

The Response of Star Ruby Grapefruit to Different *Citrus tristeza virus* Isolates

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ABSTRACT. The failure of GFMS 12 (Nartia) as a cross-protecting *Citrus tristeza virus* (CTV) isolate for Star Ruby grapefruit in South Africa necessitated the use of GFMS 35 as a substitute for the interim. Seven new CTV isolates derived from Star Ruby and Rosé grapefruit trees were evaluated and compared to GFMS 12, GFMS 35, to two isolates from mother trees at the Citrus Foundation Block that were pre-immunized with GFMS 12, two severe isolates (GFSS 1 and GFSS 5), and to trees that were planted virus-free. Trees pre-immunized with isolates GFMS 35 and GFMS 78 had the best production over a 5-yr period. These trees produced significantly better than trees that were planted virus-free, trees with mild isolates GFMS 12, GFMS 67, and those with the two severe isolates. The crop value (fruit size coupled with market prices) of trees with GFMS 35 was 5% better than that of trees with GFMS 78. Trees with GFMS 12a were third best and were 16% lower than those with GFMS 35. The latter isolate was collected from a good parent tree at the Citrus Foundation Block that was pre-immunized with GFMS 12. The results show that CTV isolate GFMS 35, which is the present pre-immunizing isolate for red grapefruit, together with isolate GFMS 78, are superior to the other isolates. However, it will be beneficial to see if the superiority will be maintained and to what extent tree life and economic production will be increased.

Shoot tip grafting is a technique to eliminate all graft-transmissible agents from citrus bud-wood sources in South Africa (5). However, the benefit of optimum growth and production of virus-free trees can not be utilized because of the abundance of *Citrus tristeza virus* (CTV) and its aphid vector, *Toxoptera citricida* (Kirkaldy) (10). Virus-free plantings become infected with various strains of CTV within a few years after planting. Many strains of CTV exist, and they usually occur as mixtures in a host (7). Factors such as virus strain, host plant and environment will determine dominance of a specific strain and this may change if any of these factors *viz.* co-infection of a new strain or extreme temperatures, are changed (4, 9). The only known method to overcome this problem is by cross-protection, a deliberate infection of virus-free material with a known CTV isolate (8). Of the commercial citrus cultivars grown in southern Africa, grapefruit is the most sensitive to stem pitting, which causes tree

decline and production of small fruit. With the initiation of the southern African Citrus Improvement Program (CIP), all grapefruit selections are pre-immunized with the GFMS 12 CTV isolate (14). This isolate originated from a 50-year-old Nartia (Marsh type) grapefruit tree in the Western Cape Province. Budwood source trees at the Citrus Foundation Block (CFB) at Uitenhage (Eastern Cape Province), which is the only source for propagating certified trees, are evaluated visually each year for decline and stem pitting symptoms. In addition, each tree of every selection is biologically indexed on an annual basis to establish the CTV severity. When there are indications of severe CTV, such a tree is terminated as a bud-wood source.

In 1993 6-yr-old Star Ruby budwood source trees were found with various degrees of stem pitting and variable fruit sizes (unpublished data). Biological indexing indicated that severe strains were dominant in four of the five budwood source trees. At first it was thought that

GFMS 12 did not protect against coinfection of severe strains. However, subsequent research showed the presence of a severe strain in the original isolate and that segregation of the strains, where the severe strain became dominant, may have occurred (13). The unsuitability of GFMS 12 as a protecting isolate for Star Ruby was also confirmed in a field trial (12). Consequently, GFMS 35 (derived from a Rosé grapefruit tree) has been approved for the pre-immunization of all red grapefruit as an interim protector (6).

The first step in searching for mild isolates for cross protection purposes is to look for old trees that are healthy and produce good quality fruit (8). The Star Ruby grapefruit industry in South Africa started in the late 1970s and therefore no trees older than 15 yr existed at the time. To overcome this problem, the best producers in the oldest plantings at Malelane, Mpumalanga Province, and Swaziland were selected. Isolates from these trees were evaluated in glasshouse tests and those with the best potential were also evaluated in the field.

