ISSN 0100-2945 http://dx.doi.org/10.1590/0100-29452016073

CITRUS TRISTEZA DISEASE SEVERITY AND AGRONOMIC PERFORMANCE OF 'PÊRA' SWEET ORANGE CULTIVARS AND SELECTIONS IN THE NORTH AND NORTHWEST REGIONS OF THE STATE OF PARANÁ, BRAZIL[1]

JOSÉ SEGUNDO GIAMPANI[2], ZULEIDE HISSANO TAZIMA[3], VIVIANE YUMI BABA[4], INÊS FUMIKO UBUKATA YADA[5], RUI PEREIRA LEITE JÚNIOR[6]

ABSTRACT - 'Pêra' sweet orange is the most important citrus cultivar in Brazil, but it is highly susceptible to *Citrus tristeza virus* (CTV), the causal agent of citrus tristeza disease. In this study, we evaluated the severity of citrus tristeza disease and its relation with the agronomic performance of seven cultivars and six selections of 'Pêra' sweet orange under field conditions in the North and Northwest regions of the State of Paraná, Brazil. The citrus tristeza disease severity was determined based on symptoms of stem pitting. The vegetative growth assessed in trees of all four locations for the 2012 season, and in two field plots for the 2011, 2012 and 2013 harvests, respectively. The cultivar 'IPR 153' (Bianchi) had the best performance concerning stem pitting severity, while Selection 27 was among the ones with the highest rate of vegetative growth. Further, Selections 27 and 12, and the cultivar 'Vimusa' had the largest fruit yields. Based on correlation analysis, stem pitting symptoms severity was negatively correlated with vegetative growth and fruit yield. Furthermore, environmental conditions may have played an important role in the severity of citrus tristeza disease and consequently on the agronomic performance of the cultivars and selections of 'Pêra' sweet orange in the North and Northwest regions of the State of Paraná.
Index Terms: *Citrus sinensis*, stem pitting, yield, vegetative growth, *Citrus tristeza virus.*

SEVERIDADE DE TRISTEZA DOS CITROS E COMPORTAMENTO AGRONÔMICO DE CULTIVARES E SELEÇÕES DE LARANJEIRA PÊRA NAS REGIÕES NORTE E NOROESTE DO ESTADO DO PARANÁ

RESUMO-A laranjeira Pêra, principal cultivar de citros no Brasil, apresenta alta suscetibilidade ao *Citrus tristeza virus* (CTV), agente causal da tristeza dos citros. No presente estudo foi avaliada a severidade de tristeza dos citros e a sua relação com o desempenho agronômico de sete cultivares e seis seleções de laranjeira Pêra, em experimentos implantados em áreas localizadas nas regiões Norte e Noroeste do estado do Paraná. A avaliação da tristeza dos citros foi realizada com base na severidade dos sintomas de caneluras. O desenvolvimento vegetativo das plantas cítricas foi avaliado nas quatro localidades em 2012, enquanto que a produção foi avaliada em experimentos em duas localidades, para as safras de 2011, 2012 e 2013. A cultivar IPR 153 (Bianchi) apresentou o melhor comportamento em relação à severidade de caneluras nas quatro localidades. A Seleção 27 está entre aqueles com os melhores índices de vigor vegetativo, e também apresentou as maiores produções, acompanhada da cultivar Vimusa e Seleção 12. Além disso, a severidade dos sintomas de caneluras apresentou correlação significativamente negativa com o desenvolvimento vegetativo e a produtividade das plantas. Cabe salientar que as condições ambientais provavelmente tiveram implicações significativas na expressão dos sintomas de tristeza e consequentemente no desempenho agronômico das cultivares e seleções de laranjeira Pêra nas regiões Norte e Noroeste do estado do Paraná.
Termos para Indexação: *Citrus sinensis*, caneluras, produção, desenvolvimento vegetativo, *Citrus tristeza virus*.

[1](Trabalho 042-15). Recebido em: 16-01-2015. Aceito para publicação em: 22-02-2016.
[2]Área de Proteção de Plantas, Instituto Agronômico do Paraná (IAPAR), Londrina-PR, Brazil. E-mail: jsgiampa@gmail.com
[3]Área de Fitotecnia, Instituto Agronômico do Paraná (IAPAR), Londrina-PR, Brazil, E-mail: zuleide@iapar.br
[4]Universidade Estadual de Londrina-(UEL), Londrina-PR, Brazil. E-mail: vybaba15@gmail.com
[5]Área de Biometria, Instituto Agronômico do Paraná(IAPAR), Londrina-PR, Brazil. E-mail: inesyada@iapar.br
[6]Área de Proteção de Plantas, Instituto Agronômico do Paraná (IAPAR), Londrina-PR, Brazil. E-mail: ruileite@iapar.br

INTRODUCTION

'Pêra' sweet orange [*Citrus sinensis* (L.) Osbeck] is the most important citrus cultivar in Brazil. Fruits of this sweet orange have an excellent quality for internal and external fresh fruit markets, as well as for industrial processing (SALIBE et al., 2002; PIO et al., 2005), thus 'Pêra' is the main choice for citrus growers, fresh fruit consumers and industry (DONADIO, 1999). This cultivar represents approximately 45% of the sweet orange trees planted in the State of Paraná, Brazil, where the citrus industry has been growing considerably in the last few years (AULER et al., 2014).

