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Evaluation of resistance to *Cowpea aphid-borne mosaic virus* in passion fruit backcrosses for recurrent selection and development of resistant cultivars

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ABSTRACT. The cultivation of passion fruit occupies an important place in Brazilian fruit culture; however, there have been successive declines in production. The main cause of this retraction in production is an increase in the incidence of *Cowpea aphid-borne mosaic virus*. This virus can cause severe deformation of the fruit and make passion fruit production inviable. Due to a lack of cultivars resistant to this virus, the Universidade Estadual do Norte Fluminense Darcy Ribeiro passion fruit breeding program has sought to develop resistant cultivars. The objective of this work was to evaluate the resistance to CABMV of a passion fruit second generation backcross (BC$_2$) segregating population via the REML/BLUP procedure aiming at selecting resistant genotypes with agronomic characteristics to start a recurrent selection program and possibly produce a new passion fruit cultivar. Virus resistance was measured by the average area under the disease progress curve using a score scale, evaluating the percentage of symptoms in the plant and young leaves. The total fruit mass produced per plant was used to assess fruit production. Based on this information, the components of variance and genetic value were estimated via REML/BLUP. The highest values estimated for genetic parameters, for the two characteristics evaluated, were

based on the average of each family. The BC_2 - 17 family showed the best additive genetic gain concerning resistance to CABMV and the worst for fruit production. The BC_2 - 293 family, had the highest estimated value of genetic gain for fruit production. The genetic variability found in the BC_2 segregating population allows us to select superior genotypes. Twenty-nine genotypes were selected to start the recurrent selection program aimed at resistance to CABMV. The - Two genotypes (BC_2 - 89 and BC_2 - 323) showed potential to be launched as CABMV resistant passion fruit cultivars.

Key words: Breeding tropical fruit; REML/BLUP; Viruses

INTRODUCTION

The cultivation of passion fruit occupies an important place in Brazilian fruit culture due to the diversified use of the fruit, its high added value, and its social importance (Meletti, 2011). According to IBGE data (2020), the production of sour passion fruit in 2019 was approximately 594 thousand metric tons. However, production has been decreasing since 2013, with a 28.95% decrease in 2019. This decline is caused by several factors, including phytosanitary problems and the lack of genotypes adapted to the growing regions (Silva et al., 2009; Cavalcante et al., 2019).

The main phytosanitary problem observed in passion fruit orchards is the virus of fruit woodiness (hardening) that is caused by the *Cowpea aphid-borne mosaic virus* (CABMV) (Sabião et al., 2011). This disease causes a reduction in the development of the plant, directly affecting the production and quality of the fruits, which become smaller, deformed, and hardened (Viana et al., 2014). The CABMV is transmitted by aphid vectors, by the grafting process, and by cultural pruning and thinning practices. The control of this virus in the producing areas is inefficient because its transmission is non-circulating and non-persistent; so chemical control of these vectors is not recommended (Gibbs and Ohshima, 2010).

The passion fruit breeding program of the Universidade Estadual do Norte Fluminense Darcy Ribeiro (UENF) seeks sources of resistance to CABMV, via the incorporation of resistance genes in a population of passion fruit from interspecific crosses between *Passiflora edulis* 'UENF Rio Dourado' (*P. edulis*) x *Passiflora setacea* (*P. setacea* - wild species) and generations of backcrosses (Santos et al., 2015; Santos et al., 2019b; Preisigke et al, 2020a and b), besides conducting studies on the inheritance of resistance (Freitas et al., 2015; Santos et al., 2019a). Studies related to the diversity of the genus *Passiflora* and breeding methods to identify genotypes with resistance to CABMV with productive potential are extremely important for the sustainability of this crop in Brazil. These studies should enable the release of resistant and adapted cultivars for the growing region, also promote the conservation and the best use of this available genetic resource.

In breeding work with passion fruit, it is necessary to use techniques that allow the prediction of the individual's genetic value through phenotypic characteristics (Ribeiro et al., 2019). The REML/BLUP method (restricted maximum likelihood/best linear unbiased prediction) supports this approach, since simultaneous fixed and random effects models allow excellent genetic gain and high selecting precision, especially in perennials (Assunção et al., 2015; Resende, 2016). They can also estimate the experimental error and the μ constant (Silva et al., 2017).

Due to the inefficiency in the control of CAMBV and its vector, the inexistence of cultivars resistant to CABMV and aiming to reduce losses in fruit production and quality, this study was aimed at evaluating resistance to CABMV of a second generation backcrossing (BC_2) population of passion fruit via REML/BLUP procedures. The objective was to select resistant genotypes with commercial agronomic characteristics to start a recurrent selection program and the possible development of a new passion fruit cultivar resistant to CABMV.

MATERIAL AND METHODS

Genealogy, acquisition, and management of the BC_2 segregating population

Figure 1 shows the stages of the passion fruit breeding program the Universidade Estadual do Norte Fluminense Darcy Ribeiro (UENF) aimed at genetic resistance to CABMV. One can observe the origin of the interspecific hybrids (IH) (*Passiflora edulis* x *Passiflora setacea*) and the first generation of backcrossing (BC_1) genotypes (selected IH x *Passiflora edulis*) in addition to the successive crosses and the analyses made in each selection cycle.

