

# Accessions of *Citrullus lanatus* var. *citroides* Are Valuable Rootstocks for Grafted Watermelon in Fields Infested with Root-knot Nematodes

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**Abstract.** Root-knot nematode-resistant rootstock lines (designated RKVL for Root-Knot Vegetable Laboratory) derived from wild watermelon (*Citrullus lanatus* var. *citroides*) were compared with wild tinda (*Praecitrullus fistulosus*) lines and commercial cucurbit rootstock cultivars for grafting of seedless triploid watermelon ‘Tri-X 313’ (*C. lanatus* var. *lanatus*) in a field infested with the southern root-knot nematode (RKN) (*Meloidogyne incognita*) in Charleston, SC, during 2009 and 2010. In both years, RKN infection was severe in ‘Emphasis’ bottle gourd, ‘Strong Tosa’ hybrid squash, and wild tinda rootstocks with galling of the root system ranging from 86% to 100%. In 2009, the RKVL wild watermelon rootstocks exhibited lower ( $P < 0.05$ ) percentages of root galling (range 9% to 16%) than non-grafted ‘Tri-X 313’ (41%), ‘Emphasis’, ‘Strong Tosa’, and the wild tinda rootstocks. The grafted wild watermelon rootstock RKVL 318 produced more ( $P \leq 0.05$ ) fruit (12 per plot) than all other entries (mean = five per plot), and it produced a heavier ( $P \leq 0.05$ ) fruit yield (29.5 kg per plot) than all entries except self-grafted ‘Tri-X 313’ (21.5 kg per plot). In 2010, soil in half of the plots was treated with methyl bromide (50%):chloropicrin (50%) (392 kg·ha<sup>-1</sup>) before planting. The RKVL wild watermelon rootstocks exhibited resistance to RKN with percentages of root system galled ranging from 11% for RKVL 316 to 56% for RKVL 301 in the untreated control plots. Fruit yields in the untreated plots were 21.9, 25.6, and 19.9 kg/plot for RKVL 301, RKVL 316, and RKVL 318, respectively. Yields were greater ( $P \leq 0.05$ ) for the three RKVL rootstocks than for ‘Strong Tosa’ (3.0 kg) and ‘Ojakkyo’ wild watermelon rootstock (2.8 kg) in the untreated plots. Yields of watermelon grafted on ‘Strong Tosa’ were nearly seven times greater ( $P \leq 0.05$ ) in the methyl bromide-treated plots than in the untreated plots. In contrast, yields of RKVL 316 and RKVL 318 were similar in both treatments and the yield of RKVL 301 was less ( $P \leq 0.05$ ) in the methyl bromide-treated plots than in the untreated plots. The three RKVL wild watermelon rootstock lines exhibited resistance to RKN. RKVL 316 had low root galling and produced the heaviest fruit yield and greatest numbers of fruit of any rootstock evaluated in 2010. The RKVL lines should be useful sources of RKN resistance for rootstocks for grafted watermelon.

The southern RKN (*Meloidogyne incognita*) is a serious constraint to U.S. watermelon production and can significantly reduce watermelon yields in the southern

United States (Davis, 2007; Sumner and Johnson, 1973; Thies, 1996). Pre-plant fumigation of soil beds with methyl bromide has been the primary method for controlling root-knot nematodes and soilborne diseases in watermelon (Thies et al., 2010). However, in accordance with the Montreal Protocol and the U.S. Clean Air Act, methyl bromide was phased out effective 1 Jan. 2005 and use of methyl bromide for pre-plant fumigation of soil has been restricted to that allowed through Critical Use Exemptions (CUE) granted to growers in specific states or geographic regions by the U.S. Environmental

Protection Agency (USEPA) (USEPA, 2011). The loss of methyl bromide for pre-plant soil fumigation was estimated to result in annual yield losses of 15% to 20% for watermelon in Georgia and Florida (Lynch and Carpenter, 1999) and the total economic impact of the methyl bromide ban for U.S. watermelon production and consumption was calculated to be –\$9,111,000 annually (Carpenter et al., 2000). Additionally, nematicides used for controlling RKN in watermelon and other vegetable crops have been lost from the U.S. market because of human health risks and groundwater contamination (Chitwood, 2003). Thus, there is increased interest in the development of alternative technologies such as use of resistant rootstocks for grafting to manage RKN in watermelon and other vegetable crops.

