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Resistance to *Cowpea aphid-borne mosaic virus* in *in vitro* germinated genotypes of *Passiflora setacea*

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Abstract- Passion fruit (*Passiflora edulis*) is a fruit species of great relevance for Brazilian economy. However, it is highly susceptible to the *Cowpea aphid-borne mosaic virus* (CABMV). The species *P.setacea*, on the other hand, is resistant to the disease. The present study aimed to identify CABMV-resistant *P. setacea* genotypes for the introgression of genes into sour passion fruit genetic breeding programs. The seeds of passion fruit genotypes were germinated *in vitro* in MS culture medium. The seedlings were acclimatized in a growth chamber at the temperature of 27 ± 2ºC, photoperiod of 16:8 hours and 80% of relative humidity. Thirty plants of each genotype were mechanically inoculated with extract prepared from leaves collected from passion fruit plants with symptoms of CABMV for the assessment of resistance to CABMV. The severity of leaf symptoms was evaluated by means of a grading scale of visual signs. After the visual evaluation and identification of the asymptomatic genotypes of *P. setacea*, the PTA-ELISA test was carried out for 30 selected genotypes. According to the visual evaluation, all *P. setacea* genotypes were highly resistant to CABMV, while the *P. edulis* genotypes were highly susceptible. Out of the 30 genotypes selected, only *Ps*RJ 4 was considered susceptible by PTA-ELISA. The other genotypes of *P. setacea* were considered resistant and present great potential for use in passion fruit genetic breeding programs.
Index terms: PTA-ELISA, fruit hardening virus, passion fruit, plant resistance.

Resistência ao *Cowpea aphid-borne mosaic* virus em genótipos de *Passiflora setacea* germinadas *in vitro*

Resumo – O maracujazeiro-azedo (*Passiflora edulis*) é uma espécie frutífera de grande importância econômica para o Brasil, porém altamente susceptível ao vírus *Cowpea aphid-borne mosaic vírus* (CABMV). A espécie *P. setacea*, em contrapartida, apresenta resistência à doença. O objetivo deste estudo foi identificar genótipos de *P. setacea* resistentes ao CABMV, visando à introgressão de genes em programas de melhoramento genético do maracujazeiro-azedo. As sementes dos genótipos de maracujazeiro foram germinadas *in vitro* em meio de cultura MS. A aclimatização das plântulas ocorreu em câmara de crescimento com temperatura de 27± 2 ºC, fotoperíodo de 16h8 e 80% de umidade relativa. Para avaliar a resistência ao CABMV, foram utilizadas 30 plantas de cada genótipo, inoculadas mecanicamente com extrato preparado a partir de folhas coletadas de plantas de maracujazeiro-azedo com sintomas de infecção pelo CABMV. A severidade dos sintomas foliares foi avaliada por meio de uma escala de notas visuais. Após a avaliação visual e a identificação dos genótipos assintomáticos de *P. setacea*, realizou-se o teste PTA-ELISA em 30 genótipos selecionados. Pela avaliação visual, todos os genótipos de *P. setacea* foram altamente resistentes ao CABMV, e os genótipos de *P. edulis,* altamente suscetíveis. Dos 30 genótipos selecionados, apenas o *Ps*RJ 4 foi considerado suscetível pelo PTA-ELISA. Os demais genótipos de *P. setacea* foram considerados resistentes e apresentam grande potencial de serem utilizados em programas de melhoramento genético do maracujazeiro.
Termos para indexação: PTA-ELISA, virose do endurecimento dos frutos, maracujazeiro, resistência de plantas.

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Introduction

Passion fruit belongs to the genus *Passiflora,* with approximately 400 species identified. Out of these, 143 species are native to Brazil, which is considered the center of genetic diversity of *Passiflora spp*. (*Passiflora in* ..., 2017). The cultivation of sour passion fruit (*P. edulis*) is very important for the agricultural sector because the fruit is widely used and accepted in the world market (Meletti, 2011). Brazil produced 700 thousand tons of passion fruit in 2015 (IBGE, 2015)and is the world's largest producer of the fruit.

