrus types grow and produce normal fruit even with high levels of blotchy mottle and CLas
- We will likely have a 20-50 year evolution in planting material to combat HLB, ultimately resulting in immunity
- Growth rate / canopy health are important, but ultimately yield and quality are paramount
- For now, what matters most is economic tolerance: the ability to produce sufficient yield of high quality fruit so that production is profitable





**Sugar Belle®: Normal fruit with blotchy mottle**

We showed considerable differences in HLB-tolerance in existing plantings. What if trees are exposed to CLas at planting?

> 6 yr replicated trial, scion/rootstock comparison

>CLas titers not significantly different HortScience 51:127-132

			Fruit/tree		Health		Change in	
<u>Scion/Rootstock</u>	<u>Mortality (%)</u>		<u>Oct 2015 (no.)</u>		<u>Oct 2015 (3 pt)</u>		<u>diam. (mm)</u>	
Fallglo/Kinkoji	20	a	28.4	b	1.9	cd	23.8	b
Hamlin/Cleopatra	20	a	18.6	bc	2.2	bc	20.4	b-d
Hamlin/Kinkoji	10	a	12.9	cd	1.9	cd	14.5	d
Ruby/Kinkoji	10	a	4.6	e	1.6	d	20.7	bc
SugarBelle/Sour	0	a	81.3	a	2.9	a	46.1	a
Tango/Kuharske	0	a	88.1	a	2.9	a	32.2	a
Temple/Cleopatra	18	a	35.6	a	2.3	ab	23.8	b

- Some scion/rootstock combinations continued to develop even with high titers of CLas and and strong mottle symptoms
- Sugar Belle<sup>®</sup>/Sour had significantly higher mottle than others
- Not “tolerant” rootstocks used so likely a scion effect





**Sugar Belle® in SW Florida, 4 years old**



# 411 Mandarin Hybrid – Resurgence against HLB!



UF 411 mandarin hybrid has shown a resurgence against HLB in various locations and on multiple rootstocks. From the cross of tolerant LB8-9 (Sugar Belle®) x sensitive Murcott.



Comparison of 50 Selections and Cultivars at Picos Farm:  
Rigorous challenge: no-choice ACP, ACP house, then field.  
At 5.5 yrs in field some are quite healthy and have grown well, while others are sickly and stunted.





## After 5.5 years in Ground: Tolerance observed from mandarin and Poncirus hybrids

		Canopy		Tree health		Canopy		Canopy
		density (%)		(5 is best)		vol (m <sup>3</sup> )		vol RGR (%)
FF1-42-70	Fortune x Encore	98.3 a		5.0 a		10.9 b-g		94 a-k
Bower	Clem x Orlando	98.0 a		4.8 a-c		9.3 c-i		103 a-g
FP6-47-119	Orange-like w/Pt	97.5 ab		4.6 a-e		17.2 ab		128 a-c
FF1-4-2	Complex w/Pt	97.0 ab		4.2 a-g		19.9 a		114 a-e
FF1-34-11	5-51-2 x 1-57-105	96.7 a-c		4.8 ab		6.6 c-j		98 a-i
Nova		96.0 a-c		3.9 a-h		3.0 h-j		51 d-o
JacksonGF		95.0 a-c		4.3 a-f		8.0 c-j		95 a-k
Clementine		94.0 a-c		4.6 a-e		7.0 c-j		144 a
FP6-49-116	SunDragon-sib	93.3 a-c		4.6 a-e		13.7 a-c		84 a-m
SunDragon		93.0 a-c		4.6 a-e		12.2 b-e		79 a-n
Valencia		90.0 a-c		4.1 a-g		5.5 d-j		28 i-o
US119	Complex w/Pt	88.8 a-c		4.6 a-e		6.6 c-j		102 a-h
FF5-51-2	Clem x Orlando	88.8 a-c		3.8 a-h		3.0 h-j		80 a-n
Temple		85.0 a-d		3.5 a-i		4.8 f-j		33 g-o
USEarlyPride		82.5 a-d		4.1 a-g		5.7 d-j		117 a-d
Carrizo		75.0 a-e		3.9 a-h		4.9 e-j		36 f-o
Flame		55.0 ef		2.0 ij		2.3 ij		26 j-o
Sunburst		53.8 ef		3.1 d-i		1.7 j		10 no
USSurprise		50.0 f		2.8 f-j		7.8 c-j		8 o