Grafting vegetables onto resistant rootstocks to prevent damage by soilborne diseases or pests has been practiced for many years in eastern Asia (Cohen et al., 2007). Grafting has proven to be a quick alternative to long-term breeding programs aimed at incorporating resistance to soilborne disease, especially fusarium wilt, into elite vegetable cultivars (Miguel et al., 2004). Rootstocks also proved to be useful in enhancing tolerance to abiotic stresses and in improving fruit yield and quality (Cohen et al., 2007; Core, 2005; Edelstein and Ben-Hur, 2006).

Grafting of watermelon is an emerging production practice in U.S. agriculture, which is being adopted largely for control of soilborne diseases such as fusarium wilt and vine decline of melon caused by *Monosporascus cannonballus*. The extensive use of methyl bromide for pre-plant soil fumigation (before 2005) and the availability of large acreages of land, which accommodated multiyear crop rotations, curtailed the buildup of soilborne disease pathogens in watermelon production fields in the United States in past decades. However, availability of agricultural land is becoming more limited in the United States and methyl bromide for pre-plant soil fumigation is only available through limited CUE allocated to growers by the USEPA (USEPA, 2011). Thus, grafting is becoming a feasible alternative for disease control in U.S. watermelon production. Although the cost of grafted seedless watermelon transplants has been estimated at \$0.75 per plant compared with \$0.28 per non-grafted plant in the United States (Taylor et al., 2008), the use of newly improved grafting equipment can reduce the labor requirements and costs of grafting. Current commercial grafting methods require maintenance of at least one rootstock cotyledon during the graft healing period to ensure high survival of grafted plants (Lee, 1994; Lee and Oda, 2003). Regrowth of the rootstock from meristematic tissue adjacent to one of the cotyledons has been a major drawback to cost-effective use of grafted plants in U.S. agriculture (Edelstein, 2004). However, Hassell et al. (2008) recently developed the cotyledon devoid method, which involves the removal of meristematic tissue

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that prevents rootstock shoot regeneration. If further testing proves this new grafting method to be successful, grafting costs could be significantly reduced as a result of complete elimination of rootstock regrowth (Memmott and Hassell, 2010).

Squash hybrid (*Cucurbita maxima* × *C. moschata*) and bottle gourd (*Lagenaria siceraria*) rootstocks are among the most commonly used rootstocks for grafting watermelon and other cucurbits because these species are not susceptible to fusarium wilt caused by *Fusarium oxysporum* f. sp. *niveum* and they have vigorous roots systems (Edelstein and Ben-Hur, 2006; Miguel et al., 2004). However, the squash hybrid rootstocks and bottle gourd rootstocks are extremely susceptible to RKNs (Thies et al., 2010), and thus these rootstocks are not suitable for grafting when fields or high tunnels are infested with RKNs. RKNs have been identified as the primary pathogen of squash hybrid and bottle gourd rootstocks worldwide (Xingping Xie, personal communication).

Germplasm with resistance to RKN should be useful for development of resistant rootstocks to manage RKN in grafted watermelon. The *C. lanatus* var. *citroides* PI collection at the USDA, ARS Plant Genetic Resources and Conservation Unit in Griffin, GA, represents wide genetic diversity and is considered a valuable germplasm source for genes conferring resistance to diseases or pests of watermelon (Jarret et al., 1997; Levi et al., 2001, 2010). In previous studies we have identified U.S. PIs of *C. lanatus* var. *citroides* that contain resistance to RKN (Thies and Levi, 2003, 2007). At the U.S. Vegetable Laboratory, USDA, ARS, we have selected and developed several improved lines derived from these wild watermelon PIs that could be useful as rootstocks for grafted watermelon (Thies et al., 2008, 2010). The objective of these studies was to evaluate the performance of these improved *C. lanatus* var. *citroides* rootstock lines vs. commercial rootstocks of bottle gourd and hybrid squash for managing RKN with and without methyl bromide treatments.