Reduced productivity and longevity have been recorded for this crop due to diseases. These are considered limiting factors for the maintenance and expansion of the culture, since they affect from the sowing to the adult stages of the plants and are harmful to roots, stem, leaves, flowers and fruits (Cerqueira-Silva et al., 2014). Passion fruit plants infected by CABMV have symptoms such as common mosaic, sometimes accompanied by wrinkling, deformations and blisters on the leaf blade. As a result of the disease, productivity is reduced quantitatively and qualitatively, since the fruits become smaller, deformed and hardened, and may even show cracks, depending on the strain of the virus (Nascimento et al., 2006; Cerqueira-Silva et al. Oliveira et al.2008; Oliveira et al., 2013). Fruit hardening virus, caused by the *Cowpea aphid-borne mosaic virus* (CABMV) is a species of the genus *Potyvirus* (Cerqueira-Silva et al., 2014; Rodrigues et al., 2015). It is transmitted by aphids, grafting and mechanically by means of buffered leaf extract and occurs in the main producing areas of Brazil (SILVA et al., 2012; GARCÊZ et al., 2015).

Wild passion fruit species have significant potential for use in breeding programs due to some of their desirable traits, such as resistance to viroses. Promising and fertile hybrids were obtained from directed crosses. Among these species, *Passiflora setacea* genotypes are resistant to CABMV and have been used in interspecific crosses with sour passion fruit (SANTOS et al., 2015).

Interspecific crosses involving *P. edulis* and *P. Setacea* were successfully performed aiming at the introgression of resistance genes. Several hybrids resistant to CABMV were identified (SANTOS et al., 2015) and backcrossed with *P. edulis*, thus generating a segregating population with resistant individuals and agronomic traits more similar to those of the commercial species (*P. edulis*) (FREITAS et al., 2015). Therefore, the identification of sources of resistance to CABMV is fundamental to breeding programs aiming to reduce productivity losses in passion fruit crops.

The present study aimed to identify *P. setacea* genotypes resistant to CABMV to be used in passion fruit genetic breeding programs.

Material and Methods

The cultivar of *P. edulis* germplasm (UENF 'Rio Dourado') was obtained from the intrapopulation recurrent selection program of the Universidade Estadual do Norte Fluminense Darcy Ribeiro (UENF), Campos dos Goytacazes, RJ. The *P. setacea* germplasm was obtained from various sources (Table 1).

Aiming to obtain the seedlings, the seed coat was removed from the seeds, which were disinfested in alcohol 70% for 30 seconds, in 0.25% sodium hypochlorite for 15 minutes and rinsed four times in deionized and autoclaved water, in a laminar flow cabinet (ALEXANDRE et al., 2009). Then, ten seeds were inserted in each glass vial (65x125 mm) containing 40 mL of MS medium and White vitamins (MURASHIGE; SKOOG, 1962), 30 g L^{-1} sucrose and 100 mg L^{-1}myo-inositol . The inoculated seeds were taken to the growth room at the temperature of 27 ± 2 ºC, photoperiod of 16:8 hours (light: dark) and 25 µmol m^{-2} s^{-1} light intensity provided by daylight fluorescent lamps (OSRAM ®, São Paulo, Brazil), for 40 days.

After *in vitro* germination, the seedlings obtained were acclimatized, transferred to 200 ml disposable plastic cups containing Basaplant® Hortaliça substrate (Base Agro, São Paulo, Brazil) and carried to a growth chamber, at 26 ± 2°C, 80% humidity and photoperiod of 16: 8 hours (light: dark).

The experiment for the assessment of passion fruit resistance to the CABMV was arranged in a completely randomized design (DIC), with three replicates. Each replicate consisted of 30 plants of each origin (Table 1), one plant per container.