**Ftp-4-13-31\***



**Ftp-4-13-7\***



**Ftp-6-13-39\***



## **Low seeded selections of Irradiated FF-5-51-2 (Clementine x Orlando)**

- Seedy original hybrid growing many places and appears to have useful HLB-tolerance, with no evidence of HLB-affected fruit
- Good midseason tangerine (late Nov early Dec)
- Easy peeling, rich flavor, segments dry when separated. Dancy+ size.
- Appears to require cross pollination
- Trialing all 3 selections in case one has problems
- May release parent 2019



# Aerial view of 5-51-2 surrounded by Early Pride



**HLB-tolerance in some but not all mandarin types.**

**BUT there is another major source of tolerance...**



*Newly released*

*First released scion for fruit use  
containing Poncirus (1/8  
Poncirus) .*

US SunDragon

Like Navel in alligator-hide

Strong tolerance to HLB at  
many locations

Released with focus on  
dooryard/niche/breeding

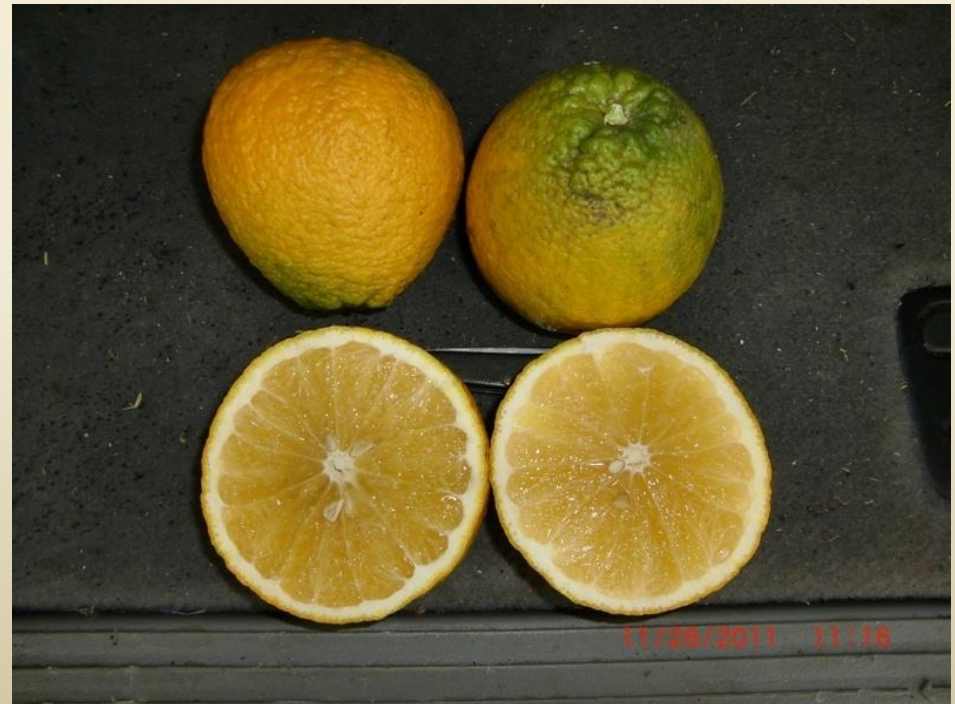
Scored highly in sweet orange  
juice trial at USHRL

Used in many crosses.

Hybrids starting to fruit!



**USDA/ARS  
Ft. Pierce, FL**





## US Sun Dragon 6-year-old trees at USDA Picos Rd Farm

**USDA/ARS  
Ft. Pierce, FL**





# SunDragon progeny selections-2018



**USDA/ARS  
Ft. Pierce, FL**

Not only interesting as a cultivar but as  
a parent as well

Recently showing promise for juice!