## Materials and Methods

### Field study, Charleston, SC, 2009

Eleven rootstock/seedless watermelon scion ('Tri-X 313') combinations, a non-grafted control ('Tri-X 313'), and a self-grafted control ('Tri-X 313') were evaluated in a field infested with the southern RKN, *Meloidogyne incognita*, at the U.S. Vegetable Laboratory, Charleston, SC. The soil was a Yonges fine loamy sand. Rootstocks were grafted to the scion triploid watermelon 'Tri-X 313' and grafted plants were transplanted on raised white plastic mulch beds at a plant density of 2.79 m<sup>2</sup>/plant on 4 June 2009. The pollinizer 'SP-4' was interplanted between every third and fourth grafted seedless watermelon.

**Rootstock genotypes.** Seedless watermelon 'Tri-X 313' was grafted onto five wild watermelon (*Citrullus lanatus* var. *citroides*) germplasm lines developed at the U.S. Vegetable

Laboratory (RKVL 301, RKVL 302, RKVL 303, RKVL 316, and RKVL 318) These five wild RKN-resistant watermelon germplasm lines were designated RKVL. 'Tri-X 313' scions also were grafted onto a bottle gourd (*Lagenaria siceraria* 'Emphasis') rootstock cultivar, a squash hybrid rootstock cultivar (*Cucurbita maxima* × *C. moschata* 'Strong Tosa'), a commercial watermelon rootstock cultivar (*C. lanatus* var. *citroides* 'Ojakkyo'), and three wild tinda (*Praecitrullus fistulosus*) rootstocks (P0004, P0005, and P0006). 'Tri-X 313' self-grafted and 'Tri-X 313' non-grafted also were included as checks in the study. The experimental design was a randomized complete block with six replicates of six plants per replicate.

**Data collected.** Watermelon fruit were harvested and fruit weight, fruit size (length × diameter), and fruit quality traits including total soluble solids (Brix) were recorded on 30 July, 3 Aug., 6 Aug., 11 Aug., 13 Aug., and 17 Aug. 2009. At the end of the harvest season on 26 Aug., roots of all plants were dug and evaluated for percent of root system galled and covered with *M. incognita* egg masses using a 0% to 100% scale where 1 = 0% to 3%, 2 = 4% to 25%, 3 = 26% to 50%, 4 = 51% to 80%, and 5 = 81% to 100% roots galled or covered with egg masses. Root systems were also rated from 0% to 100% for amount of fibrous roots, where 100% = highly fibrous root system and 0% = no fibrous roots. Nematode eggs were extracted from the roots using 1% NaOCl (Hussey and Barker, 1973) and eggs were counted using a stereomicroscope. Root galling, egg mass, and fibrous root percentages were arcsine-transformed and eggs per gram fresh root were log<sub>10</sub> (x + 1) transformed to normalize data. Analysis of variance was conducted on transformed data using the GLM procedure of SAS Version 9.1 for Windows (SAS Institute Inc., Cary, NC) and means were separated using Fisher's protected least significant difference.

### Field study, Charleston, SC, 2010

Eight rootstock/seedless watermelon scion combinations and self-grafted and non-grafted 'Tri-X 313' controls were evaluated in a *M. incognita*-infested field near that used for the 2009 study at the U.S. Vegetable Laboratory, Charleston, SC. Rootstocks were grafted to the scion triploid watermelon 'Tri-X 313' using the single cotyledon method (Hassell et al., 2008) and planted on raised white plastic mulched beds at a plant density of 2.23 m<sup>2</sup>/plant on 28 June 2010. Half of the plots were fumigated with methyl bromide-chloropicrin (50/50) at 392 kg-ha<sup>-1</sup> when beds were prepared.

