INHERITANCE OF PAPAYA RINGSPOT VIRUS-W STRAIN (WATERMELON MOSAIC VIRUS-1) RESISTANCE IN THE INTERSPECIFIC HYBRID BETWEEN *Cucurbita ecuadorensis* x C. *maxima*

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ABSTRACT

Resistance of interspecific hybrids between *C. ecuadorensis* and *C. maxima* cv. Kurokawa Delicious against the papaya ringspot virus was tested by greenhouse planting with artificial inoculation of young seedlings. PRSV-w resistance in *C. ecuadorensis* was considered to be controlled by a single dominant gene.

Introduction

Papaya ringspot virus-w disease which was previously called Watermelon Mosaic Virus-1 (WMV-1) disease is one of the most important diseases of commercial Cucurbitaceous crops cultivated in Brazil (Avila, 1982). The occurrence of this disease in Brazil was first reported in 1972 in the region of Belem in Para State (Albuquerque, 1972) which located in the northeastern part of Brazil, followed by São Paulo, Piaui and Rio Grande do Norte States in 1982, (Cost, 1972 ; Lima, 1980) and in the region of Rio São Francisco Valley in 1982 (Chioudhoury, 1982). It is considered that the use of resistant varieties is the most appropriate measure to control the disease.

Maluf *et al.* (1986) tested the resistance against the disease of thirty lines/varieties which belong to four kinds of species, and found that *C. ecuadorensis* was highly resistant. Igarashi *et al.* (1986) also showed that *C. ecuadorensis* was most resistant against several viruses such as cucumber mosaic, watermelon mosaic and zucchini yellow mosaic viruses among twenty six materials belonging to eleven different Cucurbitaceous species. Meanwhile, Thomas suggested the possibility of interspeciffic crossing with *C. maxima* and the use of this hybrid as breeding material. This study was, therefore, initiated to analyse the mode of inheritance of resistance of *C. ecuadorensis* against PRSV-w in order to obtain breeding materials for resistance to the virus disease.

Materials and methods

1 Materials

The materials shown in Table 1 such as F_1, F_2, and BC_1P_2 which were developed from an interspecific cross between *C. ecuadorensis* as resistant parent (P_1), and *C. maxima* cv. Kurokawa Delicious as susceptible parent (P_2) were used in this study.

The first interspecific crossing was carried out in May 1988 under greenhouse conditions, and the seeds of the F_1 generation were sown in August 1988, but, did not germinate well (10% only) despite the use of mature seeds. However, seeds which easily germinated showed an extremely vigorous growth (twenty liter earthen pots in the greenhouse for

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Table 1 Tested materials and number of tested plants

Materials	Symbols	No. of tested plants
C. ecuadorensis	P_1	20
C. maxima cv. Kurokawa Delicious	P_2	20
$P_1 \times P_2$	F_1	21
$F_1 \times F_1$	F_2	39
$F_1 \times P_2$	BC_1P_2	34

more than one year) and bore twenty seven fruits from two plants which were to be used as F_2 and BC_1P_2 materials. Therefore, the F_1 materials used in this experiment were propagated by cuttings in using a medium mixed with 50% of vermiculite and 50% of carbonized rice chaff.

2 Methods

1) Experimental schedule

The varieties shown in Table 1 were sown in boxes filled up with a medium consisting of a mixture of carbonized rice chaff, subsoil and fully fermented compost at a rate of 1 : 2 : 1 on 4 April 1989 and, 5 days after, transplanted in 9 cm diameter plastic pots placed on a bench in the greenhouse. Therefore the whole experiment was carried out under greenhouse conditions with a temperature ranging between 32°C in the day time and 24°C during the night.

Cutting propagation of F_1 scions (25/March, 1989)
↓
Sowing P_1, P_2, F_2 and BC_1P_2 (4/april)
↓
Transplanting all the materials to pots (9/April)
↓
lst inoculation (11/April)
↓
2nd inoculation (14/April)
↓
3rd inoculation (18/April)
↓
Visual evaluation (10/May)
↓
ELISA test (13/May)

2) Inoculum preparation

PRSV-w isolated at Anápolis in Goias State was mechanically inoculated to *C. pepo* cv. Caserta plants at the cotyledon leaf stage. The plants were grown under greenhouse conditions from 15 to 20 days after being used as an inoculum source.