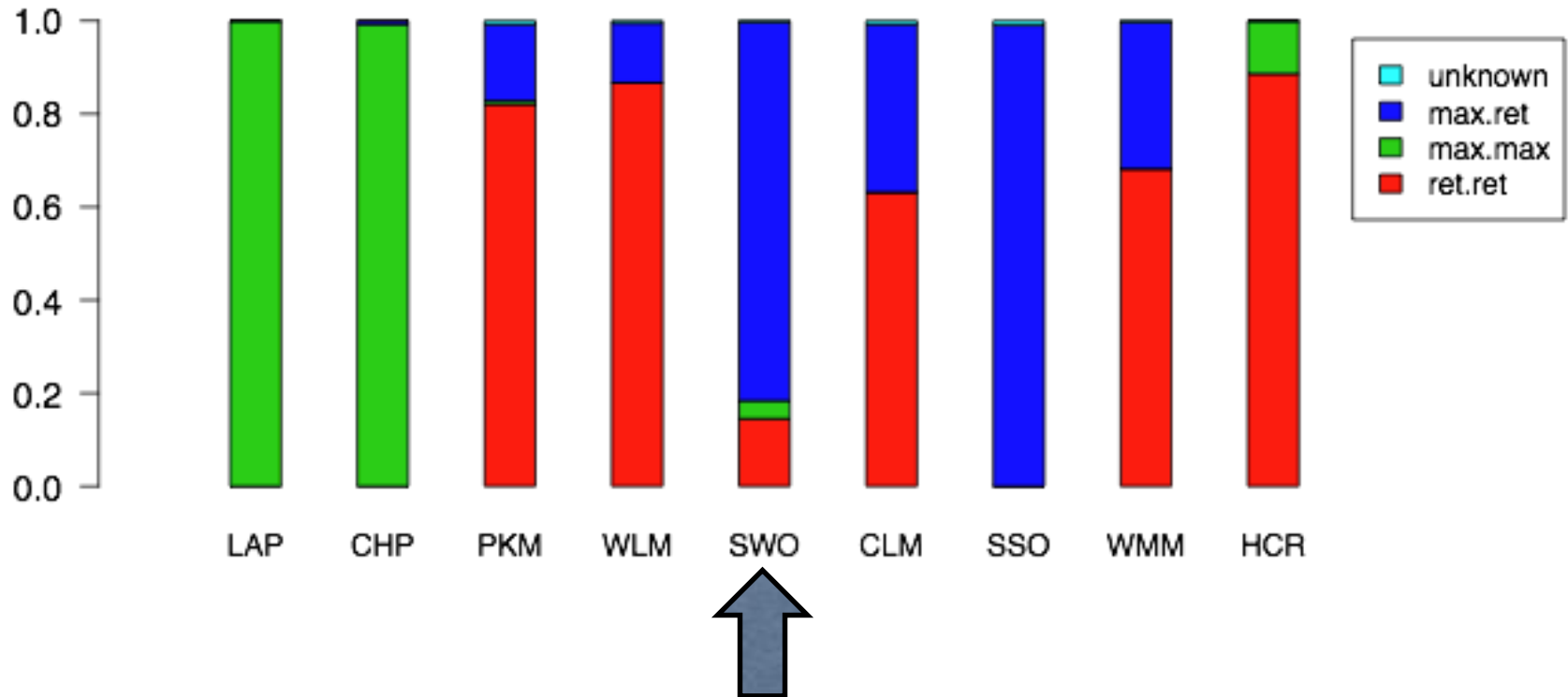




**This fruit produced juice indistinguishable from commercial sweet oranges. Midseason maturity with 37 color score and high °Brix. Recent Display Day results: blended with a high quality sweet-orange like mandarin, was the most preferred among all juice samples.**

**A seedless hybrid, 8% Poncirus**

# Genomic Makeup of Sweet Orange



***Sweet orange SWO = ((Max x Ret) x Max) x PKM***

*Max = pummelo*

*Ret = mandarin*

*PKM = Ponkan-like (also contains Max)*

**Ret:Max ~ 60:40**

# New “Sweet Orange” Hybrids Are Being Developed by Breeding

- All “true” sweet oranges are derived by accumulated mutations from a single original hybrid.
- Citrus sinensis* is NOT a true species; the original hybrid was comprised of ~ 60% mandarin and 40% pummelo.
- New sweet orange-like hybrids are derived from a similar lineage, and some have been found already with greater tolerance and are currently being tested.