As the source of inoculum, it wasused a CABMV isolate obtained from passion fruit plants with mosaic symptoms, blisters and foliar deformations, collected from the experimental area of the Colégio Agrícola Antônio Sarlo (Antônio SarloAgricultural School), in Campos dos Goytacazes, RJ. This isolate was characterized by means of RT-PCR (*Reverse Transcription - Polymerase Chain Reaction*), with primers to the cylindrical cytoplasmic inclusion (CI) protein sequence (HA et al. 2008)and *Plate Trapped Antigen - Enzyme Linked ImmunosorbentAssay* (PTA-ELISA) (SANTOS et al., 2015).

After four weeks of acclimatization, the plants were mechanically inoculated with extract from leaves of passion fruit plants with severe symptoms of CABMV infestation. The inoculum for mechanical transmission was prepared in a mortar, and the infected leaf material was macerated,at the ratio of 1 g of leaf tissue to 10 mL of 0.1 M sodium phosphate buffer solution (pH 7.0). Carborundum (600 mesh) was used as an abrasive. Forty-eight hours after the first inoculation, the plants were re-inoculated to avoid the incidence of leaks. One plant from each source treated with only buffer solution was used as control.

The results were evaluated for 30 days through daily observations of local and systemic symptoms, from the first day of inoculation. The severity of the leaf symptoms in passion fruit genotypes was visually assessed and rated according to a grading scale: 1 = no symptoms; 2 = light mosaic without leaf deformations; 3 = severe mosaic with leaf deformation; 4 = severe mosaic, blisters and leaf deformations:

The data obtained from the grading scale were used to calculate the Area Under the Disease Progress Curve (AACPD) (CAMPBELL; MADDEN, 1990) in each genotype evaluated, according to the expression.

where, $AACPD = \sum_{i=1}^{n-1} \frac{Y_i + Y_{i+1}}{2} T_{i+1} - T_i$

Y_i = proportion of the disease in thei-th observation;
T_i = time in days of thei-th observation;
n = number of observations.

The values obtained with the AACPD calculation were submitted to analysis of variance, after the normality conditions of the residues was verified by the Lilliefors test, with the aid of the Genes software system (CRUZ, 2013). The analysis adopted the following statistical model:

$Yij = \mu + Gi + eij$

μ = general constant
Gi = effect of the genotype (NID, 0, σ^2_G)
eij = random error (NID, 0, σ^2)

The mean square expectation estimates were obtained from the analysis of variance of the AACPD. The following parameters were estimated:

a) Environmental variance (σ^2_a): $\sigma_a^2 = \frac{QME}{r}$

b) Phenotypic variance (σ^2_f): $\sigma_f^2 = \frac{\sigma^2}{r} + \sigma_g^2$

c) Genotypic variance (σ^2_G): $\sigma_g^2 = \frac{QMG + QME}{r}$

d) Coefficient of genotypic variance (CV_g): $CV_g = \frac{100.\sqrt{\sigma_g^2}}{\overline{X}}$

e) Heritability (h^2): $h^2 = \frac{\sigma_g^2}{\sigma_f^2} = \frac{\sigma_g^2}{\sigma_g^2 + \sigma^2}$

f) Variation index (IV): $IV = \frac{CV_g}{CV_e}$

g) Coefficient of experimental variation (CV_e): $CV_e = \frac{100.\sqrt{\sigma_e^2}}{\overline{X}}$

After the visual evaluation and identification of the asymptomatic genotypes of *P. setacea*, the PTA-ELISA test was performed with a polyclonal antiserum specificallyagainst CABM, so as to confirm resistance to the CABMV in 30 genotypes free from the symptoms of the disease. Due to the large number of asymptomatic plants, ten plants were selected from each origin, with sufficient number of leaves to perform the PTA-ELISA test. The PTA-ELISA serological test was performed at the Agência Paulista de Tecnologia dos Agronegócios - Instituto Biológico, in the city of São Paulo - SP (Unidade Laboratorial de Referência em Fitossanidade– Reference Plant Health Laboratory Unit).