**Rootstock genotypes.** Three of the best performing wild watermelon lines (*C. lanatus* var. *citroides*) (RKVL 301, 316, and 318) were selected for testing based on data from the 2009 Charleston, SC, study and previous tests (Thies et al., 2010). Like in 2009, one bottle gourd (*L. siceraria* 'Emphasis') cultivar, one squash (*Cucurbita maxima* ×

*C. moschata* 'Strong Tosa') hybrid, and one commercial watermelon rootstock (*C. lanatus* var. *citroides* 'Ojakkyo') were included in the study. In addition, two wild tinda (*P. fistulosus*) rootstocks (P0004 and P0005) and two 'Tri-X 313' controls (self-grafted and non-grafted) also were included in the study.

**Experimental design.** The experimental design was a split-plot design where whole plots were methyl bromide treatments [non-treated control and methyl bromide (50%):chloropicrin (50%) at 392 kg-ha<sup>-1</sup>] and sub-plots were the eight rootstock/scion combinations and the two checks. The rootstock genotypes were arranged in a randomized complete block within the methyl bromide whole plot treatments. The experiment had six replicates of six plants per replicate. The pollinizer 'SP-4' was interplanted between every third and fourth grafted seedless watermelon. Watermelon fruit were harvested and yield data were collected. After the final harvest, roots of all plants were dug, washed, and evaluated for the percentage of root system galled and covered with egg masses of *M. incognita*. Root systems were also rated for percentage of fibrous roots as described for the 2009 field study.

## Results and Discussion

**Field study, 2009.** Root-knot nematode infection was severe in 'Emphasis' bottle gourd (*L. siceraria*) (86% root system galled) and 'Strong Tosa' squash hybrid (*Cucurbita maxima* × *C. moschata*) (100% galled) rootstocks (Table 1). The three tinda (*P. fistulosus*) rootstocks were also severely infected by RKN with percentages of root system galled ranging from 91% to 100%. The five wild watermelon rootstocks exhibited lower ( $P \leq 0.05$ ) percentages of root galling (range 9% to 16%) than non-grafted 'Tri-X 313' (41%), 'Emphasis', 'Strong Tosa', and the *P. fistulosus* accessions. Although 'Ojakkyo' exhibited heavier root galling (26%) than the five wild watermelon rootstocks, the differences were not significant.

Egg mass production was high for 'Emphasis' bottle gourd (40% of root system covered with egg masses) and for 'Strong Tosa' squash hybrid (67%). Two of the *P. fistulosus* rootstocks (P0004 and P0006) also had a significant portion of their root systems covered with egg masses (41% and 83%, respectively). The *P. fistulosus* rootstock P0005 had a lower ( $P \leq 0.05$ ) percentage of the root system covered with egg masses than the P0004 and P0006 rootstocks, which can be explained by very severe root galling of P0005 that resulted in loss of the egg masses from the root system. The five wild watermelon rootstocks exhibited lower ( $P \leq 0.05$ ) percentages of root systems covered with egg masses (range 2% to 3%) than 'Emphasis', 'Strong Tosa', and P0004 and P0006. 'Ojakkyo' had slightly higher percentages of the root system covered by egg masses (8%) compared with the five wild

Table 1. Percentages of root systems galled and covered with *Meloidogyne incognita* egg masses, numbers of *M. incognita* eggs per gram fresh root, percentages of root systems with fibrous roots, and watermelon fruit yields and numbers for 'Tri-X 313' seedless watermelon grafted on rootstocks of wild watermelon lines and wild tinda lines and selected commercial rootstocks, Charleston, SC, 2009.