- To be designated a sweet orange, it may be critical to have similar aroma/flavor
- New USDA hybrids shown closer to Hamlin than is Ambersweet and reported in refereed paper
- UF breeding also producing such hybrids

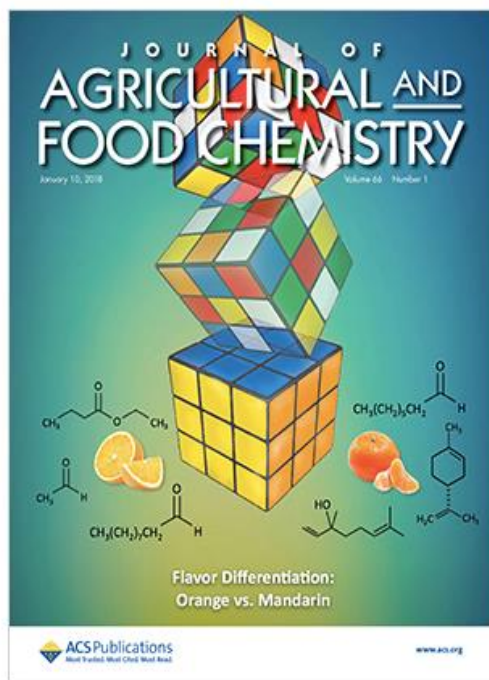
## **Volatile profile comparison of USDA sweet-orange-like hybrids vs. ‘Hamlin’ and ‘Ambersweet’**

Jinhe Bai, Elizabeth Baldwin

Randall Driggers, Jack Hearn and Ed Stover



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### About the Cover:

Sweet orange and mandarin fruits are consumed worldwide. In this study, we preliminarily decoded the characteristic flavor differences between them. Ethyl butanoate, ethyl 2-methylbutanoate, octanal, decanal, and acetaldehyde were revealed to be essential for orange-like aroma, whereas linalool, octanal,  $\alpha$ -pinene, limonene, and (*E,E*)-2,4-decadienal were considered key components for mandarin-like aroma. Furthermore, an unreleased mandarin hybrid producing fruit with orange-like flavor was used to validate the identification of characteristic volatiles in orange-like aroma.

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## Differentiation between Flavors of Sweet Orange (*Citrus sinensis*) and Mandarin (*Citrus reticulata*)

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### Supporting Information

**ABSTRACT:** Pioneering investigations referring to citrus flavor have been intensively conducted. However, the characteristic flavor difference between sweet orange and mandarin has not been defined. In this study, sensory analysis illustrated the crucial role of aroma in the differentiation between orange flavor and mandarin flavor. To study aroma, Valencia orange and LB8–9 mandarin were used. Their most aroma-active compounds were preliminarily identified by aroma extract dilution analysis (AEDA). Quantitation of key volatiles followed by calculation of odor activity values (OAVs) further detected potent components (OAV  $\geq 1$ ) impacting the overall aromatic profile of orange/mandarin. Follow-up aroma profile analysis revealed that ethyl butanoate, ethyl 2-methylbutanoate, octanal, decanal, and acetaldehyde were essential for orange-like aroma, whereas linalool, octanal,  $\alpha$ -pinene, limonene, and (*E,E*)-2,4-decadienal were considered key components for mandarin-like aroma. Furthermore, an unreleased mandarin hybrid producing fruit with orange-like flavor was used to validate the identification of characteristic volatiles in orange-like aroma.

**KEYWORDS:** sensory analysis, Valencia orange, LB8–9 mandarin, aroma extract dilution analysis, aroma profile analysis, orange-like flavor mandarin hybrid



# **NIFA Project: Accelerating implementation of HLB tolerant hybrids as new commercial cultivars for fresh and processed citrus**

**Elizabeth Baldwin, Ed Stover, Jinhe Bai, Anne Plotto, John  
Manthey, USDA-ARS Horticultural Research Laboratory  
Rhuanito Ferrarezi, Fred Gmitter and Yu Wang, CREC, UF  
Mike Roose, UC Riverside  
Goutam Gupta, New Mexico Consortium**