The second generation of backcrossing (BC_2) was obtained by crossing between first generation of backcrossing (BC_1) selected by Preisigke et al. (2020b) and *Passiflora edulis* UENF Rio Dourado (Viana et al., 2016). The crossings were made at the UENF experimental station located at Escola Agrícola Antônio Sarlo, in Campos dos Goytacazes, in the northern region of the state of Rio de Janeiro, between January and June 2017 (Table 1).

Table 1. Identification and origin of population second generation of backcrossing and their parents *Passiflora setacea*, *Passiflora edulis*, interspecific hybrids, and first generation of backcrossing (BC_1). Campos dos Goytacazes, RJ, 2018.

Description	Cross	Total number of genotypes
PS	BAG from UENF	8
PE	Cultivar UENF Rio Dourado	8
IH 5 - 14	PS 367 x PE 139 (40)	8
IH 1 - 15	PE 139 (40) x PS 367	8
BC_1 - 17	IH 5 - 14 x PE	8
BC_1 - 153	IH 5 - 14 x PE	8
BC_1 - 355	IH 1 - 15 x PE	8
BC_1 - 516	IH 1 - 15 x PE	8
BC_2 - 17	BC_1 - 17 x PE	100
BC_2 - 153	BC_1 - 153 x PE	92
BC_2 - 293	BC_1 - 293 x PE	80
BC_2 - 355	BC_1 - 355 x PE	146
BC_2 - 501	BC_1 - 501 x PE	126
BC_2 - 516	BC_1 - 516 x PE	100
Total		708

PE = *Passiflora edulis*; PS = *Passiflora setacea*; IH - interspecific hybrids selected in the study of Santos et al. (2015); BC_1 = First generation of backcrossing selected in the study of Preisigke et al. (2020b); BC_2 - Full-sib families the second backcrossing generation; Cultivar UENF Rio Dourado - passion fruit genetic breeding program by recurrent selection in the study of Viana et al. (2016).

For greater efficiency of the crosses, flowers in pre-anthesis were selected to be female parents (recipient parents), which were emasculated in the morning and protected with paper bags. The flowers selected as male parents (donor parents) were only protected with paper bags to avoid pollen contamination from other plants. From 12:00, the anthesis of the recipient parents, the pollen grains of the donor parents were

carefully rubbed manually in the stigma of the flowers of the recipient parents. After this procedure, the flowers were identified according to the cross performed and again protected with paper bags for 24 hours to check the pollination.
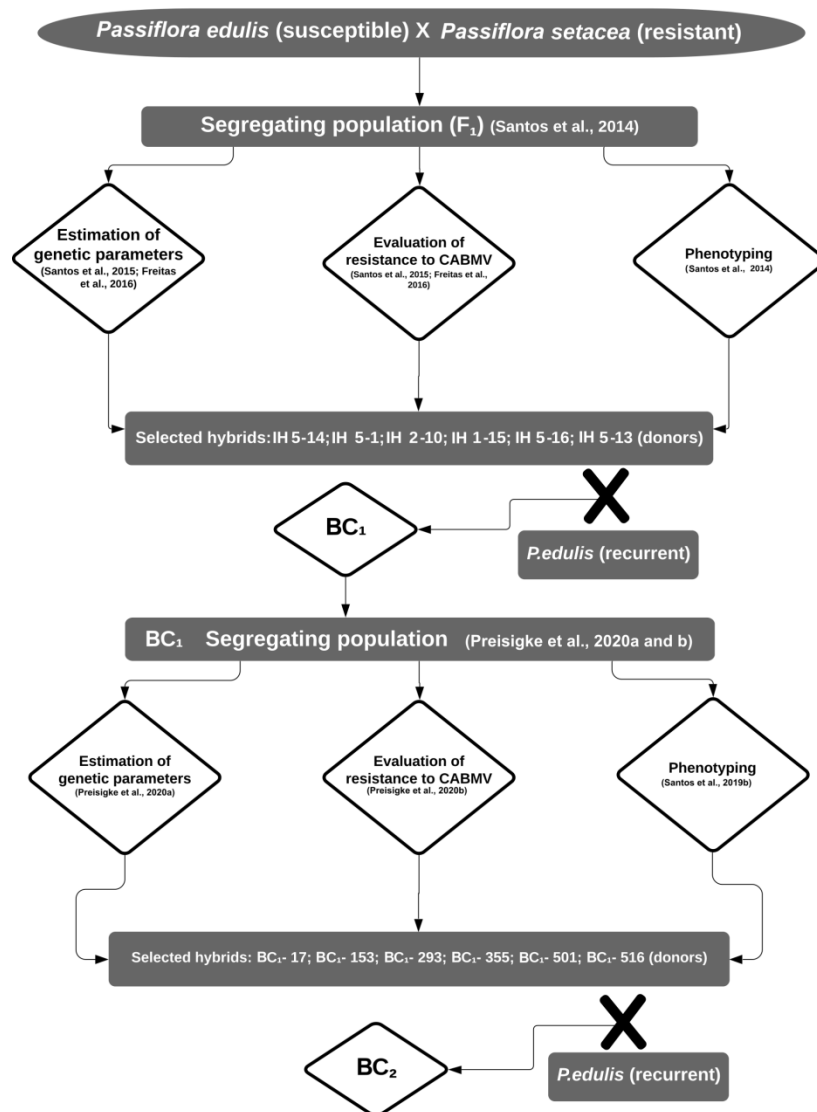

