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Effects of resistance genes, heat tolerance genes and cytoplasms on expression of resistance to *Pseudomonas solanacearum* (E.F. Smith) in potato

Pham X. Tung[1], John G.Th. Hermsen[2], Peter Vander Zaag[3] & Peter Schmiediche[4]
[1] Potato Research Station, Thaiphien, Dalat, Vietnam; [2] Department of Plant Breeding, Wageningen Agricultural University (WAU), The Netherlands; [3] International Potato Center (CIP), Box 933, Manila, The Phillippines; [4] CIP, Apartado 5969, Lima, Peru

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Summary

Effects of resistance genes and heat tolerance genes on expression of resistance to bacterial wilt caused by *Pseudomonas solanacearum* were investigated in 30 F_1 progenies from parents with different levels of bacterial wilt resistance and heat tolerance. A race 1 and a race 3 isolate of the bacterium were used for inoculation under screenhouse conditions at two locations. Results obtained indicated that with reduction in levels of parental resistance, resistance in the F_1 progenies was also reduced. Under hot conditions, a reduced heat tolerance in the genetic background also resulted in lower levels of resistance expression. The effect of heat tolerance tended to diminish at lower temperatures leaving the effect of resistance genes more consistent. There existed a strong interaction between resistance genes and genes for heat tolerance. The nature of resistance to bacterial wilt in potato and implications for breeding for resistance are discussed.

Effects of reciprocal crosses on expression of resistance to a race 1 isolate under hot screenhouse conditions, were studied in 5 sets of reciprocal F_1 progenies involving different resistant and susceptible parents. The reciprocal differences observed were not significant suggesting absence of cytoplasmic effects on expression of resistance.

Introduction

Breeding for resistance to bacterial wilt (BW), caused by *Pseudomonas solanacearum* (E.F. Smith), has been a difficult objective. Resistance is generally highly strain specific and greatly influenced by environmental conditions, especially temperature (Sequeira & Rowe, 1969; Thurston & Lozano, 1968; Ciampi & Sequeira, 1980; French & De Lindo, 1982; Tung et al., 1990b). Schmiediche (1983) and Kloos & Fernandez (1986) suggested that resistance to BW is probably a function of environmental adaptation. Tung et al. (1990a) demonstrated that genes for adaptation are involved in conferring the resistance. Heat tolerant parent clones gave higher proportions of resistant plants in their progenies under lowland tropic conditions. Potato clones with genes for both BW resistance and heat tolerance expressed a higher level of resistance under diverse environmental conditions (Tung et al., 1992). There is also evidence that under hot conditions certain heat tolerant potato clones with no known resistance could withstand BW better than other well-known resistant clones (Tung, unpublished). Resistance to BW thus seems very complex with genes for adaptation probably being involved in conditioning its expression. The extent to which heat tolerance genes help strengthen resistance under heat conditions was still not clear. Also the most effective means of combining BW resistance with heat tolerance to produce superior progenies, which are much desired in breeding potato for production in the warm tropics, needs to be determined.

The objective of the experiments described in this paper was to investigate the expression of resistance in F_1 progenies from parents with different degrees of BW resistance and heat tolerance and possible cytoplasmic effects on the expression of resistance in F_1 progenies from reciprocal crosses between different parents.

Materials and methods

In the first study, a set of potato clones (Table 1) with known genetic background for BW resistance and adaptation were used to produce sets of F_1 progenies with different average degrees of BW resistance and heat tolerance. Because most of the resistant materials currently being used in breeding programs derived their resistance from only a few original ancestors, some of the resistant parents used in this study are also more or less related. For instance, BR-63.5, BR-63.74 and BR-63.76 are full-sibs, BR-63.74 is a grandparent of 381064.3 and 381064.7, while AVRDC-1287.19 is the male parent of the latter two. In addition all clones coded BR- have virtually been developed from only two original resistant diploid *S. phureja* clones (Schmiediche, 1983).

The hybridization was conducted in the summer of 1989 at the CIP-Region VII Germpasm and Training Center at Sta. Lucia, the Philippines (800 m above sea level, asl, 14° N latitude). Crosses were made between parents which either lack or possess one or both desirable traits to be investigated: BW resistance and heat tolerance. This means four classes of parents (Table 1) and hence 16 types of single crosses including within- and between-class combinations. As there are crosses with similar reciprocal combinations of parental attributes, the number of types of crosses (hereafter called groups) to be tested was reduced to a meaningful 10 (Table 2), of which the Groups Va and Vb were similar in terms of degree of the traits of concern but differed from each other by the type of their resistant parents. Three crosses were made with their parent clones chosen at random from a desired class to represent a Group. There were thus a total of 30 single crosses forming 10 Groups differing in their levels of BW resistance and heat tolerance. For the sake of assessing the effects of differences in degree of resistance and/or heat tolerance

Table 1. Parents of the crosses studied in this article. The parents are classified according to their resistance or susceptibility to bacterial wilt (BW) and tolerance or sensitivity to heat (H).
+ symbol for the desirable traits, resistance or tolerance
− symbol for the undesirable traits, susceptibility or sensitivity

