

Carolina Strongback: A Fusarium wilt and Root knot Nematode resistant *Citrullus amarus* rootstock for watermelon production

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Fusarium wilt is one of the most devastating diseases of watermelon



- Causal agent: *Fusarium oxysporum* f. sp. *niveum* (*Fon*)
- Pathogenic races of *Fon*: 0, 1, 2, and 3
- *Fon* race 1 resistance has been incorporated into many cultivars
- There are no edible cultivars with resistance to *Fon* races 2 or 3

Root knot nematode (RKN) (*Meloydogyne spp.*)

- RKN is the most destructive nematode of watermelon in the U.S.
- Cultivated watermelon (*Citrullus lanatus*) is susceptible to RKN.
- Commercial bottle gourd, pumpkin, and Interspecific hybrid rootstocks are susceptible to RKN

Reniform nematode (*Rotylenchulus reniformis*)

- Reniform nematode causes damage on numerous cucurbits
- Found in tropical & subtropical areas
- Rio Grande Valley in Texas and throughout the southeastern U.S.
- Can reduce fruit size and lower sucrose levels

Fusarium wilt and plant parasitic nematodes

- *Fusarium oxysporum* f. sp. *niveum* can remain viable in soil for more than 30 years.
- Most growing areas in the southeastern US have issues with southern root knot nematode.
- The loss of methyl bromide as an affordable soil fumigant has required a search for alternative control options.

Soilborne disease and pest control options:

- Chemicals
- Biological control
- Plant host resistance
- Grafting

WATERMELON

- *Citrullus lanatus*- Cultivated watermelon
 - Sweet (High Brix)
 - Mostly red flesh
 - Mostly large fruit
 - Often susceptible to pathogens and pests

Citrullus amarus- Citron melon

- Not sweet (often bitter)
- Mostly green or white
- Resistance to many pathogens and pests

Citrullus amarus is a valuable genetic resource, particularly for disease resistance alleles

Fusarium wilt Race 2



ROOT KNOT NEMATODE

Meloidogyne incognita

ROOT STOCK ID	TYPE	REACTION
Macis	<i>Lagenaria</i>	Susceptible
FR Strong	<i>Lagenaria</i>	Susceptible
WMXP 3945	<i>Lagenaria</i>	Susceptible
Emphasis	<i>Lagenaria</i>	Susceptible
Geo Sprint	<i>Lagenaria</i>	Susceptible
Ojakkyo	<i>Citrullus amarus</i>	Least Susceptible
Shintosa Camel	Inter-specific Hybrid	Very Susceptible
WR-15006	Inter-specific Hybrid	Very Susceptible
Carnivor	Inter-specific Hybrid	Very Susceptible
WMXP 3943	Inter-specific Hybrid	Very Susceptible
Strong Tosa	Inter-specific Hybrid	Very Susceptible
Gladiator	Inter-specific Hybrid	Very Susceptible
Iron Cap	Inter-specific Hybrid	Very Susceptible

GRAFTING WATERMELONS

Grafting rootstocks for watermelon:

- Bottle gourd
 - Pumpkin
 - Hybrid squash
-
- Not a host for *Fusarium oxysporum* f. sp *niveum* (“Resistant”)
 - Highly susceptible to RKN
 - May impart an off “squash” flavor to the fruit

Goals for a new rootstock

- Reduction in rootstock costs.
- Easy and compatible grafting characteristics.
- Fusarium wilt resistance.
- Nematode resistance.
- High yields.



← **USVL246-FR²**

USVL252-FR²



Genetic Mapping of QTL Associated with resistance to *Fusarium oxysporum* f. sp. *niveum* (Fon) race 2



USVL246-FR² and USVL252-FR²

Breeding lines:

❖ Released in 2015

- Twenty-five thousand seeds provided nationally and internationally, to more than 20 Seed companies
- Published in 2016: HortScience 51:1065-1067
- Being used by several major seed companies in development of Fusarium wilt resistant watermelon cultivars
- USVL246-FR2 has been completely sequenced, annotated and released.