This report is on the field evaluation of the best isolates identified, and the objective of the study was to find superior CTV isolates for pre-immunization that will maximize the profitability (productive life and quality) of Star Ruby grapefruit.

MATERIALS AND METHODS

Plant material and virus isolates. Virus-free Star Ruby trees on Swingle citrumelo rootstock were grown under aphid-free conditions using standard nursery practices. When the scions developed to approximately 5 mm in diameter, they were bud-inoculated with different CTV isolates. These isolates were selected from healthy-looking trees and they showed potential as cross-protecting isolates in glasshouse tests. The following treatments were applied in replicates of five:

1. GFMS 12 (derived from Nartia grapefruit A, the standard at the time);
2. GFMS 12a (derived from Star Ruby mother tree at CFB pre-immunized with GFMS 12; showing mild stem pitting);
3. GFMS 12b (derived from GFMS 12 pre-immunized mother tree at CFB; displaying severe stem pitting and small fruit);
4. GFMS 35 (derived from Rosé grapefruit at Komatipoort; Marsh grapefruit trees pre-immunized with this isolate performed better than trees with GFMS 12 over a 12-year period (11); this is the present pre-immunizing isolate for red grapefruit);
5. GFMS 65 (derived from a Star Ruby tree at Tambankulu Estates, Swaziland);
6. GFMS 67 (similar to 5);
7. GFMS 71 (derived from old budwood source Star Ruby, Esselen Nursery, Malelane);
8. GFMS 73 (similar to 7);
9. GFMS 77 (similar to 7);
10. GFMS 78 (derived from 10-yr-old planting, F. Esselen, Malelane);
11. GFSS 1 (derived from 5-yr-old Marsh grapefruit tree with severe stem pitting, Nkwaleni Valley);
12. GFSS 5 (derived from 5-yr-old Star Ruby grapefruit with severe stem pitting, F. Esselen, Malelane).
13. Control (trees planted as virus-free).

After DAS-ELISA (double antibody sandwich enzyme-linked immuno-assay) using polyclonal antiserum (1) confirmed infection, a field trial was planted at Nelspruit in a randomized block. Tree size was measured and calculated as a cylinder and half sphere according to Burger et al. (3). Fruit were harvested, graded in export sizes (2), and weighed. Tree health was monitored by evaluating stem pitting and decline.

The approximate monetary value of the fruit associated with each CTV isolate was calculated, and a projec-

tion of income was made for a hectare planting (6 × 3 spacing). The average production over three years of such a planting was determined and the value calculated according to fruit size distribution of each treatment. The value of the crop in relation to fruit size was determined by calculating the average value per export box (15 kg) for 10 yr. The highest price equalled a value of ten while the other values were calculated accordingly. The value of the crop per hectare for each treatment was determined by multiplying the production of a specific fruit size (export boxes) by the value for that size.

RESULTS AND DISCUSSION

Growth, production and disease rating of the 7-yr-old Star Ruby trees on Swingle citrumelo rootstock are given in Table 1. The canopy volumes of the trees of all the treatments were equivalent, except for trees pre-immunized with GFMS 65 and the two severe isolates, GFSS 1 and GFSS 5. Canopy sizes of these trees were significantly smaller

when compared to trees pre-immunized with GFMS 12a, GFMS 12b, GFMS 35, GFMS 73, GFMS 76, as well as those that were planted virus-free. Trees pre-immunized with GFMS 35 produced the largest crop and were significantly larger than trees pre-immunized with GFMS 12, GFMS 67, GFMS 71, GFMS 73, as well as the trees that were planted virus-free and those with the two severe isolates. The highest yield efficiency (kg/canopy volume) was achieved by trees pre-immunized with GFMS 65 (the smallest trees with a mild isolate) but was not significantly better than those of trees pre-immunized with isolates GFMS 35, GFMS 67, GFMS 77 and GFMS 78. The highest cumulative yield over four seasons was produced by trees pre-immunized by GFMS 35, but this was not significantly better than those of trees pre-immunized with GFMS 12a, GFMS 12b, GFMS 65, GFMS 76, GFMS 77, and GFMS 78. High yields can reduce fruit size and therefore the value of the crop. However, trees with GFMS 35 also had the best