A main restriction for planting 'Pêra' sweet orange in Brazil is the susceptibility to *Citrus tristeza virus* (CTV), the causal agent of citrus tristeza disease (MÜLLER et al., 1999, SALIBE et al., 2002). Citrus tristeza is one of the most important diseases for citrus production in several regions around the world, because of the endemic nature of the virus and their vectors, as well as the susceptibility of certain agronomic important citrus rootstock and scion cultivars (ROSSETTI, 2001). Under the Brazilian conditions, all sweet orange trees are infected with CTV, as the brown citrus aphid (*Toxoptera citricida* Kirk.), the main vector of the virus, is widespread in all citrus growing areas (BORDIGNON et al., 2003b).

Symptoms of citrus tristeza can vary according to the aggressiveness of the viral complex, its multiplication in plant tissue and environmental conditions, particularly temperature (BENNETT; COSTA, 1949; BORDIGNON et al., 2003a). The citrus scion and rootstock cultivars and their interactions can also determine the severity of the disease symptoms exhibited by the citrus tree (BENNETT; COSTA, 1949; BORDIGNON et al., 2003a). In Brazil, the damage caused by the disease is usually associated with stem pitting, which results from the development of a mass in the phloem parenchyma cells that penetrates into the xylem, causing vessel malformation and even destruction of the xylem vessels and parenchyma (TANAKA et al., 2009). In more severe cases, the citrus tree may show atrophy, small chlorotic leaves, and small and malformed fruits (SOUZA; MÜLLER, 2006). Although it does not usually kill the tree, the disease reduces tree vigor and production, resulting in significant economic losses (DAWSON et al., 2013).

Planting of 'Pêra' sweet orange was discouraged in the late 1950s due to the susceptibility to CTV. Nevertheless, this sweet orange became the leading citrus cultivar in Brazil due to the fruit quality for fresh market and industrial processing. The cross protection technique with mild complexes of the virus allowed growing 'Pêra' sweet orange grafted on CTV tolerant rootstock under an endemic presence of the virus (TEÓFILO SOBRINHO et al., 2001). Nowadays, more than 90 million cross protected 'Pêra' trees were planted in Brazil (MORENO et al., 2008). However, CTV continues to be a threat due to the possibility of the development of new severe CTV complexes (FOLIMONOVA, 2013). Reduction of protection against CTV in cross protected 'Pêra' trees has been reported (MÜLLER et al., 1999; CORAZZA-NUNES et al., 2001; SOUZA et al., 2002).

Changes in the CTV protective complex and the presence of severe strains of the virus have been reported in the State of Paraná, Brazil (CARRARO et al., 2003; COSTA et al., 2010; CORAZZA et al., 2012; ZANUTTO et al., 2013). Consequently, the use of a single mild strain in a cross protection program for 'Pêra' sweet orange may represent a high risk (MÜLLER et al., 1999). Furthermore, the use of mild strains of the virus from cross protection programs from other Brazilian States may not provide adequately protection against CTV complexes present in Paraná (CORAZZA et al., 2012; ZANUTTO et al., 2013). Therefore, the evaluation of the agronomic performance of different cultivars of 'Pêra' sweet orange and their reaction to citrus tristeza under different environmental conditions is certainly an important strategy to select the best ones to be planted in the different regions of the State of Paraná. This study was carried out to evaluate the reaction of 'Pêra' sweet orange cultivars and selections to citrus tristeza based on stem pitting severity and to establish a correlation with agronomic performance under the environmental conditions of the North and Northwest regions of the State of Paraná, Brazil.

MATERIALS AND METHODS

Plant material and CTV complexes

Seven cultivars and six selections of 'Pêra' sweet orange (*Citrus sinensis* (L.) Osbeck), grafted on Rangpur lime (*Citrus limonia* Osbeck) rootstock, were included in this study. The field plots were established in the municipalities of Arapongas (latitude of 23° 27' 39.23" S, longitude of 51° 24' 51.45" W and altitude of 759 m), Cambé (latitude of 23° 04' 50.43" S, longitude of 51° 18' 4.18" W and altitude of 456 m) and Londrina (latitude of 23° 21' 50.41" S, longitude of 51° 10' 24.58"

W and altitude of 535 m), located in the North region, and Paranavaí (latitude of 23° 05' 41.49" S, longitude of 52° 26' 32.55" W and altitude of 469 m), located in the Northwest region. According to the Brazilian Soil Classification System (SANTOS et al., 2006), the experiments were conducted on a Typical Eutroferric Red Nitosol with a moderate A horizon and clayey texture, Typical Eutroferric Red Latosol with a moderate A horizon and very clayey texture, Eutroferric Latosolic Red Nitosol with a moderate A horizon and very clayey texture, and Typical Dystrophic Red Latosol with a moderate A horizon and medium texture for the municipalities of Arapongas, Cambé, Londrina and Paranavaí, respectively. The average annual temperatures are 20.6 ºC, 21.4 ºC, 21.1 ºC and 22.2 ºC, for these municipalities, respectively, and the climate of both regions is classified as Cfa type (CAVIGLIONE et al., 2000).

The nursery trees were planted on the second half of 2000 in a spacing of 7.0 x 4.0 m, without irrigation. The experimental design was completely randomized, with 13 treatments and five replicates for each treatment. The citrus trees were grown according to the cultural and phytosanitary practices recommended for the North and Northwest regions of the State of Paraná, Brazil (IAPAR, 1992).