Leaf samples (1 g) of healthy*P. edulis* were used as a negative control (-) and were ground in coating buffer (1.59 g of Na_2CO_3 and 2.93 g of $NaHCO_3$for500 mL of H_2O, 0.05M, pH 9.6) at the ratio of 1/5 (g / ml). Leaves of *P. edulis* with mosaic symptoms and leaf deformations (infected with CABMV), macerated in 1/5 (g / mL) cover buffer were used as a positive control (+). Foliar samples of *P. setacea* were ground in a solution (0.1126 g DIECA, 0.0372 g and 0.057 g sodium thioglycolate) 1/5 ratio (g / mL) and diluted in PBSTPo (1200 mL of PBS-T, 2% of polyvinylpyrrolidone, PVP) 1/1 ratio (mL / mL). The extracts from positive and negative controls and samples were applied to 96-well polystyrene plates incubated at 37 °C, for 2 hours. Then, the plates were washed three times with phosphate buffer with Tween (PBST) (0.05% *Tween*-20® in 0.1M PBS, pH 7.4), blocked with 1% skimmed milk, diluted in PBSTPo and incubated at 37°C, for 2 hours. The plates were re-washed with PBST, followed by the specific antiserum against CABMV (AS-CABMV - Gentle supplied by Dr. JAM Rezende, ESALQ, USP), previously adsorbed, and diluted 1: 2000 in the presence of PBSTPo. The plates were incubated again at 37°C for 2 hours.The plates were washed (three times with PBST) and then it was added the alkaline phosphatase conjugatedanti-rabbit(Sigma) diluted in PBSTPo at the ratio of 1: 30000. Then, the plates were incubated at 37°C for 2 hours. After the last wash with PBST, the substrate (p-nitrophenylphosphate) was applied. After approximately 30 minutes, the plates were read using the *Microplate reader 3550-UV* (Bio-Rad) device, at the wavelength of 405 nm. The results were analyzed by the ratio between the mean of three readings of the infected samples and the reading of healthy samples (I/S). The samples were considered positive when the average of the absorbance readings was three times higher than that obtained for the negative control (SILVA et al., 2012).

Table 1-Description of germplasm origin.UENF, Campos dos Goytacazes-RJ, 2013.

Germplasm	Origin
P. setacea – BA	Germplasm Bank, genotype UESC 365– UESC
P. setacea – RJ	Germplasm Bank– UENF
P. setacea– MG	Nova Porteirinha (MG)
P. edulis – UENF 'Rio Dourado'	Passion fruit intrapopulacional recurrent selection

Results and Discussion

Significant difference was observed at 1% probability for AACPD, which indicates variability among the passion fruit genotypes investigated. The coefficient of variation was considered low (7.97%), which indicates good experimental accuracy during the conduction of the trial (Table 2).

The environmental variance was considered low, which reveals little effect of the environment on the expression of resistance to CABMV; this was already expected, since the plants were subjected to controlled temperature, humidity and photoperiod conditions. The estimates for genotypic variation (σ^2_G = 777.08) and genotypic variation coefficient (CV_g = 64.91) were considered high. This evidences the existence of wide genetic variability among genotypes, which can be exploited in passion fruit breeding programs (Table 3).

The heritability in the evaluated population was considered high (h^2 = 99.94%),which indicates low environmental influence. This demonstrates that resistance to CABMV for this population is exclusively genetic. High heritability values (99%) for resistance to CABMV were found in a segregating population resulting from the cross between *P. edulis* and *P. setacea* (SANTOS et al., 2015). Freitas et al., (2015) also found high values (94%) for resistance to CABMV in a population obtained from backcrossing between *P. edulis* and an interspecific hybrid (*P. edulis* x *P. setacea*). These results agree with those obtained in the present study. High heritability values may indicate that the trait under study is controlled by few genes. Studies indicate that resistance to CABMV is polygenic, but there may be gain in selection, provided that large populations and more complex methods are used, such as backcrossing and recurrent selection (FREITAS et al., 2015). There are no difficulties to obtain resistant cultivars when trait heritability is high (JUHÁSZ et al., 2008).This is true for both oligogenic and polygenic control of genetic resistance.

Table 2 - Summary of the analysis of variance for the variable AACPD in the genotypes tested for resistance to CABMV.UENF, Campos dos Goytacazes-RJ, 2013.