3) Method of inoculation

"Caserta" leaves showing severe mosaic symptoms were triturated at the rate of 1 : 10 (weight volume) in 0.02 M phosphate buffer, pH 7.0 with 0.1% of sodium sulfite. The plants were dusted with carborundum before inoculation. The tested materials were first

Symptoms	Grade value	
Mosaic	+	Severe systemic mosaic
	−	No symptoms
Chlorotic spots	+	Systemically spreading
	±	Few chlorotic spots scattered on the upper leaves inoculated
	−	No symptoms

inoculated to the first unfolded leaves and re-inoculated twice to the second and third leaves at four day intervals.

4) Evaluation

The symptoms of the tested plants were visually evaluated one month after the first inoculation. The plants that exhibited doubtful symptoms were tested by indirect ELISA for the presence of PRSV-w infection. The classification of symptoms and susceptibility index based on visual evaluation is shown on Table 2.

Results and discussion

The results of the inoculation test on the F_1, F_2 and BC_1P_2 of the interspecific hybrid between *C. ecuadorensis* and *C. maxima* are presented in Table 2. None of the plants of *C. ecuadorensis* used as resistant parent showed any symptoms and they grew well. On the contrary, *C. maxima* cv. Kurokawa Delicious used as susceptible parent showed severe systemic mosaic symptoms.

As shown in Table 2 in the case of the F_1 plants, most of the plants showed a sound growth, two inoculatedplants showed faint chlorotic spots on the upper leaves and PRSV -w was not detected by ELISA.

Segregations of susceptible and resistant individuals in the F_2 and BC_1P_2 were 29 : 10 (for 3 : 1 $50<P<70$) and 18 : 16 (for 1 : 1 $70<P<80$) respectively, and agreed with the ratio expected from single dominant gene. Therefore, PRSV-w resistance derived from *C. ecuadorensis* was estimated to be controlled by a single dominant gene based on the segregation ratios in the F_1, F_2 and BC_1P_2 generations. *C. ecuadorensis*, which has been known to be a promising material for breeding for resistance to several major viruses such as cucumber mosaic (CMV), tomato ringspot (TRSV), papaya ringspot (PRSV) (Thomas and Robiujm) and zucchini yellow mosaic (ZYMV) viruses (Maluf *et al.*, 1986), was able to hybridize with the cultivar *C. maxima* which is hardly resistant to those viruses. Fertility as well as germinability of the F_2 and BC_1P_2 generations was similar to those of other *C. maxima* cultivars. Moreover distinct segregations of flesh colour and reduction of bitterness were observed in the F_2 and BC_1P_2 populations. Therefore, it is considered that it may be possible to utilize these interspecific progenies as breeding materials to develop *C. maxima* varieties resistant to virus diseases and/or use them as bridge materials to hybridize with other species like *C. moschata* in order to develop a variety like 'Tetsukabuto' in future.

Table 2 Reactions of progenies of interspecific hybrid crossed with *C. ecuadorensis* and *C. maxima* to inoculation of PRSV-w using carborundum rubbing method

	Visual evaluation								
Symbols	Mosaic		Chlorotic S			No. of plants	Chi square value probability		
	(+)	(−)	(+)	(±)	(−)				
P_1	0	0	0	0	0	20			
P_2	20	0	0	0	0	20			
F_1	0	0	0	2	19	21			
F_2	5	0	6	0	28	39	3 : 1	0.213	$50<P<70$
BC_1P_2	18	0	0	16	0	34	1 : 1	0.116	$70<P<80$
	ELISA evaluation								
	Positive(+)		Negative(−)						
F_1	0		0						
F_2	10		29			39	3 : 1	0.281	$50<P<70$
BC_1P_2	19		15			34	1 : 1	0.470	$50<P<70$

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