Assessment of the severity of citrus tristeza symptoms in 'Pêra' sweet orange cultivars and selections

'Pêra' sweet orange cultivars and selections were evaluated in regard to the severity of tristeza stem pitting symptoms caused by the CTV complex on the second half of 2011. Circular and lignified branches with approximately 0.5 to 1.0 cm in diameter and 15 cm long were collected from the middle part of each quadrant, comprising a total of eight branches per tree. The branches were autoclaved at 120 ºC for five minutes to facilitate bark removal. Stem pitting was evaluated based on the intensity and severity, using a scale from 0 to 5, adapted from Meissner Filho et al. (2002), where 0 = no stem pitting; 1 = few surface stem pitting; 2 = sparse moderate stem pitting; 3 = intermediate surface stem pitting, rarely deep; 4 = large amount of surface and deep stem pitting; and 5 = branch completely covered by stem pitting.

Evaluation of vegetative growth

Vegetative growth of the 'Pêra' sweet orange cultivars and selections was evaluated on April 2012. Canopy height and diameter (m) were measured by using a graduated scale, and the trunk circumference was determined 10 cm below and 10 cm above the grafting line. The vegetative tree vigor index was determined by using the following equation (BORDIGNON et al., 2003b): [height (m) + mean canopy diameter (m) + (trunk diameter below grafting line (cm)/100 x 10)]. The canopy volume was determined by using the equation $V = \frac{2}{3} \pi R^2 H$ (MENDEL, 1956), where V, R and H represents the canopy volume (m^3), canopy radius (m) and tree height (m), respectively.

Evaluation of fruit yield

Fruit yield was determined in the experiments of Cambé and Londrina, for the 2010/2011, 2011/2012, and 2012/2013 harvests. Fruit size was evaluated by using wood boards with openings of 54, 64, and 73 mm in diameter. The fruits were classified as very small (<54 mm), small (54-64 mm), medium (64-73 mm), and large (>73 mm). The number and mass of the fruits were also determined for each class. The total yield of each tree was determined based on the number and mass of the fruits comprising all four different fruit classes. The yield efficiency index (kg m^{3-1}) was determined based on the ratio between the mean annual yield (kg) and canopy volume (m^3).

Data analysis

Data on citrus tristeza severity, yield and vegetative tree growth rates were analyzed individually by ANOVA test, followed by an analysis combining all the disease severity data of the four locations and all the thirteen 'Pêra' sweet orange cultivars and selections. The Scott-Knott test at 5% significance level was applied to group means by using the Sisvar software (FERREIRA, 2010). The percentage of fruits per size class was transformed using the arcsine square root of x/100 equation. The Pearson linear correlation between disease severity and agronomic variables was calculated using the t-test.

RESULTS AND DISCUSSION

Citrus tristeza severity differed significantly among the cultivars and selections of 'Pêra' sweet orange included in this study (Table 1). The cultivar 'Gullo', 'IPR 153' (Bianchi), 'IPR 158' (Vac. 3), 'IPR 159' (Vac. 4), 'Morretes', and Selection 27 had the best performance regarding citrus tristeza stem pitting severity, with an average score below 2.30, considering the data of all four locations (Table 1). In contrast, the other 'Pêra' cultivars and selections had stem pitting severity above 2.40 (Table 1). Based on each location individually, the lowest stem pitting severity was observed for the trees in the plot of Cambé, with 1.86 disease severity score, while the highest level was for the trees in Arapongas, with an average of 2.70 on stem pitting severity (Table 1). The expression of citrus tristeza stem pitting symptoms is usually highly dependent on environmental conditions (DODDS et al., 1987; SALIBE et al., 2002). Further, multiplication of CTV is also dependent of environmental conditions, especially during colder seasons, when the concentration of the virus attains higher titers in the plant tissue and consequently increases the expression of disease symptoms (DODDS et al., 1987). Therefore, stem pitting was more severe in trees of the locations with lower temperatures, such as Arapongas and Londrina (Table 1). These results are in agreement with those obtained in other studies, as the highest severity levels of citrus tristeza disease stem pitting in 'Pera' sweet orange were observed for trees in regions with milder climates (SALIBE et al., 2002).

The cultivar 'IPR 153' (Bianchi) had the best performance in regard to stem pitting severity in all four locations. Some cultivars of 'Pêra', as 'Gullo', 'Morretes', 'D-6', 'IPR 159' (Vac. 4), 'IPR 158' (Vac. 3), and Selections 11, 12, 15 and 27 performed very well in regard to citrus tristeza in more than one location (Table 1). Further, the cultivar 'Morretes' was the only one that did not show any significant difference between citrus tristeza disease severity and field plot location (Table 1). Selections 11 and 12 were the most influenced by the environmental conditions. Trees of these two selections had the highest disease severity score in Arapongas where disease severity attains the highest levels (Table 1). In addition to environmental conditions, severity of citrus tristeza disease may vary according to the aggressiveness of the viral complex present in the citrus tree. The citrus tristeza disease severity data obtained in this study are strongly consistent with those of Costa et al. (2010) that evaluated the clones 'Pêra Bianchi 89 C', 'Pêra D-6 111 A', 'Pêra Vacinada 59 B', and 'Pêra Vimusa 66 B', and Selection 37, also in the North region of the State of Paraná. Although 'Pêra Bianchi 89 C' had shown the highest citrus tristeza disease severity index among all 'Pêra' cultivars 'Gullo' (Costa et al*.,* 2010), in our studies 'IPR 153' (Bianchi), the same cultivar as 'Pêra Bianci 89 C', had a disease severe index of only 2.07, based on data from four different locations (Table 1). In another study with accessions of Pêra sweet oranges from the Citrus Germplasm Collection of the Instituto Agronômico do Paraná-IAPAR, in Londrina, I-58 Vacinada 3, I-59 Vacinada 4, and I-89 Bianchi, here designated IPR 158, IPR 159 and IPR 153, respectively, showed severe stem pitting symptoms (BABA et al., 2014). These differences in citrus tristeza disease severity may be due to the presence of different CTV haplotypes, in combination with differences in sensibility of the plant tissues to the virus, besides the effect of environmental conditions (BABA et al., 2014), as discussed previously in this study.