Rootstock accession	Percentage root system galled <sup>a</sup>	Percentage root system covered with egg masses <sup>a</sup>	Eggs per gram fresh root <sup>b</sup>	Percentage root system with fibrous roots <sup>c,x</sup>	Total fruit wt (kg/plot) <sup>w</sup>	No. of fruit/plot <sup>w</sup>
Wild watermelon ( <i>Citrullus lanatus</i> var. <i>citroides</i> )						
RKVL 301	9 f <sup>a</sup>	3 d	24 fg	92 ab	20.0 b	7 bc
RKVL 302	10 f	2 d	21 g	71 cd	14.8 bc	5 bc
RKVL 303	16 ef	3 d	58 efg	98 b	16.6 bc	6 bc
RKVL 316	14 ef	3 d	31 fg	83 ab	18.8 bc	8 b
RKVL 318	13 ef	2 d	16 g	87 ab	29.5 a	12 a
'Ojakkyo'	26 def	8 cd	34 efg	74 cd	19.8 b	7 bc
Bottle gourd ( <i>Lagenaria siceraria</i> )						
'Emphasis'	86 c	40 bc	835 ab	33 e	17.2 bc	7 bc
<i>(Cucurbita maxima × C. moschata)</i>						
'Strong Tosa'	99 b	67 ab	3137 a	35 f	11.6 cde	3 cde
Tinda ( <i>Praecitrullus fistulosus</i> )						
P0004	98 bc	41 b	284 bcd	2 f	6.2 e	2 e
P0005	91 bc	17 cd	128 cde	6 f	4.7 e	2 e
P0006	100 a	83 a	448 bc	1 f	7.0 de	3 de
Watermelon checks ( <i>C. lanatus</i> var. <i>lanatus</i> )						
'Tri-X 31' non-grafted	41 d	4 d	140 cde	54 de	15.7 bc	6 bc
'Tri-X 313' self-grafted	31 de	13 cd	91 def	51 de	21.5 ab	8 e

<sup>a</sup>Data were arcsine transformed before analysis; non-transformed data are shown.

<sup>b</sup>Data were  $\log_{10}(x + 1)$  transformed before analysis; non-transformed data are shown.

<sup>c</sup>Root systems were rated on a 0% to 100% scale, where 100% = 100% of root system had fibrous roots and 0% = root system had no fibrous roots.

<sup>w</sup>Six plants per plot ( $3.6 \times 4.6 = \approx 2.8 \text{ m}^2$  per plant).

<sup>x</sup>Means within a column followed by the same letter are not significantly different ( $P \leq 0.05$ ) according to Fisher's protected least significant difference.

watermelon rootstocks, but the differences were not significant.

RKN reproduction was lowest for the five wild watermelon rootstocks and 'Ojakkyo'. Two of the wild watermelon rootstocks, RKVL 302 and RKVL 318, had fewer ( $P \leq 0.05$ ) eggs of *M. incognita* per gram fresh root than 'Tri-X 313' (non-grafted and self-grafted), 'Emphasis', 'Strong Tosa', P0004, P0005, and P0006. 'Emphasis' and 'Strong Tosa' supported the greatest numbers of *M. incognita* eggs per gram fresh root (835 and 3137, respectively).

The wild watermelon rootstocks RKVL 301, RKVL 303, RKVL 316, and RKVL 318 had greater ( $P \leq 0.05$ ) amounts of fibrous roots than 'Tri-X 313' (non-grafted and self-grafted), 'Emphasis', 'Strong Tosa', P0004, P0005, and P0006 (Table 1). Large amounts of fibrous roots are often associated with the ability of a host plant to tolerate attack by plant-parasitic nematodes. We have often observed that many of the *Citrullus lanatus* var. *citroides* germplasm accessions have excellent fibrous root systems when grown in fields with high soil populations of RKN.