FV	GL	QM
Genotype	3	23324.40**
Error	116	11.72**
Mean	119	42.94
CV (%)		7.97

FV = source of variation, GL = degree of freedom, QM = average square. **Significant at 1% (P<0.01) by the F test.

Table 3 - Estimates of the genetic parameters obtained from the AACPD for *Passiflora edulis* and *Passiflora setacea*. UENF, Campos dos Goytacazes-RJ, 2013

Genetic Parameters	Values
Environmental variance (σ^2_a)	0.39
Phenotypic variance (σ^2_f):	777.48
Genotypic variance (σ^2_g)	777.08
Coefficient of genotypic variation (CV_g)	64.91
Variation Index (IV)	8.14
Heritability (h^2)	99.94

In this study, it was not possible to discriminate moderately resistant plants, since all genotypes of *P. setacea* were highly resistant, while those of *P. edulis* were highly susceptible. This can be easily observed by the high heritability value (Table 3), which corroborates that *P. setacea* resistance and *P. edulis* susceptibility were both highly inheritable traits for the population evaluated.

Throughout the visual evaluation period, wide symptom variation was observed in the different genotypes of passion fruit. The *P. setacea* genotypes did not show CABMV symptoms. Therefore, they were graded 1 at the end of the 30 days of evaluation. In contrast, all genotypes of *P. edulis* showed characteristic symptoms of CABMV infection, such as severe mosaic and leaf deformation, and were graded 4 at the end of the evaluation.

The presence of CABMV symptoms in *P. edulis* plants was observed nine days after the first inoculation. From the 11th day of monitoring, it was verified that some plants already presented grade 4. For the genotypes of *P. edulis*, differences were observed between the genotypes for the severity of CABMV symptoms until the 28th day of evaluation. On day 30, all plants were graded 4 (data not shown).

AACPD values ranged from 59.5 to 90.5 for the *P. edulis* genotypes. The lowest value was obtained for the genotype *Pe* 19 and the highest values, for the genotypes *Pe* 9, 14, 15, 16 and 20. The genotypes of *P. setacea* obtained the lowest values for AACPD (29) in all genotypes (Table 4).

The results for the AACPD values obtained were previously observed for *P. edulis* and *P.setacea* by Santos et al (2015). The authors assessed the presence of CABMV symptoms in *P. edulis* and *P. setacea* plants, and fond susceptibility and resistance, respectively, in these species. On the other hand, Maciel et al. (2009) evaluated the reaction of 16 *Passiflora* species to the infection with four Brazilian isolates of the CABMV and observed that *P. setacea* was resistant only to the isolate CABMV-RJ. Oliveira et al. (2013) evaluated the reaction of four *P. setacea* genotypes to the CABMV, under natural occurrence in the field and verified that only one genotype (BGM237) was considered resistant, in the three types of evaluations for the virus. Therefore, variability was observed for the reaction to CABMV within the *P. setacea* species. Besides, the evaluations for reaction to CABMV should be performed at the intraspecific level, mainly if these genotypes are used as parents in breeding programs aimed at the introgression of resistance. After visual evaluation and identification of asymptomatic *P. setacea* genotypes, the PTA-ELISA test was performed to corroborate resistance to CABMV. For such, 30 genotypes of *P. setacea* were randomly selected, ten from each origin. Out of the 30 samples analyzed, only the genotype *Ps*RJ 4 reacted with the se of the specific antiserum against CABMV. The samples were considered positive when the mean of the absorbance readings (405nm) was three times higher than those obtained for the negative control. Thus, the results corroborated the resistance of the genotypes *Ps*RJ 3, *Ps*RJ 5, *Ps*RJ 7, *Ps*RJ 10, *Ps*RJ 11, *Ps*RJ 13, *Ps*RJ 14, *Ps*RJ 17, *Ps*RJ 18, *Ps*BA 6, *Ps*BA 11, *Ps*BA 12, *Ps*BA 13, *Ps*BA 14, *Ps*BA 15, *Ps*BA 16, *Ps*BA 17, *Ps*BA 21, *Ps*BA 22, *Ps*MG 2, *Ps*MG 8, *Ps*MG 9, *Ps*MG 11, *Ps*MG 12, *Ps*MG 13, *Ps*MG 14, *Ps*MG 18, *Ps*MG 19 and *Ps*MG 24 to CABMV (Table 5).