The cultivars and selections of 'Pêra' showed differences in regard to vegetative tree growth related to the location of the field plot, except for Paranavaí (Tables 2 and 3). In the field plots of Arapongas and Londrina, Selection 27 had the largest vegetative tree growth with vigor index of 10.23 and 11.40, respectively (Tables 2 and 3). In contrast, the vegetative tree growth of this Selection did not differ from the ones of the cultivars 'IPR 153' (Bianchi) and 'Morretes', and Selections 11, 12, 14, 15, and 37 in the field plot of Cambé (Table 2). In this location, these 'Pera' cultivars and selections had the highest values for this parameter (Table 2).

In Arapongas and Londrina field plots, the vegetative growth of the 'Pêra' sweet orange trees may have been determined by the combination of the aggressiveness of the CTV complex present in the clone or selection with environmental conditions of these locations. The development of the citrus trees, including canopy diameter and volume, vegetative vigor index and tree height, was negatively correlated with the severity of the disease symptoms in these two locations (Tables 2 and 3). These data confirm the results obtained in previous studies, where a negative correlation between these citrus trees development variables and stem pitting severity were already established for 'Pêra' sweet orange cultivars of the Citrus Germplasm Collection of IAPAR (BABA et al., 2014).

The yield of 'Pêra' sweet orange cultivars and selections were evaluated for three seasons in Cambé and Londrina field plots (Tables 4 and 5). In Cambé, the cultivars and selections did not differ

significantly in regard to fruit yield, which ranged from 141.33 kg for 'D-6' up to 221.36 kg per tree for 'IPR 153' (Bianchi) (Table 4). The highest yield efficiency indexes were obtained for trees of the cultivars 'IPR 159' (Vac. 4), 'Vimusa', 'D-6', 'Gullo', and 'IPR 158' (Vac. 3) with more than 3.8 kg m^{3-1} (Table 4). However, the number of fruits and yield were negatively correlated with citrus tristeza disease severity (Tables 4 and 5). In Londrina, the highest yield was obtained for 'Vimusa' and Selections 12 and 27, with more than 110 kg/tree (Table 5). These 'Pêra' cultivar and selections also had the highest numbers of fruits, ranging from 864 up to and 1054 fruits per tree (Table 5). The number of fruits, yield and fruit mass were also negatively correlated with stem pitting severity (Table 5). Furthermore, fruit size was significantly correlated with stem pitting severity, whereas fruit class <54 was positively correlated and fruit class 64-73 was negatively correlated with disease severity (Table 5). These data indicates a strong relationship between high citrus tristeza disease severity and reduced fruit size for 'Pêra' sweet orange cultivars and selections.

Based on the results obtained in our studies, citrus tristeza disease severity in 'Pêra' sweet orange cultivars and selections was highly related to environmental conditions. Further, the disease severity was negatively correlated with most of the agronomic characteristics of the citrus trees in regard to both, vegetative growth and yield. In other studies with 'Valência' sweet orange, the most affected agronomic characteristics of the citrus trees by citrus tristeza disease were fruit mass and tree yield, followed by vegetative traits, as tree vigor and height, and trunk diameter (BORDIGNON et al., 2003b). On the other hand, industrial characteristics of the fruits were barely affected (BORDIGNON et al., 2003b). Baba et al. (2014) also evaluated the effect of the citrus tristeza disease severity on the agronomic performance of 24 accessions of 'Pêra' and related sweet orange. They observed the highest correlations between disease severity and yield, number of fruits, and vegetative vigor index. In the present study, the vegetative vigor index did not correlate with the citrus tristeza disease severity for the citrus trees in the Cambé and Paranavaí field plots (Tables 2 and 3), unlike the results obtained for the Arapongas and Londrina field plots (Tables 2 and 3). Therefore, environmental conditions may have played a major role on the development of citrus tristeza disease and vegetative and yield characteristics of 'Pêra' sweet orange cultivars and selections. Further, strong negative correlations between disease severity and tree growth and fruit characteristics were observed basically in areas with mild climate conditions, where citrus tristeza disease symptoms were more severe.

TABLE 1 - Stem pitting severity due to citrus tristeza caused by *Citrus tristeza virus* (CTV) in cultivars and selections of 'Pêra' sweet orange in four locations of the State of Paraná, Brazil.