Figure 1. Flowchart of the steps of the passion fruit breeding program aimed at resistance to *Cowpea aphid-borne mosaic virus* of the Universidade Estadual do Norte Fluminense Darcy Ribeiro. Campos dos Goytacazes, RJ, 2020. F1 - first generation; IH - interspecific hybrids; BC_1 - first generation of backcrossing; BC_2 - full-sib families the second backcrossing generation; *P. edulis = Passiflora edulis* Cultivar UENF Rio Dourado; *P. setacea = Passiflora setacea*.

The fruits from the crosses were protected with a nylon net until their complete ripening. Then, the harvested fruits were taken to the laboratory for the removal,

washing and drying of the seeds. The drying process of the seeds was done at room temperature. After complete drying, seeds were stored in paper bags lined with plastic bags and kept in a refrigerator at +10°C in the LMGV of UENF until sowing.

BC_2 seeds were sown in Styrofoam trays of 200 cells containing commercial substrate in a greenhouse in September 2017. After emergence of two pairs of definitive leaves, the seedlings were transferred individually to plastic polyethylene bags containing commercial substrate. In January 2018, the seedlings were planted in the experimental area of the UENF located at the Escola Agrícola Antônio Sarlo with AW climate - tropical with dry winter (Köppen and Geiger, 1928) and sandy-clay texture soil.

The experimental design used was a randomized block with six replications, unbalanced for plants within families composed of six full-sib families of the BC_2, their parents - *P. setacea* (resistant parent), *P. edulis* Cultivar UENF Rio Dourado (recurrent and susceptible parent), two interspecific hybrids (IH) and four full-sib genotypes of the BC_1, totaling 708 individuals (Table 1). All *P. edulis* plants used in the experiment were obtained from seeds of Cultivar UENF Rio Dourado (Viana et al., 2016). The plants of the interspecific hybrids and the first generation of backcross were obtained from cuttings and kept in a greenhouse until planting.

A vertical trellis system with posts 2.5 m high, spaced at 4.0 m, and no. 12 wire at 1.80 m from the ground was used to conduct the plants in the field. The distance between planting rows was 3.5 m, and crop management was carried out according to the requirement of the crop. In the fertilization, 200 g of lime, 400 g of simple superphosphate and 250 g of potassium chloride per planting hole were used considering the soil analysis carried out by FunderNor. In the monthly fertilization, 80g of potassium chloride and 100g of urea was used. A drip irrigation system was used to meet the water needs of the crop. (Faleiro et al., 2016).

Evaluation of resistance to CABMV, fruit production and estimates of genetic parameters

The experimental area has a history of the natural occurrence of the virus of the virus and the aphid responsible for the transmission of the disease. Consequently, the virus was not inoculated into the plants and the evaluations were carried out based on the natural occurrence of CABMV. The pruning equipment used during the conduction of the plants was not sanitized to rule out the escape effect of the virus so that the transmission of the virus occurred in all genotypes under study, thus, ensuring that all plants had contact with the virus.

The first symptoms of CABMV appeared 50 days after the implantation of the experiment (March 2018); thereafter evaluations for the severity of the virus started and were performed every fortnight during 18 months. The first symptoms of the virus in *P. edulis* plants appeared at 60 days, and by 240 days, all genotypes showed symptoms of the disease. The first flowers and flower buds were observed 50 days after the implementation of the experiment, with the first fruits harvested in June 2018.

The evaluations regarding the severity of the virus in the genotypes were carried out visually for the young leaves, and the percentage of symptoms in the plant was evaluated using the diagrammatic scale of scores adapted by Oliveira et al. (2013) (Table 2). In Figure 2, we can see photos of the symptoms of CABMV in young leaves and the percentage of symptoms in the plant at each disease severity level.

Based on these data, the area under the disease progress curve (AUDPC) was estimated for young leaves and the percentage of symptoms in the plant, by the following estimator:

$$AUDCP = \sum_{i=1}^{n-1} \frac{Y_i + Y_{i+1}}{2}(T_{i+1} + T_I) \qquad \text{(Eq. 1)}$$

Where: Yi = proportion of the disease in the i-th observation; Ti = time in days of the i-th observation; n = number of observations (Campbell and Madden, 1990). After estimating the AUDPC's for young leaves and the percentage of symptoms in the plant, a simple arithmetic mean was performed between them to obtain the average area under the disease progress curve (AAUDPC) according to Preisigke et al. (2020b). About fruit production (FP), all the fruits produced by each genotype from June 2018 to July 2019 (harvest 2018/2019) were weighed on a precision scale.