Santos et al. (2015) used PTA-ELISA to corroborate the resistance to CABMV in interspecific hybrids of *Passiflora*. According to the authors, out of the 31 genotypes analyzed, three reacted to the antiserum specific against CABMV. The authors also considered positive samples, i.e. susceptible samples, when the mean of the absorbance readings (A 405 nm) was three times higher than those obtained for the negative control.

The genotype *Ps*RJ 4, described as resistant by visual evaluation, was considered susceptible by hePTA-ELISA test, possibly because more time was necessary for the virus to express the symptoms in the plant. Evaluations should be longer and the serological test should be repeated. Santos et al. (2006) assessed the resistance to CABMV in interspecific hybrids of *Passiflora* (*P.edulis* x *P. setacea*) and observed that some hybrids that were considered resistant, that is, asymptomatic by visual analysis, were indicated as susceptible by the PTA-ELISA test, which is in agreement with thefindings of this study.

Thus, in view of some disagreements between the visual evaluation and the PTA-ELISA test, it is considered that both evaluations should be carried out to corroborate the resistance to the CABMV virus in asymptomatic *P. setacea* genotypes.

Table 4 - AACPD values and classification of genotypes based on the visual evaluation of the symptoms caused by the CABMV virus.UENF, Campos dos Goytacazes-RJ, 2013.

Genotypes	Value	Classification[1]	Genotypes	Value	Classification[1]
*Ps*MG 1	29	R	*Ps*RJ 1	29	R
*Ps*MG 2	29	R	*Ps*RJ 2	29	R
*Ps*MG 3	29	R	*Ps*RJ 3	29	R
*Ps*MG 4	29	R	*Ps*RJ 4	29	S
*Ps*MG 5	29	R	*Ps*RJ 5	29	R
*Ps*MG 6	29	R	*Ps*RJ 6	29	R
*Ps*MG 7	29	R	*Ps*RJ 7	29	R
*Ps*MG 8	29	R	*Ps*RJ 8	29	R
*Ps*MG 9	29	R	*Ps*RJ 9	29	R
*Ps*MG 10	29	R	*Ps*RJ 10	29	R
*Ps*MG 11	29	R	*Ps*RJ 11	29	R
*Ps*MG 12	29	R	*Ps*RJ 12	29	R
*Ps*MG 13	29	R	*Ps*RJ 13	29	R
*Ps*MG 14	29	R	*Ps*RJ 14	29	R
*Ps*MG 15	29	R	*Ps*RJ 15	29	R
*Ps*MG 16	29	R	*Ps*RJ 16	29	R
*Ps*MG 17	29	R	*Ps*RJ 17	29	R
*Ps*MG 18	29	R	*Ps*RJ 18	29	R
*Ps*MG 19	29	R	*Ps*RJ 19	29	R
*Ps*MG 20	29	R	*Ps*RJ 20	29	R
*Ps*MG 21	29	R	*Ps*RJ 21	29	R
*Ps*MG 22	29	R	*Ps*RJ 22	29	R
*Ps*MG 23	29	R	*Ps*RJ 23	29	R
*Ps*MG 24	29	R	*Ps*RJ 24	29	R
*Ps*MG 25	29	R	*Ps*RJ 25	29	R
*Ps*MG 26	29	R	*Ps*RJ 26	29	R
*Ps*MG 27	29	R	*Ps*RJ 27	29	R
*Ps*MG 28	29	R	*Ps*RJ 28	29	R
*Ps*MG 29	29	R	*Ps*RJ 29	29	R
*Ps*MG 30	29	R	*Ps*RJ 30	29	R
*Ps*BA 1	29	R	*Pe*1	75.5	S
*Ps*BA 2	29	R	*Pe*2	75.5	S
*Ps*BA 3	29	R	*Pe*3	88.5	S
*Ps*BA 4	29	R	*Pe*4	87.5	S
*Ps*BA 5	29	R	*Pe*5	72.5	S
*Ps*BA 6	29	R	*Pe*6	86.5	S
*Ps*BA 7	29	R	*Pe*7	85.5	S
*Ps*BA 8	29	R	*Pe*8	84.5	S
*Ps*BA 9	29	R	*Pe*9	90.5	S
*Ps*BA 10	29	R	*Pe*10	84.5	S