Cultivar or Selection	Location				Mean stem pitting score
	Arapongas	Cambé	Londrina	Paranavaí	
'D-6'	2.63a[1] B[2]	2.07 b A	2.90 b B	2.04 a A	2.41b
'Gullo'	2.25a B	1.61 a A	2.50 a B	2.11 b B	2.12a
'IPR 153'(Bianchi)	2.41a B	1.69 a A	2.49 a B	1.68 a A	2.07a
'IPR 158' (Vac. 3)	2.64a B	1.50 a A	2.69 b B	2.27 b B	2.28a
'IPR 159' (Vac. 4)	2.84b B	1.65 a A	2.81 b B	1.84 a A	2.28a
'Morretes'	2.33a A	2.04 b A	2.28 a A	2.06 a A	2.18a
Selection 11	3.25b C	1.82 a A	2.51 a B	2.46 b B	2.51b
Selection 12	2.97b C	1.84 a A	2.54 a B	2.45 b B	2.45b
Selection 14	3.10b B	2.32 b A	2.93 b B	2.26 b A	2.65b
Selection 15	2.73a B	1.86 a A	3.00 b B	2.15 b A	2.44b
Selection 27	2.36a B	1.58 a A	2.27 a B	2.33 b B	2.13a
Selection 37	3.10b B	2.00 b A	2.43 a A	2.18 b A	2.43b
'Vimusa'	2.50a B	2.14 b A	2.72 b B	2.30 b A	2.42b
Mean stem petting score per location	2.70 C	1.86 A	2.62 C	2.16 B	
CV (%)[3]	14.96	15.19	13.89	14.27	14.69

[1]Stem pitting severity. Means followed by the same lowercase letter in the column did not differ according to the Scott-Knott test ($p<0.05$).
[2]Means followed by the same uppercase letter in the same row did not differ according to the Scott-Knott test ($p<0.05$).
[3]CV (%) = coefficient of variation.

TABLE 2 - Vegetative growth and correlation with citrus tristeza stem pitting severity in cultivars and selections of 'Pêra' sweet orange in Arapongas and Cambé, State of Paraná, Brazil.

Cultivar or Selection	Location							
	Arapongas				Cambé			
	Tree height (m)	Canopy diameter (m)	Canopy volume (m^3)	Vegetative vigor index	Tree height (m)	Canopy diameter (m)	Canopy volume (m^3)	Vegetative vigor index
'D-6'	2.68 a[1]	3.37 a	15.93 a	7.73 a	3.31 a	4.27 a	32.83 a	9.41 a
'Gullo'	2.58 a	3.50 a	16.62 a	7.77 a	3.81 b	4.66 b	43.68 b	10.37 b
'IPR 153' (Bianchi)	3.23 b	3.85 a	25.80 b	8.99 b	4.13 c	5.73 d	71.20 c	12.13 c
'IPR 158' (Vac. 3)	2.89 a	3.50 a	18.96 a	8.12 a	3.76 b	4.93 b	48.27 b	10.66 b
'IPR 159' (Vac. 4)	3.09 b	4.10 b	27.32 b	9.06 b	3.70 b	4.94 b	47.51 b	10.67 b
'Morretes'	2.78 a	3.64 a	20.23 a	8.22 a	4.02 c	5.26 c	58.80 c	11.42 c
Selection 11	3.13 b	3.54 a	20.65 a	8.72 a	4.16 c	5.28 c	60.87 c	11.74 c
Selection 12	3.25 b	3.87 a	26.11 b	9.09 b	4.08 c	5.13 c	56.50 c	11.55 c
Selection 14	3.20 b	3.59 a	22.18 a	8.71 a	4.07 c	5.09 c	55.72 c	11.35 c
Selection 15	3.30 b	3.98 b	27.47 b	9.33 b	4.20 c	4.97 b	54.77 c	11.53 c
Selection 27	3.53 b	4.55 c	38.78 c	10.23 c	4.21 c	5.27 c	61.49 c	11.76 c
Selection 37	2.98 a	3.60 a	20.81 a	8.61 a	4.01 c	5.21 c	57.81 c	11.46 c
'Vimusa'	2.86 a	3.78 a	21.88 a	8.52 a	3.36 a	4.85 b	42.45 b	10.13 b
CV (%)[2]	8.08	10.44	26.45	8.06	6.01	6.80	16.45	6.03
Correlation between vegetative growth and disease severity[3]	-0.06 ^{ns}	-0.45 **	-0.39 **	-0.25 **	-0.19 ^{ns}	-0.19 ^{ns}	-0.20 ^{ns}	-0.18 ^{ns}

[1]Means followed by the same lowercase letter in the column did not differ according to the Scott-Knott test ($p<0.05$).
[2]CV (%) = Coefficient of variation.
[3]Pearson's linear correlation coefficient between vegetative growth and stem pitting severity. **Significant at $p<0.05$; ^{ns}not significant.

TABLE 3 - Vegetative growth and correlation with citrus tristeza stem pitting severity in cultivars and selections of 'Pêra' sweet orange in Londrina and Paranavaí, State of Paraná, Brazil.

Cultivar or Selection	Location							
	Londrina				Paranavaí			
	Tree height (m)	Canopy diameter (m)	Canopy volume (m^3)	Vegetative vigor index	Tree height (m)	Canopy diameter (m)	Canopy volume (m^3)	Vegetative vigor index
'D-6'	2.35a[1]	3.43a	14.98a	7.16a	3.11a	3.40a	20.91a	8.19a
'Gullo'	3.26c	4.21b	30.78b	9.16b	3.37a	4.12a	30.31a	9.23a
'IPR 153' (Bianchi)	2.97b	3.95b	24.83a	8.60b	3.76a	3.88a	31.44a	9.67a
'IPR 158' (Vac. 3)	3.22c	3.93b	26.21a	9.02b	3.24a	3.96a	27.97a	8.82a
'IPR 159' (Vac. 4)	3.28c	4.53c	35.28b	9.78c	3.13a	3.43a	20.96a	7.90a
Morretes	3.46c	4.34c	33.85b	9.52c	3.28a	4.10a	29.29a	9.19a
Selection 11	3.07b	3.96b	25.53a	8.88b	3.50a	4.24a	33.50a	9.98a
Selection 12	3.70d	4.34c	37.22b	10.07c	3.13a	3.71a	23.39a	8.85a
Selection 14	2.83b	3.73a	21.16a	8.26b	3.10a	3.84a	25.70a	8.89a
Selection 15	3.33c	4.17b	30.63b	9.52c	3.50a	4.00a	34.23a	9.86a
Selection 27	4.14e	5.13d	58.15c	11.40d	3.05a	4.04a	27.29a	9.14a
Selection 37	3.51c	4.02b	29.62b	9.40c	3.62a	4.18a	34.48a	9.81a
'Vimusa'	3.63d	4.59c	40.25b	10.05c	3.14a	4.04a	27.19a	9.10a
CV (%)[2]	8.73	8.79	22.38	6.98	14.68	16.83	41.43	13.86
Correlation between vegetative growth and disease severity[3]	-0.34**	-0.46**	-0.45**	-0.41**	-0.12ns	0.13ns	0.02ns	0.07ns