TABLE 1
THE AVERAGE TREE SIZE, PRODUCTION, YIELD EFFICIENCY AND STEM PITTING RATING OF 7-YR-OLD STAR RUBY TREES PRE-IMMUNIZED WITH DIFFERENT CTV ISOLATES.

Tristeza isolate	Tree canopy size (m ³) *	2003 yield (kg) *	Yield efficiency (kg/m ³) *	Stem pitting rating**	Cumulative yield (kg) 1999-2003*	Relative crop value (ZAR)/ha/yr
Control	7.2 a	39 c	5.4 ef	0.6 abc	87 d	6658
GFMS 12	5.9 abc	46 bc	7.8 bcde	2.8 e	111 cd	8426
GFMS 12a	7.6 a	64 ab	8.4 bcd	0.2 ab	158 abc	11696
GFMS 12b	7.3 a	63 ab	8.6 bcd	2.5 e	153 abc	11534
GFMS 35	7.1 a	76 a	10.7 abc	0.0 a	174 a	13588
GFMS 65	4.0 bcd	57 abc	14.3 a	2.5 e	134 abcd	10147
GFMS 67	5.4 abc	55 bc	10.2 abcd	2.2 de	118 bcd	8862
GFMS 71	6.5 ab	53 bc	8.2 bcde	1.4 cd	123 bcd	9186
GFMS 73	7.6 a	51 bc	6.7 de	1.1 bc	123 bcd	9133
GFMS 76	7.5 a	59 abc	7.9 cde	1.2 bcd	136 abcd	10240
GFMS 77	5.5 abc	56 abc	10.2 ab	0.9 abc	148 abc	11197
GFMS 78	6.3 ab	65 ab	10.3 abcd	0.0 a	164 ab	12877
GFSS 1	3.4 cd	11 d	3.2 f	3.0 e	26 e	1533
GFSS 5	1.9 d	11 d	5.8 ef	2.6 e	22 e	1461

*Figures in each column followed by the same letter do not differ significantly at the 5% level (Fisher's LSD).

**Stem pitting rating: 1 = Smooth trunk; 2 = Occasional visible pits; 3 = Mild pitting; 4 = Moderate pitting; 5 = Severe pitting.

crop value, 5% better than that of trees with GFMS 78, which was second best. Trunks of trees with isolates GFMS 35 and GFMS 78 showed no external pitting. Except for the trees with the two severe isolates, severe pitting occurred in trees pre-immunized with GFMS 12 (Nartia), GFMS 12b, GFMS 65 and GFMS 67. No decline occurred in any treatment but one tree with GFSS 5 died at an early stage.

Trees pre-immunized with GFMS 35, the present cross protecting isolate for Star Ruby in the CIP, gave the best results followed by isolates GFMS 78 and GFMS 12a. The latter was derived from the original mother tree for Star Ruby budwood at the CFB. Trees with GFMS 35 produced significantly better than trees with mild isolates GFMS 12 or GFMS 67, those that were planted

virus-free, and the two severe isolates. The total crop value (fruit size and market prices) of trees with GFMS 35 and GFMS 78 averaged 23% better than the rest.

GFMS 35 is recommended for use as a pre-immunizing isolate for Star Ruby grapefruit in the southern Africa citrus industry. This isolate contains no severe strains (6) and the chances of detrimental strain shifts within Star Ruby trees, which are planted in different climatic areas, are minimized.

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