Table 2. Scale of scores used for the evaluation of symptoms induced by *Cowpea aphid-borne mosaic virus* in sour passion fruit, adapted from Oliveira et al. (2013). Campos dos Goytacazes, RJ, 2019.

| Score	Visual symptoms	
	Young leaves	**Plant**
1	Leaf without mosaic symptoms	Plant without symptoms
2	Mild mosaic, node formation of leaves	Moderate quantity of leaves with mild mosaic and wrinkling
3	Mild mosaic, blisters, and deformation of leaves	Moderate quantity of leaves with mosaic and leaf wrinkling
4	Severe mosaic, blisters, and deformation of leaves	Large quantity of leaves with mosaic, severe wrinkling and leaf deformation
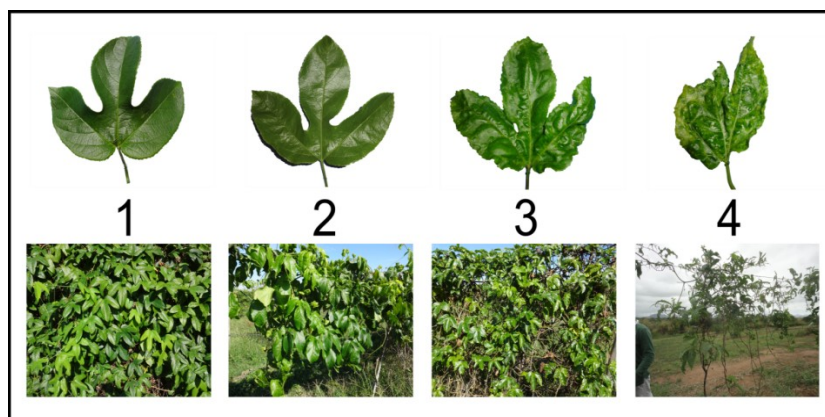
Figure 2. Images of the scale for assessing *Cowpea aphid-borne mosaic virus* symptoms in young leaves of passion fruit and percentage of symptoms in the plant. Campos dos Goytacazes, RJ, 2020. Photo: Sandra Preisigke.

The genetic parameters were estimated by REML and the individual additive genetic effects by BLUP for the two characteristics evaluated (resistance to CABMV and FP). Data were analyzed using the Selegen REML/BLUP statistical software, model 147 (Resende, 2016). The analysis followed the statistical model y= Xr+Zg+Wp+e, where *y* is the vector of data, *r* is the vector of the replicate effects (assumed to be fixed) added to the overall mean, *g* is the vector of the genotypic effects individual (assumed to be random), *p* is the vector of plot effects (random), and *e* is the vector of errors or residuals (random). The capital letters represent the incidence matrices for the said effects. The following components of variance (REML) were estimated: σ^2_g: genotypic variance between full-sibs progenies, equivalent to 1/2 of the additive genetic variance plus 1/4 of the dominance genetic variance, ignoring epistasis; σ^2_{plot}: environmental variance between plots; σ^2_{within}: residual variance within the plot; σ^2_f: individual phenotypic variance; h^2_a: individual narrow-sense heritability, obtained by ignoring the fraction (1/4) of the genetic dominance variance; h^2_{mp}: mean heritability of progenies, assuming complete survival; h^2_{ad}: additive heritability within the plot, obtained by ignoring the fraction (1/4) of the genetic variance of dominance; c^2_{plot}: coefficient of determination of plot effects; Acprog: progeny selection accuracy, assuming complete survival; and General average.

Ranking and selection of genotypes

The ranking of the 30 best genotypes for the characteristic resistance to CABMV, we considered a cutoff for AACPDM from 620.00 to 699.00 since for the disease characteristic it is necessary to select the genotypes with the lowest values of AACPDM. In addition to this criterion, only individuals who produced fruit during the evaluation period were considered in the evaluation. For the ranking of the 30 genotypes with the highest fruit production, the sequence established by the BLUP's was considered.

For the selection of the 29 best genotypes to continue the passion fruit program aiming at resistance to CABMV, four criteria were used. This was done to maintain the genetic diversity of future generations without narrowing the genetic base to guarantee the success and continuity of the passion fruit breeding program. In addition, for fruit production criteria, we considered the selection of at least one genotype from each evaluated family, increasing the cut-off grade to 799.00 and based on the breeder's experience in the field.

RESULTS

Estimates of genetic parameters

The estimates of genetic parameters for the characteristics of resistance to CABMV and fruit production can be seen in Table 3. For the AAUDPC variable, the lowest estimated values were found for the genotypic variance (σ^2_g), individual narrow-sense heritability (h^2_a), additive heritability within the plot (h^2_{ad}) and coefficient of determination of plot effects (c^2_{plot}). The highest estimated values were observed for

progeny selection accuracy (Acprog) and heritability of the average of progenies (h^2_{mp}). The estimate of the genotypic variance (σ^2_g) for AAUDPC was low, with only 1.30% of phenotypic variance (σ^2_f). Concerning accuracy, 75% of the predicted values were explained by the model.