[1]Means followed by the same lowercase letter in the column did not differ according to the Scott-Knott test ($p<0.05$).
[2]CV (%) = Coefficient of variation.
[3]Pearson's linear correlation coefficient between vegetative growth and stem pitting severity. **Significant at $p<0.05$; nsnot significant.

TABLE 4 - Fruit yield of cultivars and selections of 'Pêra' sweet orange in Cambé, State of Paraná, Brazil, for the 2010/2011, 2011/2012 and 2012/2013 harvests and correlation with citrus tristeza stem pitting severity.

Cultivar or Selection	Number of fruits	Annual yield (kg/pl)	Fruit mass(g)	Fruit class (% of fruits per size class (mm)[2])				Yield efficiency index (kg m^{-3})	
				<54	54-64	64-73	>73		
'D-6'	781.53 a[1]	141.33 a	180.38 a	2.06 a	20.46 a	49.49 a	32.65 b	4.28	b
'Gullo'	923.93 a	170.18 a	184.26 a	2.17 a	15.98 a	47.83 a	37.21 b	3.97	b
'IPR 153' (Bianchi)	1298.07 a	221.36 a	170.64 a	4.67 a	26.49 a	50.09 a	26.42 a	3.12	a
'IPR 158' (Vac. 3)	1085.73 a	184.10 a	169.09 a	2.92 a	28.31 a	48.88 a	26.21 a	3.83	b
'IPR 159' (Vac. 4)	1171.53 a	210.95 a	181.01 a	2.20 a	21.53 a	51.69 a	29.72 a	4.46	b
'Morretes'	1062.27 a	182.96 a	172.71 a	2.54 a	24.90 a	48.89 a	29.41 a	3.10	a
Selection 11	1089.60 a	206.85 a	189.23 a	2.19 a	23.81 a	52.35 a	27.04 a	3.40	a
Selection 12	1145.40 a	198.04 a	173.64 a	8.25 a	22.32 a	43.83 a	34.93 b	3.50	a
Selection 14	983.20 a	179.16 a	181.65 a	3.36 a	23.59 a	51.67 a	27.39 a	3.26	a
Selection 15	1075.13 a	192.71 a	179.83 a	3.65 a	26.07 a	51.27 a	25.60 a	3.51	a
Selection 27	1106.40 a	183.89 a	164.18 a	6.67 a	28.13 a	50.54 a	21.74 a	2.96	a
Selection 37	1151.47 a	200.19 a	173.38 a	5.06 a	25.77 a	53.39 a	22.99 a	3.48	a
'Vimusa'	983.00 a	176.34 a	178.62 a	2.00 a	21.56 a	47.64 a	33.55 b	4.35	b
CV (%)[3]	17.85	19.26	6.58	101.61	22.96	8.81	19.42	14.20	
Correlation between vegetative growth and disease severity[4]	-0.29**	-0.27**	0.01^{ns}	-0.07^{ns}	0.21^{ns}	0.13^{ns}	-0.18^{ns}	-0.01^{ns}	

[1]Means followed by the same lowercase letter in the column did not differ according to the Scott-Knott test ($p<0.05$).
[2]Data transformed by using arcsine square root of x/100. [3] CV (%) = coefficient of variation.[4]Pearson's linear correlation coefficient between yield and stem pitting severity. **Significant at $p<0.05$; ^{ns}not significant.

TABLE 5 - Fruit yield of cultivars and selections of 'Pêra' sweet orange in Londrina, State of Paraná, Brazil, for the 2010/2011, 2011/2012 and 2012/2013 harvests and correlation with citrus tristeza stem pitting.