*Ps*BA 11	29	R	*Pe*11	86.5	S
*Ps*BA 12	29	R	*Pe*12	86.5	S
*Ps*BA 13	29	R	*Pe*13	77.5	S
*Ps*BA 14	29	R	*Pe*14	90.5	S
*Ps*BA 15	29	R	*Pe*15	90.5	S
*Ps*BA 16	29	R	*Pe*16	90.5	S
*Ps*BA 17	29	R	*Pe*17	87.5	S
*Ps*BA 18	29	R	*Pe*18	88.5	S
*Ps*BA 19	29	R	*Pe*19	59.5	S
*Ps*BA 20	29	R	*Pe*20	90.5	S
*Ps*BA 21	29	R	*Pe*21	82.5	S
*Ps*BA 22	29	R	*Pe*22	88.5	S
*Ps*BA 23	29	R	*Pe*23	85.5	S
*Ps*BA 24	29	R	*Pe*24	78.5	S
*Ps*BA 25	29	R	*Pe*25	88.5	S
*Ps*BA 26	29	R	*Pe*26	88.5	S
*Ps*BA 27	29	R	*Pe*27	87.5	S
*Ps*BA 28	29	R	*Pe*28	87.5	S
*Ps*BA 29	29	R	*Pe*29	88.5	S
*Ps*BA 30	29	R	*Pe*30	88.5	S

Table 5 - AACPD values and classification of genotypes based on the visual evaluation of the symptoms caused by the CABMV virus.UENF, Campos dos Goytacazes-RJ, 2013.

Genotypes	Absorbance	ELISA[1]	Final Assessment[2]
*Ps*MG2	0.19	-	R
*Ps*MG8	0.16	-	R
*Ps*MG9	0.21	-	R
*Ps*MG11	0.16	-	R
*Ps*MG12	0.19	-	R
*Ps*MG13	0.19	-	R
*Ps*MG14	0.33	-	R
*Ps*MG18	0.21	-	R
*Ps*MG19	0.20	-	R
*Ps*MG24	0.20	-	R
*Ps*BA6	0.17	-	R
*Ps*BA 11	0.16	-	R
*Ps*BA 12	0.15	-	R
*Ps*BA 13	0.14	-	R
*Ps*BA 14	0.14	-	R
*Ps*BA 15	0.16	-	R
*Ps*BA 16	0.19	-	R
*Ps*BA 17	0.18	-	R
*Ps*BA 21	0.17	-	R
*Ps*BA 22	0.19	-	R
*Ps*RJ 3	0.20	-	R
*Ps*RJ 4	0.80	+	S
*Ps*RJ 5	019	-	R
*Ps*RJ 7	0.22	-	R
*Ps*RJ 10	0.13	-	R
*Ps*RJ 11	0.18	-	R
*Ps*RJ 13	0.15	-	R
*Ps*RJ 14	0.17	-	R
*Ps*RJ 17	0.16	-	R
*Ps*RJ 18	0.17	-	R
P. edulis (+)	1.54	+	S
P. edulis (-)	0.23	-	R
Negative Contro (buffer)	0.08	-	-

[1](+) = Positive reaction to the presence of the virus; (-) = negative reaction to the presence of the virus. [2]R = resistance; S = susceptibility.

Conclusions

Out of the 30 *P. setacea* genotypes inoculated with CABMV and submitted to the PTA-ELISA test, 29 were considered resistant. These genotypes can be eventuallyused in passion fruit genetic breeding programs.

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