The estimated values for genotypic variance (σ^2_g) and the coefficient of determination of plot effects (c^2_{plot}) were the lowest for the FP variable. The highest estimate values were for individual narrow-sense heritability (h^2_a), heritability of the average of progenies (h^2_{mp}), additive heritability within a plot (h^2_{ad}), and accuracy (Acprog). The estimate of the genotypic variance (σ^2_g) for FP was 27.83% of the phenotypic variance (σ^2_f) with an accuracy of 98%, which explains the predicted values.

Table 3. Estimates of genetic parameters via REML procedure, for resistance to *Cowpea aphid-borne mosaic virus* based on the average area under the disease progress curve (AAUDPC) and for fruit production (FP) in passion fruit genotypes from the second generation of a backcrossing segregating population. Campos dos Goytacazes, RJ, 2019.

Genetic Parameters	Characteristics	
	AAUDPC	FP
σ^2_g	89.24	2.46
σ^2_{plot}	191.02	0.34
σ^2_{within}	6582.15	6.04
σ^2_f	6862.42	8.84
h^2_a	0.03	0.56
h^2_{mp}	0.56	0.96
h^2_{ad}	0.01	0.41
C^2_{plot}	0.03	0.04
Acprog	0.75	0.98
General average	774.49	2.65

(σ^2_g): genotypic variance between full-sib progenies; (σ^2_{plot}): environmental variance between plots; (σ^2_{within}): residual variance within plot; (σ^2_f): individual phenotypic variance; (h^2_a): individual narrow-sense heritability; (h^2_{mp}): mean heritability of progenies; (h^2_{ad}): additive heritability within plot; (C^2_{plot}): coefficient of determination of plot effects; (Acprog): progeny selection accuracy.

Ranking of families and genotypes

The ranking of the families that were evaluated is shown in Table 4. The BC2 - 17 family stood out, with the best family estimates for genetic gain in relation to resistance to CABMV and the lowest for fruit production. The BC_2 - 293 family, had the highest estimated value of genetic gain for FP and the third best for resistance. The predicted genetic gain ranged from 0% to 7.65% for the AAUDPC trait and from 2.44% to 0.00% for FP.

In the ranking of the 30 best genotypes for the AACPDM characteristic, only genotypes from the families BC_2 - 17 (40%), BC_2 - 153 (40%), BC_2 - 293 (10%) and BC_2 - 501 (10%) were ranked with the lowest predicted estimates for resistance (Table 5). Genetic values ranged from -7.28 to -3.89 and gains from 0.60 to 1.92%.

The families BC_2 - 153 (1 individuals), BC_2 - 293 (19 individuals), BC_2 - 355 (9 individuals) and BC_2 - 501 (1 individual) participated in the ranking of the 30 best genotypes with the highest predicted FP estimates (Table 6). The estimated values for the genetic effect ranged from 7.54 to 2.97 and the estimated genetic gains from 7.54 to 4.06%.

Table 4. Ranking of the six passion fruit families from second generation of backcrossing for predicted estimates of additive genetic value, genetic gain, and new average, estimated via REML/BLUP for average area under the disease progress curve and fruit production. Campos dos Goytacazes, RJ, 2019.

Family	Additive genetic value	Genetic gain	New average
	AAUDPC		
17	-5.62	0	774.49
153	-3.83	1.12	775.62
293	-2.46	2.36	776.86
501	-2.38	3.97	778.47
355	6.65	7.15	781.64
516	7.65	7.65	782.15
Family	**FP**		
293	2.44	2.44	5.09
355	1.14	1.79	4.44
501	-0.07	1.17	3.82
153	-1.01	0.62	3.28
516	-1.16	0.27	2.92
17	-1.33	0	2.65

AAUDPC: average area under the disease progress curve; FP: fruit production (kg).

Table 5. Ranking of the 30 passion fruit genotypes from the second generation of backcrossing with the lowest predicted estimates for additive genetic value, genetic gain, and new mean, estimated via REML/BLUP for average area under the disease progress curve. Campos dos Goytacazes, RJ, 2019.