The wild watermelon rootstock RKVL 318 produced significantly more fruit (12 per plot) ( $P \leq 0.05$ ) than any other entry and also produced the heaviest yield (29.5 kg per plot) ( $P \leq 0.05$ ) compared with all other entries except self-grafted 'Tri-X 313' (21.5 kg per plot). Although RKVL 301, RKVL 303, and RKVL 316 were resistant to southern RKN with low root galling and low nematode reproduction, their fruit numbers and yields were not significantly greater than non-grafted 'Tri-X 313'. 'Strong Tosa' and

the three *P. fistulosus* rootstocks produced the lowest yields in the study, which is associated with the extensive root galling, root damage, and poor fibrous root systems (range less than 1% to 35% fibrous roots) observed for these rootstocks. No significant differences were detected among rootstocks for total soluble solids and fruit size (length  $\times$  diameter) (data not shown).

All five of the wild watermelon lines exhibited resistance to southern RKN in this test. Furthermore, the wild watermelon rootstock RKVL 318 produced the most fruit and highest yield of any of the rootstock entries in the test. The results of this study suggest that these wild watermelon rootstocks (*C. lanatus* var. *citroides*) possess useful resistance to RKN in the field.

These results are consistent with our earlier observations of resistance to RKN in seedling evaluations of *Citrullus* accessions in greenhouse experiments (Thies and Levi, 2003, 2007).

*Field study, 2010: Untreated control plots.* The three RKVL wild watermelon rootstocks exhibited resistance to RKN with percentages of root system galled ranging from 11% for RKVL 316 (Fig. 1A) to 56% for RKVL 301 in the untreated control plots (Table 2). 'Ojakkyo' had 52% of the root system galled. In the 2010 field test, the RKN pressure was extremely high as illustrated by the extreme root galling on 'Emphasis' bottle gourd (Fig. 1B) and non-grafted 'Tri-X 313' seedless watermelon, which had 97% and 96% of the root systems galled, respectively,

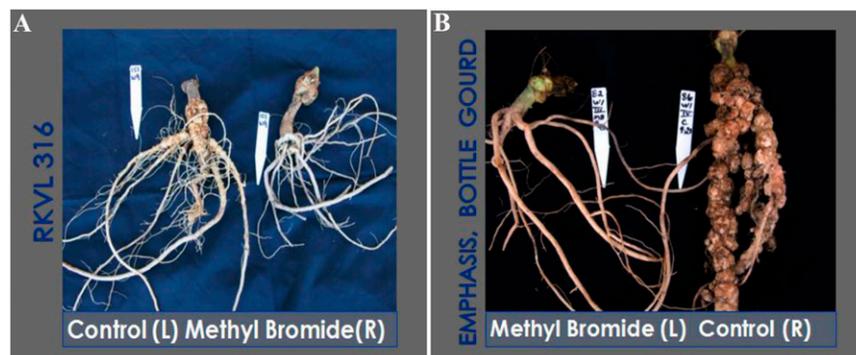


Fig. 1. (A) Root-knot nematode (RKN)-resistant Root-Knot Vegetable Laboratory (RKVL) 316 wild watermelon breeding line: root system from untreated control plot (left) with very minimal RKN galling and root system with no galling (right) from methyl bromide-treated plot. (B) RKN-susceptible 'Emphasis' bottle gourd: root system with no RKN galling from methyl bromide-treated plot (left) and root system with severe RKN galling from untreated control plot (right). Sept. 2010, Charleston, SC.

Table 2. Percentages of root systems galled by *Meloidogyne incognita*, numbers of *M. incognita* eggs per gram fresh root, percentages of root systems with fibrous roots, and watermelon fruit yields and numbers for ‘Tri-X 313’ seedless watermelon grafted on rootstocks of wild watermelon lines and wild tinda lines and selected commercial rootstocks grown in field plots treated with methyl bromide (50%):chloropicrin (50%) at 392 kg·ha<sup>-1</sup> before planting or left untreated, Charleston, SC, 2010.