# NIFA Project: Focused on bringing HLB-tolerant cultivars to the industry

**Screening for tolerance:**  
Screen for HLB in the field, develop resistance markers, release tolerant cultivars

**Screening for flavor:**  
Screen hybrids for orange/grapefruit like and overall citrus flavor quality

**Classification:**  
Work with industry to classify hybrids as orange or grapefruit  
And develop blends

↑ Yield



# The Current Situation

- OJ standards require at least 90% sweet orange
- Genomics have unequivocally shown that sweet orange is not a true species, but a single hybrid
- No other industry is limited by law to such a narrow genetic base
- Does it make sense to be limited legally by an antiquated and arbitrary taxonomic designation?

# How do we make use of HLB-tolerant types?

- Standard of Identity Orange Juice-146.135 - CFR - Code of Federal Regulations Title 21
- “Orange juice is the unfermented juice obtained from mature oranges of the species *Citrus sinensis* or of the citrus hybrid commonly called "Ambersweet"
- Within existing regulations, we could use the “Ambersweet model” to have individual hybrids classified as sweet oranges based on aroma volatiles
- Sugar Belle juice already shows potential utility, at least for blending under current standards



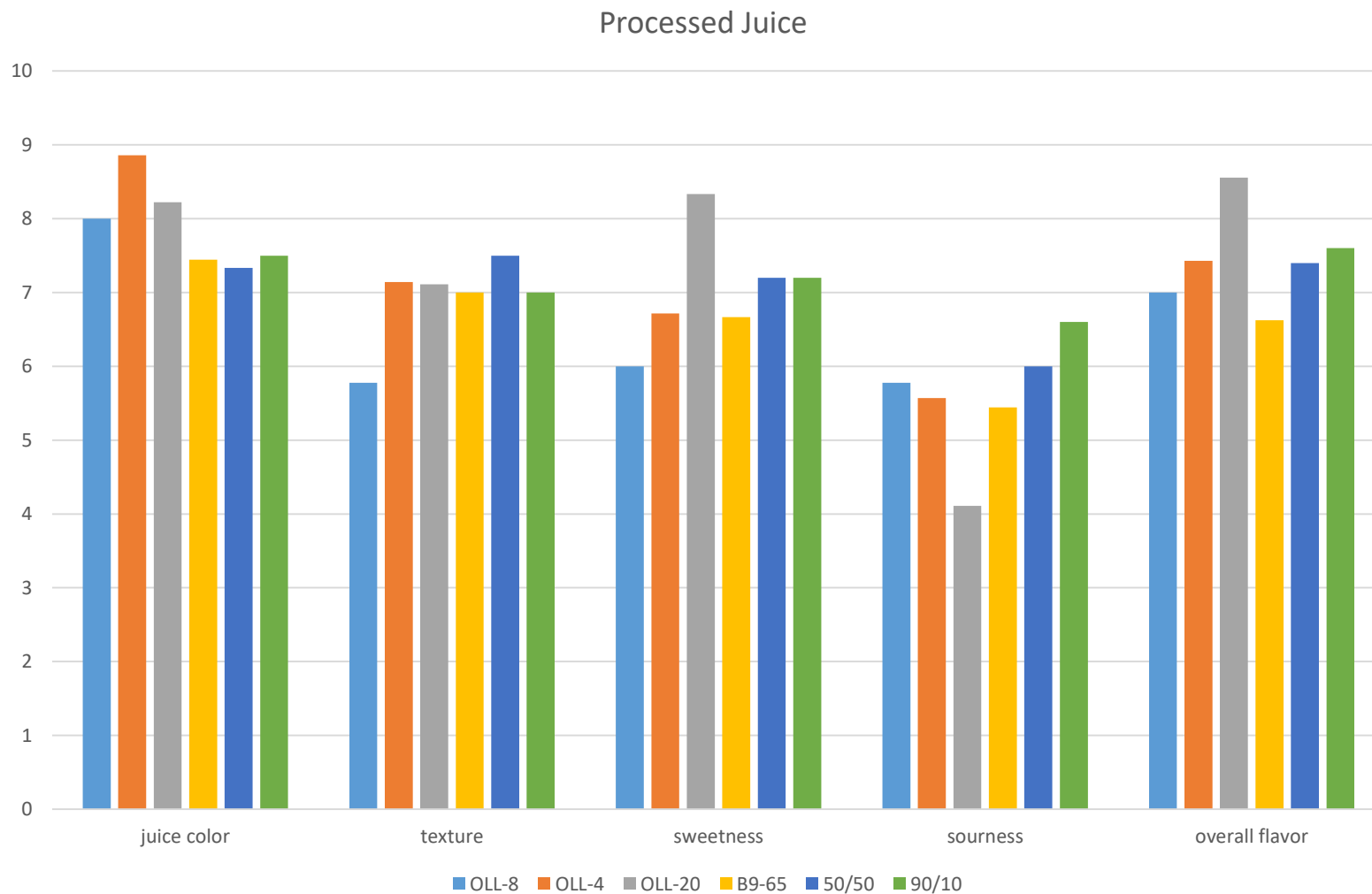
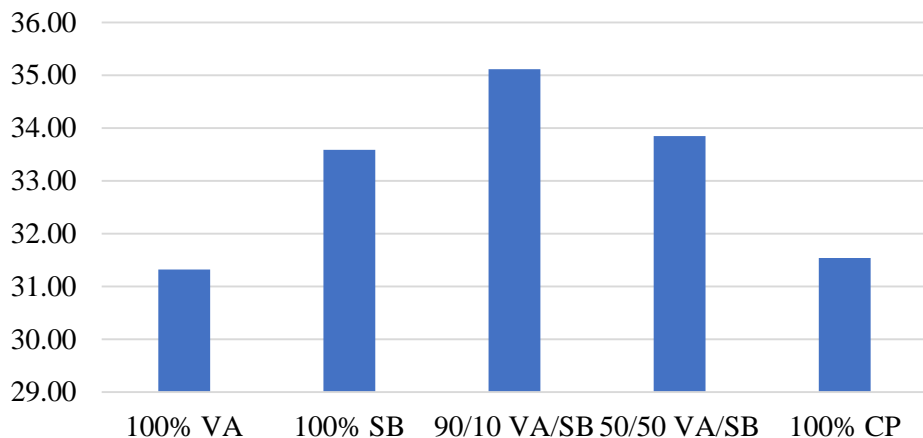
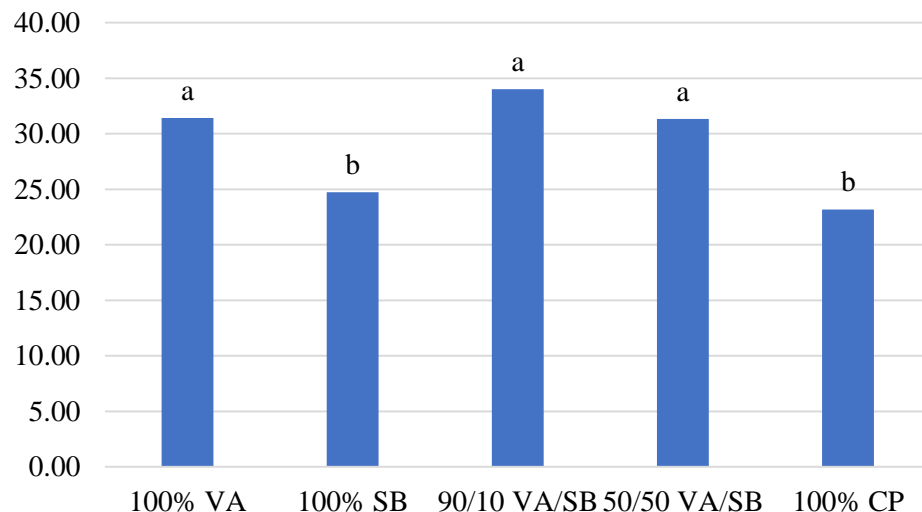


Figure 2. February 19 2019 Juice Display Results (subjective mean scores from display attendees; scale of 0-10 with 10=perfect score). 50/50 is a 1:1 blend of pasteurized Valquarius and LB8-9 Sugar Belle® juice; 90/10 a 9:1 blend of the same.