Rank	Genotype (family)	AAUDPC			
		Phenotypic value	Genetic value	Gain (%)	New adjusted average
1	433 (17)	622.50	-7.28	0.60	626.23
2	431 (17)	645.00	-6.97	0.62	648.99
3	616 (17)	667.50	-6.77	0.64	671.75
4	138 (17)	686.25	-6.69	0.66	690.75
5	619 (17)	675.00	-6.67	0.67	679.55
6	142 (17)	690.00	-6.64	0.69	694.77
7	133 (17)	693.75	-6.59	0.71	698.68
8	419 (17)	678.75	-6.52	0.73	683.69
9	307 (17)	686.25	-6.49	0.75	691.37
10	422 (17)	682.50	-6.47	0.76	687.72
11	301 (17)	697.50	-6.34	0.78	702.96
12	418 (17)	693.75	-6.31	0.80	699.30
13	410 (153)	660.00	-5.04	1.21	667.96
14	539 (153)	663.75	-5.03	1.22	671.87
15	223 (153)	675.00	-4.99	1.24	683.37
16	120 (153)	686.25	-4.87	1.29	695.11
17	393 (153)	675.00	-4.84	1.31	683.83
18	103 (153)	693.75	-4.77	1.32	702.94
19	100 (153)	693.75	-4.77	1.34	703.05
20	108 (153)	697.50	-4.72	1.36	706.97
21	237 (153)	697.50	-4.68	1.37	707.09
22	221 (153)	697.50	-4.68	1.39	707.20
23	392 (153)	693.75	-4.58	1.41	703.52
24	399 (153)	697.50	-4.53	1.42	707.44
25	89 (501)	630.00	-4.29	1.63	640.24
26	577 (501)	600.00	-4.29	1.64	609.86
27	323 (293)	637.50	-4.12	1.80	648.96
28	376 (501)	633.75	-4.03	1.83	645.37
29	324 (293)	652.50	-3.92	1.90	664.92
30	156 (293)	652.50	-3.89	1.92	665.03

Phenotypic value: intensity of damage to the plant caused by *Cowpea aphid-borne mosaic virus* (CABMV) at the end of the evaluations; New adjusted average expected average of the next generation in relation to the intensity of damage to the plant caused by CABMV; AAUDPC: average area under the disease progress curve.

Table 6. Ranking of the 30 passion fruit genotypes from second generation of backcrossing with the highest predicted estimates for additive genetic value, genetic gain, and new mean, estimated via REML/BLUP for fruit production. Campos dos Goytacazes, RJ, 2019.

Rank	Genotype (family)	Fruit production			
		Phenotypic value	Genetic value	Gain (%)	New adjusted average
1	157 (293)	17.62	7.54	7.54	18.95
2	583 (355)	17.16	6.46	7.00	18.36
3	692 (355)	16.62	5.83	6.61	17.72
4	632 (293)	12.33	5.77	6.40	13.12
5	548 (355)	14.12	5.22	6.17	14.99
6	323 (293)	11.48	4.81	5.94	12.16
7	334 (293)	11.26	4.72	5.77	11.91
8	482 (293)	9.56	4.52	5.61	10.10
9	246 (501)	13.50	4.35	5.47	14.24
10	214 (355)	11.15	4.24	5.35	11.75
11	496 (293)	8.69	4.16	5.24	9.15
12	156 (293)	9.05	4.04	5.14	9.52
13	154 (293)	8.95	4.00	5.05	9.40
14	162 (293)	8.53	3.83	4.97	8.95
15	501 (293)	7.70	3.76	4.88	8.08
16	483 (293)	7.41	3.64	4.81	7.77
17	643 (293)	6.76	3.50	4.73	7.08
18	545 (355)	9.67	3.41	4.66	10.12
19	678 (355)	9.52	3.37	4.59	9.96
20	332 (293)	7.86	3.34	4.53	8.22
21	159 (293)	7.28	3.32	4.47	7.61
22	441 (355)	8.84	3.30	4.42	9.23
23	315 (293)	7.72	3.28	4.37	8.06
24	322 (293)	7.66	3.26	4.32	7.99
25	495 (293)	6.26	3.17	4.27	6.53
26	319 (293)	7.04	3.00	4.23	7.34
27	45 (355)	7.12	2.99	4.18	7.42
28	595 (355)	8.63	2.98	4.14	8.99
29	328 (293)	6.99	2.98	4.10	7.28
30	238 (153)	11.53	2.97	4.06	12.00

Phenotypic value: fruit production per plant (Kg); New average: expected average of the next generation in relation to fruit plant production (Kg)

Genotype selection

Table 7 shows the 29 genotypes selected to continue the passion fruit breeding program aiming to obtain resistance to CABMV. Among these genotypes, only six (89, 103, 138, 156, 323 and 376) are present among the 30 best for AAUDPC and nine genotypes (156, 162, 238, 323, 332, 441, 483, 495 and 583) among the 30 best for FP. Only genotypes 156 and 323 were ranked high for the two traits. Genotype 89 stood out for resistance to CABMV and genotype 323 for FP.

Table 7. Genotypes selected to continue the passion fruit breeding program aimed at resistance to *Cowpea aphid-borne mosaic virus* at the Darcy Ribeiro State University of Northern Rio de Janeiro based on average area under the disease progress curve and fruit production. Campos dos Goytacazes, RJ, 2019.