Rootstock accession	Methyl bromide treatment	Percentage root system galled <sup>z</sup>	Eggs per gram fresh root <sup>y</sup>	Percentage root system with fibrous roots <sup>z,x</sup>	Total fruit wt (kg/plot) <sup>w</sup>	No. of fruit per plot <sup>w</sup>
<i>Wild watermelon (Citrullus lanatus var. citroides)</i>						
RKVL 301	–	56 bc <sup>v</sup>	39 d–g	40 cd	21.9 ab	4.2 a
	+	0 e	16 f–i	46 bc	6.4 d–g	1.6 b–d
RKVL 316	–	11 d	179 b–e	30 cd	25.6 a	4.5 a
	+	0 e	23 g–i	86 ab	15.2 a–g	2.6 a–d
RKVL 318	–	40 c	148 bc	55 a–c	19.9 a–e	3.5 ab
	+	0 e	22 g–i	54 a–c	6.0 e–g	3.5 ab
‘Ojakkyo’	–	52 bc	474 b	37 c	2.8 g	0.5 d
	+	2 e	2 hi	42 bc	8.9 b–g	1.7 b–d
<i>Bottle gourd (Lagenaria siceraria)</i>						
‘Emphasis’	–	97 a	3950 a	66 ab	7.2 c–g	1.4 b–d
	+	0 e	6 g–i	2 f	15.4 a–g	3.0 a–c
<i>Hybrid squash (Cucurbita maxima × C. moschata)</i>						
‘Strong Tosa’	–	96 a	2462 b	5 ef	3.0 g	0.6 cd
	+	7 e	0 i	99 c–e	20.0 a–c	3.7 a
<i>Tinda (Praecitrullus fistulosus)</i>						
P0004	–	97 a	440 abc	3 f	2.8 g	0.5 f
	+	0 e	18 hi	—	16.7 a–f	2.5 a–f
P0005	–	91 b	239 a–d	3 f	13.4 a–g	1.4 c–f
	+	0 e	12 e–h	—	15.8 a–g	3.5 a–d
<i>Watermelon checks (C. lanatus var. lanatus)</i>						
‘Tri-X 313’ non-grafted	–	97 a	119 b–d	1 ef	15.3 a–d	2.8 a–d
	+	0 e	120 e–h	30 cd	21.0 a	4.2 a
‘Tri-X 313’ self-grafted	–	76 bcd	188 bcd	1 ef	20.0 a–d	3.9 a
	+	0 e	68 fgh	30 cd	26.1 a	4.5 a

<sup>z</sup>Data were arcsine transformed before analysis; non-transformed data are shown.

<sup>y</sup>Data were log<sub>10</sub>(x + 1) transformed before analysis; non-transformed data are shown.

<sup>x</sup>Root systems were rated on a 0% to 100% scale, where 100% = 100% of root system had fibrous roots and 0% = root system had no fibrous roots.

<sup>w</sup>Six plants per plot.

<sup>v</sup>Means within a column followed by the same letter are not significantly different ( $P \leq 0.05$ ) according to Fisher’s protected least significant difference.

compared with 86% and 41% of root systems galled for ‘Emphasis’ and non-grafted ‘Tri-X 313’, respectively, in 2009. The root systems of ‘Strong Tosa’ hybrid squash, *P. fistulosus* P0004 and P0005, and self-grafted ‘Tri-X 313’ watermelon rootstocks also were severely galled (96%, 97%, 91%, and 76% of root systems galled, respectively). The three RKVL wild watermelon rootstocks and ‘Tri-X 313’ self-grafted produced the heaviest fruit yields of all entries in the test in the untreated control plots (Table 2). Fruit yields in the untreated plots were 21.9, 25.6, and 19.9 kg/plot for RKVL 301, RKVL 316, and RKVL 318, respectively (Table 2). Yields in the untreated plots were greater ( $P < 0.05$ ) for plants grafted on the RKVL rootstocks, P0005 tinda (13.4 kg), or the non-grafted or self-grafted ‘Tri-X 313’ than for those grafted on ‘Strong Tosa’ (3.0 kg), ‘Emphasis’ (7.2 kg), ‘Ojakkyo’ (2.8 kg), and P0004 tinda (2.8 kg), but significant differences in yield were not detected between plants grafted on the RKVL rootstocks and non-grafted or self-grafted ‘Tri-X 313’ (15.3 kg and 20.0 kg, respectively).