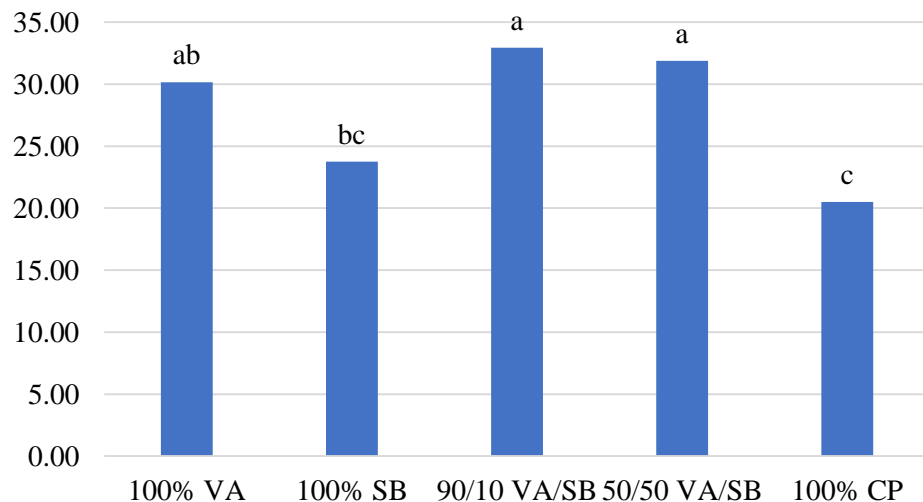
Overall Appearance



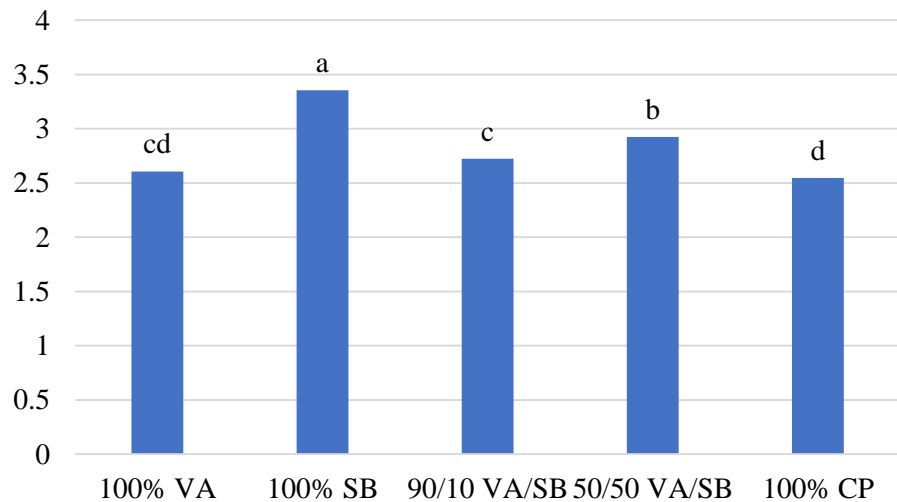
Overall Liking



Overall Flavor Liking

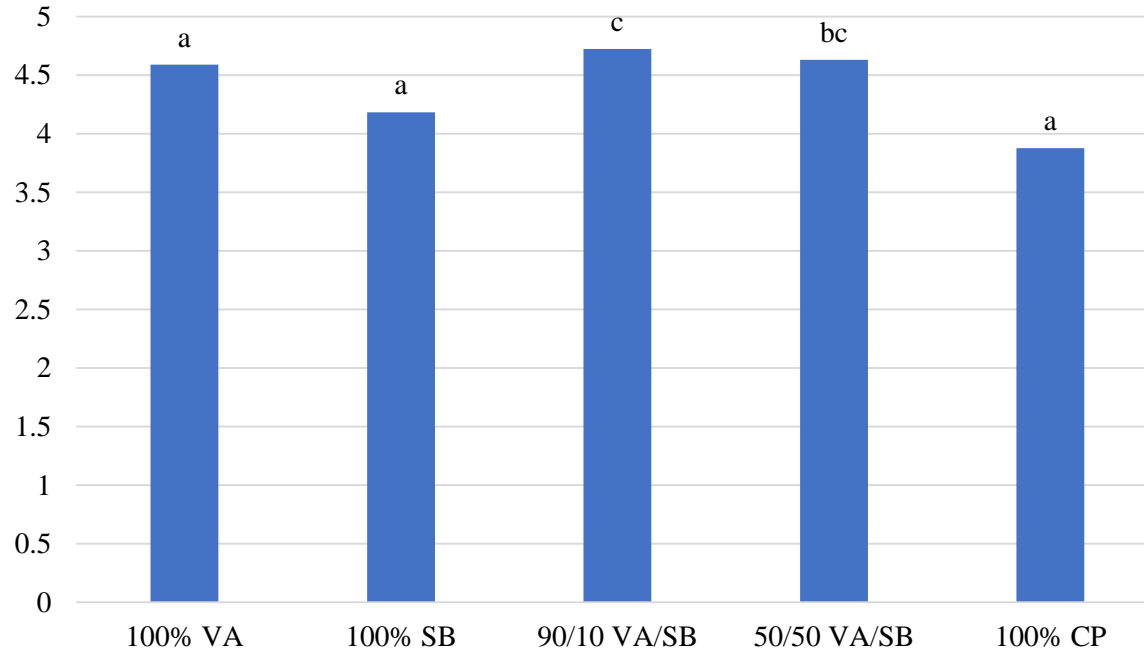


Color



**100% VA: 100% Valquarius; 100% SB: 100% Sugar Belle; 90/10 VA/SB: 90% Valquarius & 10% Sugar Belle; 50/50 VA/SB: 50% Valquarius & 50% Sugar Belle; 100% CP: 100% commercial product.**

# Price willingness to pay



**100% VA: 100% Valquarius; 100% SB: 100% Sugar Belle; 90/10 VA/SB: 90% Valquarius & 10% Sugar Belle; 50/50 VA/SB: 50% Valquarius & 50% Sugar Belle; 100% CP: 100% commercial product.**

# Using tolerant hybrids: flexibility for innovation & maintaining quality-1

- No limitations to producing “Citrus Juice” but there is enormous value in the name “Orange Juice”
- No other industry is limited by law to such a narrow genetic base. Not a true species but a single hybrid
- The 90% sweet orange OJ standards might benefit from reconsideration.
- Examples from the wine industry are supportive: varietal designation in US wine only requires 75% composition from the designated variety, and only 51% for designation as a *Vitis labrusca* variety (CFR, 1996).

## Using tolerant hybrids: flexibility for innovation & maintaining quality-2

- Consider broadening the definition of orange to reflect that it is just one reticulata & maxima introgression hybrid
- Regulatory language like: “Orange juice is the unfermented juice obtained from mature fruit of the sweet orange (conforming to the genotype known as *Citrus sinensis*), or from fruit with an ancestral interspecific pedigree similar to that of the sweet orange and with color, flavor and organoleptic properties typical of sweet orange.”



# Using tolerant hybrids: flexibility for innovation & maintaining quality-3

- Many of the HLB-tolerant selections identified are early to mid-season and may provide an invaluable supplement to Hamlin/Mids
- Ultimately we MAY want to open the door to even broader tolerant hybrid use.
- Perhaps the individual fruit need not be similar to sweet orange as long as the ultimate blend is very similar to OJ?
- While the focus of this talk is OJ, the same thinking can be applied to GFJ

# Considering Hybrids for Processing

- This ABSOLUTELY must be an industry-driven process, and we are hoping that this information stimulates discussion
- Clearly, implementation requires substantial research including:
  - production
  - quality parameters
  - acceptable maturity windows
  - transportation/handling (softer fruit?)
  - blending for uniform products
  - consumer acceptance

# Work underway

- With grower/collaborators there are large scale trials being planted to provide critical information and material for processing tests
- UF, USDA, and industry are all conducting small-scale assessments of processing potential
- Any change that occurs will be implemented over many years with successes and disappointments
- Please understand, we do not claim to have the ideal solution ready for implementation.
- Due to the time required for new plantings and changes in regulations, we need to start discussions and evaluations now!





**THANK YOU!**