Cultivar or Selection	Number of fruits	Annual yield (kg/pl)	Fruit mass (g)	Fruit class (% fruits per size class (mm)[2])				Yield efficiency index (kg m^{3-1})
				<54	54-64	64-73	>73	
'D-6'	411.68a[1]	52.04a	122.50a	18.79a	41.58b	38.88a	8.05a	3.46b
'Gullo'	632.71a	85.25b	134.18a	15.47a	41.95b	42.62a	8.42a	2.76a
'IPR 153' (Bianchi)	552.85a	86.36b	155.27b	9.98a	27.67a	50.41b	20.97c	3.45b
'IPR 158' (Vac. 3)	431.15a	60.69a	140.99b	7.06a	39.44b	46.69b	12.66a	2.35a
'IPR 159' (Vac. 4)	662.91a	82.37b	124.56a	16.32a	46.89b	37.35a	7.60a	2.35a
'Morretes'	752.86b	89.97b	118.94a	19.43a	45.42b	36.05a	8.18a	2.68a
Selection 11	630.92a	94.14b	148.24b	10.56a	32.20a	50.80b	15.98b	3.67b
Selection 12	864.25c	110.43c	128.83a	14.65a	41.41b	42.32a	11.21a	2.85a
Selection 14	577.18a	74.63b	127.82a	14.93a	42.13b	41.12a	10.36a	3.49b
Selection 15	714.03b	94.21b	130.61a	12.90a	41.10b	44.45b	7.71a	3.06b
Selection 27	1054.52c	138.82c	130.80a	13.18a	39.45b	44.67b	11.62a	2.43a
Selection 37	425.18a	55.74a	131.69a	8.23a	41.06b	46.32b	8.33a	1.89a
'Vimusa'	914.78c	120.31c	131.80a	14.76a	39.21b	41.74a	16.03b	3.01b
CV (%)[3]	26.80	29.35	8.16	46.16	12.55	14.60	30.86	16.85
Correlation[4]	-0.27**	-0.35**	-0.36 **	0.36 **	0.23ns	-0.34**	-0.17ns	0.11ns

[1]Means followed by the same lowercase letter in the column did not differ according to the Scott-Knott test ($p<0.05$).
[2]Data transformed by using arcsine square root of x/100. [3] CV (%) = coefficient of variation.
[4]Pearson's linear correlation coefficient between yield and stem pitting severity.
**Significant at $p<0.05$; nsnot significant.

CONCLUSIONS

Cultivars 'IPR 153' (Bianchi), Gullo, Morretes, 'IPR 158' (Vac. 3), and 'IPR 159' (Vac. 4), and Selection 27 of 'Pêra' sweet orange have the lowest levels of citrus tristeza disease severity.

Expression of citrus tristeza disease symptoms is more severe in Arapongas and Londrina, where mild climate are prevalent. Under this condition, Selection 27 has high vegetative growth and yield, besides low citrus tristeza disease symptoms.

Severity of citrus tristeza disease symptoms is negatively correlated with vegetative growth and yield for areas with mild environmental conditions, such as Arapongas and Londrina.

ACKNOWLEDGEMENTS

To Araucária Foundation for the Scientific and Technological Development of Paraná (Fundação Araucária de Apoio ao Desenvolvimento Científico e Tecnológico do Paraná) for financial support; to Paraná Agronomic Institute (Instituto Agronômico do Paraná – IAPAR) for infrastructure and field support, and to João Henrique Caviglione for providing climate data analysis and soil classification.

REFERENCES

AULER, P.A.M.; LEITE JÚNIOR, R.P.L.; TAZIMA, Z.H.; ANDRADE, P.F.S. A citricultura no Paraná. **Citricultura Atual**, Cordeirópolis, n. 99, p.17-20, 2014.

BABA, V.Y.; GIAMPANI, J.S.; TAZIMA, Z.H., YADA, I.F.U., PACCOLA-MEIRELLES, L.D.; LEITE JÚNIOR, R.P. Agronomic performance of Pera and related sweet orange accessions naturally infected with *Citrus tristeza virus* in northern Paraná State State, Brazil. **Tropical Plant Pathology**, Brasília, v.39, n.6, p. 442-448, 2014.

BENNETT, C.W.; COSTA, A.S. Tristeza disease of citrus. **Journal of Agricultural Research**, Washington, v.78, n.8, p.207-237, 1949.

BORDIGNON, R.; MEDINA FILHO, H.P.; MULLER, G.W.; SIQUEIRA, W.J. A tristeza dos citros e suas implicações no melhoramento genético de porta-enxertos. **Bragantia**, Campinas, v.62, n.3, p.345-355, 2003a.

BORDIGNON, R.; MEDINA FILHO, H.P.; SIQUEIRA, W.J.; PIO, R.M. Efeito da tristeza em caracteres vegetativos, produtivos e industriais da laranjeira 'Valência' enxertada em híbridos segregando para tolerância. **Bragantia**, Campinas, v.62, n.2, p.207-215, 2003b.

CARRARO, B.P.; NUNES, W.M.C.; CORAZZA-NUNES, M.J.; MACHADO, M.A.; STACH-MACHADO, D.R. Avaliação de complexos do *Citrus tristeza virus* da região Norte do Paraná por meio de testes imunológicos e SSCP do gene da capa protéica. **Acta Scientiarum**, Maringá, v.25, n.2, p.269-273, 2003.

CAVIGLIONE, J. H.; KIIHL, L. R. B.; CARAMORI, P. H.; OLIVEIRA, D. **Cartas climáticas do Paraná**. Londrina: IAPAR, 2000. CD-ROM.

CORAZZA-NUNES, M.J.; MACHADO, M.A.; MÜLLER, G.W.; STACH-MACHADO, D.R.; SOUZA, A.A.; NUNES, W.M.C. Evaluation of *Citrus tristeza virus* (CTV) complexes in pre-immunized Marsh seedless grapefruits. **Summa Phytopathologica**, Jaboticabal, v.27, p.11-16, 2001.

CORAZZA, M.J.; ZANUTTO, C.A.; ZANINELI-RÉ, M.L.; MÜLLER, G.W.; NUNES, W.M. de C. Comparison of *Citrus tristeza virus* (CTV) isolates by RFLP analysis of the coat protein nucleotide sequences and by the severity of the symptoms. **Tropical Plant Pathology**, Brasília, v.37,n.3, p.179-184, 2012.