**Methyl bromide-treated plots.** Methyl bromide was highly effective in controlling RKN in all rootstock entries with essentially no galling observed for any entry (Table 2). Likewise, numbers of *M. incognita* eggs

per gram fresh root were low for all of the rootstock accessions and checks (ranging from 0 eggs for ‘Strong Tosa’ to 120 eggs for self-grafted ‘Tri-X 313’) in methyl bromide-treated plots.

Fruit yields of seedless watermelon grafted on ‘Strong Tosa’ were nearly seven times greater ( $P < 0.05$ ) in the methyl bromide-treated plots than in the untreated plots. Although significant differences for fruit yields of seedless watermelon grafted on ‘Emphasis’ rootstocks were not detected between methyl bromide-treated and untreated plots, fruit yields were more than two times greater for plants grown in the methyl bromide plots than in untreated plots. Similarly, yields were numerically, although not significantly, greater for ‘Tri-X 313’ watermelon (non-grafted) and ‘Tri-X 313’ grafted on ‘Ojakkyo’ in the methyl bromide-treated plots than in untreated plots. In contrast, yields of seedless watermelon grafted on RKVL 301 were 71% greater ( $P \leq 0.05$ ) in the untreated plots than in methyl bromide-treated plots. Similar trends (although not statistically significant) in fruit production were observed for RKVL 316 and RKVL 318, which produced 41% and 70% heavier fruit yields, respectively, in the untreated plots than the methyl bromide-treated plots. The greater yields from RKVL plants

in the untreated plots may have been in response to RKN infection because RKVL 301 plants as well as those of RKVL 316 and RKVL 318 usually have larger, more fibrous root systems when exposed to RKN than when grown in absence of RKN (J.A. Thies, unpublished data).

The three RKVL wild watermelon rootstock lines RKVL 301, RKVL 316, and RKVL 318 (developed at the U.S. Vegetable Laboratory, USDA, ARS, Charleston, SC) exhibited resistance to southern RKN in the 2010 test. RKVL 316 had low root galling in both the untreated and methyl bromide-treated plots. RKVL 301 and RKVL 316 produced heavier fruit yields in untreated plots than ‘Ojakkyo’ wild watermelon, ‘Emphasis’ bottle gourd, ‘Strong Tosa’ hybrid squash, and P0004 tinda rootstocks. The bottle gourd and hybrid squash rootstocks were highly susceptible to RKN with severe root galling and very low yields, demonstrating that bottle gourd and hybrid squash are unsuitable for use in RKN-infested fields without methyl bromide or other nematicide treatment. Our results confirm those of our previous greenhouse and field studies, indicating that the RKVL wild watermelon rootstock lines (*C. lanatus* var. *citroides*) possess useful resistance to RKN in the field. These rootstock lines should be useful

sources of RKN resistance for rootstock breeding programs and also useful in the development of RKN-resistant watermelon cultivars.

Grafting allows a rapid response to the development of new races of a pathogen if resistant rootstocks are available and provides an alternative to breeding new resistant watermelon cultivars for controlling soil-borne diseases. In Japan and other parts of Asia, watermelons have been grafted on cucurbit rootstocks to suppress fusarium wilt in watermelon caused by *F. oxysporum* Schlechtend.: Fr. f. sp. *niveum* (E.F. Sm.; W.C. Snyder and H.N. Hans) that causes disease in watermelon but not in cucurbit rootstocks (Cohen et al., 2007; Miguel et al., 2004). However, the cucurbit rootstocks presently used for grafting watermelon are highly susceptible to RKN. The *C. lanatus* var. *citroides* rootstocks may offer a solution for suppressing RKN and be useful to rotate with the susceptible cucurbit rootstocks if watermelon is grown in successive years in the same fields.

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