Carolina Strongback



Carolina Strongback

Excellent grafting qualities:

- Uniform seedling growth
- Performs well under LED lights
- Thick hypocotyl
- 90-100% grafting “takes”
- Adapts well to newest grafting methods.



Carolina Strongback



Carolina Strongback

- *Citrullus amarus*
- Developed, PVP and Released, USDA-ARS & Clemson University
- High Levels of Tolerance to *Fusarium oxysporum* f. sp. *niveum* races 1 & 2
- High Levels of Tolerance to Root Knot & Reniform Nematode
- High Levels of Tolerance to Bacterial Fruit Blotch
- Produces an Abundance of Seed
- Open pollinated



YIELD

Rootstock	Fruit Number	Fruit Weight	Average Fruit Wt.	% Fruit Count				
<u>Treatment</u>	<u>per Plot</u>	<u>lb/Plot</u>	<u>lb/Plot</u>	<u>Under 9 Lb</u>	<u>60 Count</u>	<u>45 Count</u>	<u>36 Count</u>	<u>30 Count</u>
Non-Grafted	21.00 a	301.00 a	14.33 a	9.52 a	42.86 b	23.81 b	19.05 b	4.76 b
Carnivor	21.00 a	273.15 b	11.09 b	4.76 b	57.14 b	33.33 a	0.00 c	4.76 b
Strong Tosa	22.00 a	291.71 ab	13.26 a	4.55 b	22.73 c	8.09 c	59.09 a	3.57 b
Macis	14.00 b	137.30 c	9.81 b	4.61 b	70.94 a	24.45 b	0.00 c	0.00 b
Emphasis	15.00 b	151.41 c	10.09 b	3.98 b	68.21 a	23.82 b	3.99 c	0.00 b
Carolina Strongback	23.00 a	380.30 a	16.53 a	0.00 c	26.09 c	34.78 a	17.39 b	21.74 a

INTERNAL FRUIT QUALITY

Treatment	Cut Length (cm)	Cut Width (cm)	Total Soluble Solids (Heart)	pH (Heart)	Heart Firmness			Locule Firmness		
Non-grafted	26.88	22.75	11.53	5.96	3.32	2.27	2.62	1.82	1.53	1.69
Carnivor	25.31	21.36	11.39	5.76	4.72	4.91	4.58	2.08	2.20	2.17
Strong Tosa	24.08	20.83	10.87	5.81	4.44	4.30	4.19	1.90	1.93	2.01
Emphasis	25.72	22.00	11.32	5.91	5.61	4.52	4.94	3.49	3.85	3.90
Macis	24.42	20.15	10.57	5.57	6.02	6.08	6.66	2.77	2.28	2.63
Carolina Strongback	25.20	21.63	11.77	5.89	4.24	4.53	4.93	2.95	1.94	2.92



Carnivor and Carolina Strongback in nematode infested field



**Triploid grafted to
Carnivor Rootstock**



**Triploid grafted to
Carolina Strongback Rootstock**

Carolina Strongback:

- ❖ Plant Variety Protection (PVP)
- ❖ Licensed to Syngenta Seed Company

2021 - Breeder Assignments:

- Australia
- Chile
- Israel
- Costa Rica
- Turkey
- Mexico
- European Union
- All 1,500,000 seeds generated sold to Australia in 2021 (First year)
- 10,000,000 seeds generated for 2022 growing season

Carolina Strongback: Cucumber (*Cucumis sativus*)

- Preliminary study (2020) on use of Carolina Strongback for greenhouse cucumber production is promising.
- Yields were much heavier for grafted verses non-grafted (more than 2-fold)
- Fruit produced ten days earlier on rootstocks and produced longer.