COSTA, A.T.; NUNES, W.M.C; ZANUTTO, C.A; MÜLLER, G.W. Stability of *Citrus tristeza virus* protective isolates in field conditions. **Pesquisa Agropecuária Brasileira**, Brasília, v.45, n.7, p.693-700, 2010.

DAWSON, W.O.; GARNSEY, S.M.; TATINENI, S.; FOLIMONOVA, S.Y.; HARPER, S.J.; GOWDA, S. *Citrus tristeza virus*- host interactions. **Frontiers in Microbiology**, Lausanne, v.4, Article 88, p.1-10, 2013. doi:10.3389/fmicb.2013.00088

DODDS, J.A.; JORDAN, R.L.; ROISTACHER, C.N. & JARUPATI, T. Effects of strain, host, time of harvest and virus concentration on double-stranded RNA analysis of *Citrus tristeza virus*. **Phytopathology**, St. Paul, v.77, p.442-447, 1987.

DONADIO, L.C. **Laranja Pêra**. Jaboticabal: FUNEP, 1999. 51p. (Boletim Citrícola, 11).

FERREIRA, D. F. **Sisvar**: Sistema de análise de variância para dados balanceados, versão 5.3 Build 75. Lavras: DEX/ UFLA, 2010. Software.

FOLIMONOVA, S.Y. Developing an understanding of cross-protection by *Citrus tristeza virus*. **Frontiers in Microbiology**, Lausanne, v.4, Article 76, p.1-9, 2013. doi: 10.3389/fmicb.2013.00076.

IAPAR. **A citricultura do Paraná**. Londrina: IAPAR, 1992. 288p. (Circular, 72)

MEISSNER FILHO, P.E.; SOARES FILHO, W. dos S.; VELAME, K.V.C.; DIAMANTINO, E.P.; DIAMANTINO, M.S.A.S. Reação de porta-enxertos híbridos ao *Citrus tristeza virus*. **Fitopatologia Brasileira**, Brasília, v.27, n.3, p.312-315, 2002.

MENDEL, K. Rootstock-scion relationships in Shamouti trees on light soil. **Ktavim**, Rehovot, v.6, p.35-60, 1956.

MORENO, P.; AMBRÓS, S.; ALBIACH-MARTÍ, M.R.; GUERRI, J.; PEÑA, L. *Citrus tristeza virus*: a pathogen that changed the course of the citrus industry. **Molecular Plant Pathology**, Oxford, v.9, n.2, p.251-268, 2008.

MÜLLER, G.W.; TARGON, M.L.N. E MACHADO, M.A. Trinta anos de uso do clone pré- imunizado "Pêra" IAC na citricultura paulista. **Laranja**, Cordeirópolis, v.20, n. 2, p. 399-408, 1999.

PIO, R. M.; FIGUEIREDO, J. O.; STUCHI, E. S.; CARDOSO, S. A. B. Variedades copas. In: MATTOS JUNIOR, D.; PIO, R.M.; DE NEGRI, J. D.; POMPEU JUNIOR, J. (Ed.). **Citros**. Campinas: Instituto Agronômico. FUNDAG, 2005. Capítulo 3, p.37-60.

ROSSETTI, V.V. **Manual ilustrado de doenças dos citros**. Piracicaba: Fealq/Fundecitrus, 2001. 207 p.

SALIBE, A.A.; TEÓFILO SOBRINHO, J.; MÜLLER, G.W. Sinopse de conhecimentos e pesquisas sobre a laranja 'Pêra'. **Laranja**, Cordeirópolis, v. 23, n. 1, p. 231-245, 2002.

SANTOS, H. G. D.; JACOMINE, P. K. T.; ANJOS, L. H. C. D.; OLIVEIRA, V. Á. D.; OLIVEIRA, J. B. D.; COELHO, M. R.; LUMBRERAS, J. F.; CUNHA, T. J. F. **Sistema brasileiro de classificação de solo**. 2. ed. Rio de Janeiro: EMBRAPA - Centro Nacional de Pesquisa de Solos, 2006. 306p.

SOUZA, A.A.; MÜLLER, G.W. A premunização no controle da tristeza dos citros. **Laranja**, Cordeirópolis, v.27, n.1, p. 57-70, 2006.

SOUZA, A.A.; MÜLLER, G.W.; TARGON, M.L.P.N.; COLETA-FILHO, H.D.; MACHADO, M. Avaliação de haplótipos do gene do capsídeo do *Citrus tristeza virus* em plantas pré-imunizadas de laranja 'Pêra'. **Summa Phytopathologica**, Botucatu, v.28, n.2, p.154-159, 2002.

TANAKA, F.A.O.; KITAJIMA, E.W.; MACHADO, M.A.; MACHADO, S.R. Ontogênese de caneluras em pedúnculo de flores de laranjeira doce infectados pelo vírus da tristeza dos citros estirpe "Capão Bonito". **Summa Phytopathologica**, Botucatu, v.35, n.4, p.316-321, 2009.

TEÓFILO SOBRINHO, J.; MÜLLER, G.W.; FIGUEIREDO, J.O. de; LARANJEIRA, F.F.; SALIBE, A. A. Laranja 'Pêra IAC 2000'. **Laranja**, Cordeirópolis, v.22, n.2, p.495-501, 2001. **)**

ZANUTTO, C.A; CORAZZA, M.J; NUNES, W.M.C; MÜLLER, G.W. Evaluation of the protective capacity of new mild *Citrus tristeza virus* (CTV) isolates selected for a preimmunization program. **Scientia Agricola**, Piracicaba, v.70, n.2, p.116-124, 2013.