Rank	Genotype (family)	AAUDPC				Genotype (family)	Fruit production			
		Phenotypic value	Genetic value	Gain	New average		Phenotypic value	Genetic value	Gain	New average
1	138	686.25	-6.69	0.66	690.75	583	17.16	6.46	7.00	18.36
2	103	693.75	-4.77	1.32	702.94	323	11.48	4.81	5.94	12.16
3	89	630	-4.29	1.63	640.24	156	9.05	4.04	5.14	9.52
4	238	727.5	-4.28	1.66	739.58	162	8.53	3.83	4.97	8.95
5	323	637.5	-4.12	1.80	648.96	483	7.41	3.64	4.81	7.77
6	376	633.75	-4.03	1.83	645.37	332	7.86	3.34	4.53	8.22
7	156	652.5	-3.89	1.92	665.03	441	8.84	3.30	4.42	9.23
8	391	746.25	-3.87	1.96	760.85	495	6.26	3.17	4.27	6.53
9	325	675	-3.62	2.13	689.41	238	11.53	2.97	4.06	12.00
10	332	682.5	-3.51	2.26	697.94	42	6.76	2.85	3.92	7.02
11	95	690	-3.48	2.30	705.86	166	6.00	2.80	3.80	6.23
12	74	690	-3.48	2.32	705.99	325	6.40	2.74	3.75	6.64
13	565	660	-3.47	2.36	675.55	571	7.46	2.04	3.29	7.71
14	166	686.25	-3.43	2.43	702.94	95	6.01	1.45	2.73	6.17
15	377	682.5	-3.37	2.55	699.90	74	5.25	1.14	2.49	5.38
16	495	701.25	-3.35	2.57	719.27	281	3.41	1.09	2.43	3.49
17	572	671.25	-3.32	2.59	688.63	593	3.60	0.93	2.34	3.68
18	571	682.5	-3.17	2.90	702.30	627	0.32	0.88	2.30	0.33
19	627	705	-3.05	3.08	726.69	556	4.00	0.63	2.10	4.08
20	556	735	-2.46	4.26	766.35	89	3.79	0.54	2.00	3.87
21	162	765	-2.36	4.45	799.02	565	2.78	0.13	1.70	2.83
22	551	780	-1.85	5.39	822.06	138	4.21	0.02	1.61	4.28
23	281	667.5	5.02	7.00	714.20	572	2.38	-0.03	1.58	2.42
24	583	641.25	5.06	7.03	686.36	551	1.76	-0.28	1.38	1.78
25	42	663.75	5.06	7.05	710.53	54	2.77	-0.45	1.26	2.80
26	593	660	5.31	7.12	706.96	376	0.74	-0.61	1.15	0.75
27	441	731.25	6.17	7.52	786.24	103	2.00	-0.69	1.12	2.02
28	483	697.5	6.56	7.77	751.68	377	0.37	-0.76	1.06	0.37
29	54	757.5	7.23	8.47	821.68	391	0.81	-1.07	0.88	0.82

Phenotypic value: intensity of damage to the plant caused by CABMV at the end of the evaluations; New adjusted average expected average of the next generation in relation to the intensity of damage to the plant caused by CABMV; AAUDPC: average area under the disease progress curve; Phenotypic value: fruit production per plant (Kg); New average: expected average of the next generation in relation to fruit plant production (Kg).

DISCUSSION

Estimates of genetic parameters

Estimates of genetic parameters are effective in breeding programs, as they assist in the indication of superior genotypes, in the selection of parents for crosses and the prediction of genetic gains (Cruz et al., 2014). This facilitates decision making in future stages of the breeding program and helps plan more efficient breeding strategies, selection of genotypes and maintenance of the gains and genetic variability of the populations under study.

The estimates of the genotypic variances ($\sigma_2 g$) were low for both AAUDPC and FP, showing that the environmental effects had more effective participation in the estimated values than the genotypic effects (Table 3). Environmental variation is not favorable to genetic improvement and should be minimized to increase the precision of estimated values through strategies that aim to reduce their impacts.

These interactions of genotypes with the environment directly influence heritability estimates (Ferreira et al., 2016) as well as the type of genetic inheritance that controls the trait. As the inheritance that controls resistance to CABMV is polygenic and highly influenced by the environment (Freitas et al., 2015), this may explain the low values predicted for genetic variance found in this study.

Although interspecific crosses can be used in a breeding method aiming at introgression of characteristics controlled by one or a few genes (Freitas et al., 2015), the Passion Fruit Breeding Program of UENF used this method followed by backcrossing to incorporate various inheritance genes. Since resistance to CABMV involves a complex of inheritance genes, identifying CABMV-resistant genotypes is difficult for breeders.

The estimated values of heritability for AAUDPC and FP showed that the best form of selection for these characteristics is based on the family (h^2_{mp}), since heritability is involves the heritable portion of phenotypic variance (Ramalho et al., 2012a). Therefore, the selection of families is recommended, as they will provide more significant genetic gains and more accurate selection for the two characteristics that we evaluated.