Carolina Strongback: Issues

- Seven- to ten-day lag in fruit set compared to non-grafted

Possible causes:

- Too vigorous vine growth
- Needs more cold soil tolerance
- Flowering time

Genetic Mapping of QTL Associated with resistance to *Fusarium oxysporum* f. sp. *niveum* (*Fon*) race 2

- Initial cross of USVL246-FR2 (resistant to *Fon* race 1 & 2) x USVL114 (susceptible to both races) both *Citrullus amarus*

USVL246-FR²

Resistant to
Fon race 1 and 2

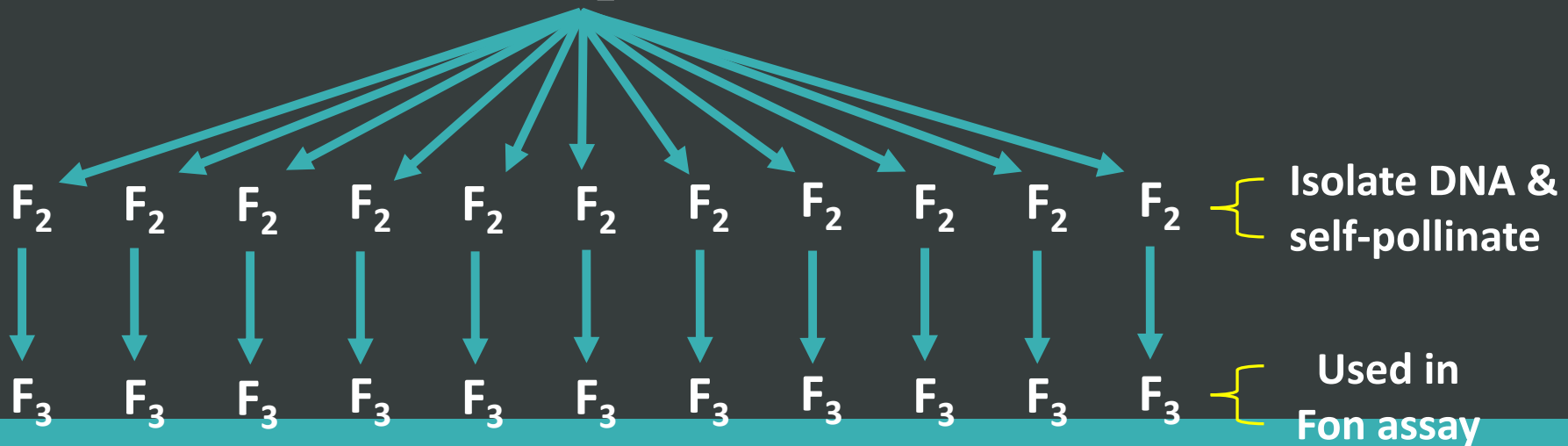
X

USVL114

Susceptible to
Fon race 1 and 2



F₁ { Self-pollination



F₇ { Recombinant Inbred Line (RIL)

Future Citroides Rootstocks

- Six (USVL246-FR2 x USVL114) RILs.
 - High tolerance to *Fon* race 1 & 2
 - High level of resistance to Root Knot nematode
- RILS performed well in grafting study.
- Made reciprocal F_1 hybrids with Carolina Strongback.
- Field testing this season in South Carolina.



Future Citroides Rootstocks

- Currently we are identifying and mapping genes associated with cold tolerance in watermelon in hopes of use in cold-tolerant rootstock
- We have identified several lines from the USVL246-FR2 x USVL114 RILS that flower and set fruit two to three weeks earlier than Carolina Strongback and USVL246-FR2 for introgression into Carolina Strongback
- We will be looking at identifying *C. amarus* lines from our collection or from the RILs with tolerance to wet conditions



Carolina Strongback: Issues

- Does not work with Cantaloupe (*Cucumis melo*)
 - Graft incompatibility?
 - Genetic?
 - Physical issues?

Carolina Strongback: Cantaloupe failure



Thank you!

- USDA Project 6080-22000-025-00
- NIFA-SCRI 6080-21000-018-08 (CucCAP)
- NIFA-SCRI 6080-22000-028-22 (Grafting)



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