However, it should be noted that for the FP characteristic, individual selection and selection within the family is also viable, as the estimated values for individual narrow-sense heritability and additive heritability within the family were above 40%. Ferreira et al. (2016) evaluated FP of passion fruit progenies of the second cycle of recurrent selection, and they obtained a lower result for individual narrow-sense heritability and higher for additive heritability within the family than what we found here. This fact may mean that these populations are still very segregating, and that mixed modeling is effective for genotype selection through characteristics of low heritability. Santos et al. (2015) also explained that based on the predicted genetic gains values, superior parents can be selected for the following generations of the breeding program.

The observed accuracy for AAUDPC and FP were classified as high, indicating the reliability of the predicted values concerning the real values. In the selection process in breeding programs, accuracy values above 70% are sought. Preisigke et al. (2020a) found an accuracy value for FP classified as low. Thus, it can be concluded that the high precision of the predicted genetic values aids in the more reliable selection of superior genotypes.

Ranking of families and genotypes

The ranking of families showed the BC_2 - 17 family as the most resistant and least productive (Table 4). This is an indication that the more resistant the genotype is, the more it concentrates blocks of alleles of the genotypes that are genetically closer to *P. setacea*. Thus, they have fewer characteristics of the cultivar Rio Dourado. This fact shows that the UENF Passion Fruit Breeding Program aiming at resistance to CABMV must opt for new breeding methods to rescue these desirable characteristics of commercial cultivars. The recurrent selection proves to be a promising method for selecting resistant passion fruit genotypes with desirable agronomic traits. According to Ramalho et al. (2012b), recurrent selection is an efficient method to increase the performance of the population to be improved through the accumulation of favorable alleles, maintaining genetic variability, and thus allowing genetic gains in the following cycles.

The BC_2 - 293 family showed better performance for the predicted estimates for FP and with an additive genetic value of -2.46 concerning resistance to CABMV, being

favorable for the selection of superior parents within this family (Table 4). According to Freitas et al. (2015), nonspecific crosses involving *P. edulis* and *P. setacea*, followed by backcrossing with *P. edulis*, originate a segregating population with resistant individuals with agronomic characteristics similar to those of commercial species.

Although the selection had to be made based on the additive genetic values, the ranking of the 30 best genotypes for the AAUDPC trait considered the fruit production and maximum AAUDPC score of 699 (Table 5). The use of these criteria in the AAUDPC ranking occurred because, for the formation of a base population of a breeding program, the parents involved should have high performance for the characteristics to be selected (Hallauer et al., 2010).

The average genetic increments predicted for AAUDPC were low but desirable, because when it comes to diseases, the smaller the gains, the better the genotypes. Also, as the genetic nature of CABMV resistance is quantitative, linked to several genes, there is the possibility of indicating genotypes with different levels of resistance. Therefore, evaluating the individual genetic gains of the genotypes is essential for the good performance of the different stages of a breeding program.

Most genotypes had low FP, but genotypes 157, 583, and 692 stood out with FP above 15 kg plant^{-1} (Table 6). The genetic gains of the 30 genotypes with higher yields were low when compared to the results found by Ferreira et al. (2016), and Silva et al. (2017) in a passion fruit population originated from recurrent selection. According to Meletti and Brückner (2001), interspecific crossings cause some problems, such as male sterility, low pollen viability, and low flower emission, causing low fruit yield in segregating populations of backcrossing. Also, weobserved a high rate of abortion of flowers and poor fruit formation

Genotype selection

The four criteria used to select the parents of future targeted crosses were aimed at maintaining maximum genetic variability, preventing the narrowing of the genetic basis and the genetic erosion of future populations in the program. Also, it is essential that in the selection process, the observations made by the breeder on the performance of the genotypes during the evaluation period of the experiment be considered.

Most parents selected to continue in the program are not among the top 30 ranked for both AAUDPC and FP (Table 7). The breeder should plan the best selection strategy to ensure significant genetic gain and select efficient parents. According to Borém and Miranda (2013), the ability to choose the genotypes that will be the parents of future generations of crossbreeding is decisive for the success in the genetic improvement of plants. A strategy for making selection based on fruit characteristics is the simultaneous selection of traits through a selection index. The objective of this strategy is to choose superior genotypes, based on the set of variables that accumulate the maximum characteristics of interest to the breeder, resulting in the best simultaneous gains (Cruz et al., 2014).

It is crucial to choose individuals with good agronomic characteristics for the development of new cultivars with desirable traits. (Borém and Miranda, 2013). Therefore, genotypes 89 and 323 can be indicated as candidate cultivars resistant to CABMV with acceptable agronomic characteristics.

CONCLUSIONS

The genetic variability existing in the BC_2 segregating population allows us to select superior genotypes through the characteristics of resistance to CABMV and FP. The genotypes 42, 54, 74, 89, 95, 103, 138, 156, 162, 166, 238, 281, 323, 325, 332, 376, 377, 391, 441, 483, 495, 551, 556, 565, 593, 571, 572, 583 and 627 are indicated to start the recurrent selection program aiming at resistance to CABMV. Genotypes 89 and 323 have the potential to be launched as new CABMV resistant passion fruit cultivars adapted to the Northern Fluminense region of the state of Rio de Janeiro.

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CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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