Class	Parent clone	Class characteristics BW/H	Source of resistance
1	378597.1	+/+	*Solanum phureja*
	AVRDC-1287.19	+/+	*S. raphanifolium* *S. chacoense*
	381064.3	+/+	*S. phureja*, AVRDC-1287.19
	381064.7	+/+	*S. phureja*, AVRDC-1287.19
2	BR-63.5	+/−	*S. phureja*
	BR-63.74	+/−	*S. phureja*
	BR-63.76	+/−	*S. phureja*
	BR-112.113	+/−	*S. phureja*
	Cruza-148	+/−	Unknown
3	LT-8	−/+	
	DTO-28	−/+	
	DTO-33	−/+	
	7XY.1	−/+	
	Atlantic	−/+	
4	I-1039	−/−	
	I-1035	−/−	
	CFK-69.1	−/−	
	Serrana	−/−	
	P-7	−/−	
	Conchita	−/−	

on levels of resistance expression, Groups were tentatively joined in two ways into two clusters of three Categories: 1, according to their degree of resistance regardless of their degree of heat tolerance, Resistance categories and 2, according to their degree of heat tolerance regardless of their degree of resistance, Heat tolerance categories (Table 2). This categorization assumes that the interaction effects between resistance genes and those for adaptation in all F_1s are all of the same magnitude and positive and negative effects will cancel out each other. Thus Resistance Categories will, on the average, have the same level of heat tolerance. Similarly, Heat tolerance Categories will have the same average level of BW resistance. Otherwise, this categorization is just a rough approximation which facilitates assessing the average effects of levels of resistance and heat tolerance.

The crosses were tested for resistance to a race 1 (WP-17) and a race 3 (WP-156) isolate under screenhouse conditions at two locations: Los Banos (150 m asl) and Sta. Lucia (800 m asl). The BW resistance tests were conducted during the periods February 28 – March 15, and March 10 – 25, 1990, at Los Banos and Sta. Lucia, respectively. At each location the F_1 progenies were tested in a splitplot randomized complete block design (RCBD) with isolates of *P. solanacearum* as main plots and the potato progenies as subplots in three replications of 30 plants.

F_1 true seeds were sown in heat sterilized volcanic soil in 0.50 m × 0.50 m boxes. Seedlings were transplanted, at the first true leaf stage, into 8 cm diameter clay-pots, containing the same type of soil. The plants were irrigated with tap water at 2-day intervals and fertilized weekly with a 1% solution of 14% N, 5% P and 12% K. Mancozeb (0.3%) and cypermethrin (0.1%) were sprayed every week to prevent infestation by fungi and insect pests. The test plants were trimmed to have one main stem one week prior to inoculation with a sterilized knife. Four weeks after transplanting, the plants were inoculated by clipping off the third and the fourth leaves, from the soil surface with a sterilized scalpel previously dipped into a 10^8 cells/ml inoculum suspension of the bacterium. For each isolate, a separate scalpel was used. For each test, plants (5 per isolate) of the susceptible clone 7XY.1, raised from apical cuttings of a disease free stock, were also inoculated to check for effectiveness of the inocula. This clone has been observed to wilt within 5 to 7 days after inoculation (DAI). During the incubation period, the inoculated plants were maintained in the screenhouse, and the daily maximum and minimum temperatures were recorded. On the 15 DAI, when check plants were completely wilted, disease incidence was recorded as percentage of plant survival in the populations and the disease index of individual plants was determined based on the following index scale: 1, no

Table 2. Groups and categories of F_1's obtained from 9 basically different combinations of parental clones. BW, bacterial wilt; H, heat. + = BW resistant/H tolerant; − = BW susceptible/H sensitive

Group	Parental attributes		Group characteristics	Categories for	
	Female BW/H	Male BW/H		Resistance	Tolerance
I	+/+	+/+	++/++	A	P
II	+/−	+/+	++/−+	A	Q
III	+/−	+/−	++/−−	A	R
IV	+/+	−/+	+−/++	B	P
Va	+/+	−/−	+−/+−	B	Q
Vb	+/−	−/+	+−/−+	B	Q
VI	+/−	−/−	+−/−−	B	R
VII	−/+	−/+	−−/++	C	P
VIII	−/+	−/−	−−/+−	C	Q
IX	−/−	−/−	−−/−−	C	R

wilt symptoms; 2, one or two leaves wilted; 3, half of the leaves wilted; 4, 3/4 of the leaves wilted; and 5, plant completely wilted or dead.

The second study was conducted to investigate the possible effect of the heat tolerant cytoplasm on the level of resistance to wilt. Four pairs of reciprocal F_1 progenies were produced using the clones BR-63.5 and BR-112.113 as resistant parents which have heat sensitive BW resistance derived from *S. phureja*. The clones I-1039 and DTO-33 were used as heat sensitive and tolerant parents, respectively, both being BW susceptible. The progenies were tested for resistance to isolate WP-17 in a screenhouse at Los Banos during the period 5–20 February, 1990, along with the reciprocal crosses of susceptible parents as checks. The experiment was conducted in an RCBD with two replications of 50 plants. All cultural practices, inoculation method, and temperature reading followed the same procedure as described in the first study. Percentage of survival was recorded at 3-day intervals and disease index at 15 DAI.

For all experiments, Arc-sin transformation of percentage of survival was used for analysis of variance. For the disease indices, the average value of the plants of each replicate was determined. As these values are usually normally distributed, no transformation was attempted.

Results

During the first study, the average max/min temperatures were 33.4/24.5° C (mean 28.9° C) at Los Banos and 29.7/19.8° C (mean 24.8° C) at Sta. Lucia. Under these conditions, though not favourable for tuberization, the potato plants still used to grow well, and hardly any abnormal growth or plant death occurred before inoculation. So wilting which occurred after inoculation was considered to be due entirely to *P. solanacearum* infection.

In general, wilt incidence was much higher at Los Banos than at Sta. Lucia for both % survival (Table 3) and disease index (Table 4). As found earlier (Tung et al., 1992), isolate WP-156 was much more virulent than WP-17 (Tables 3 & 4). Statistical analysis showed highly significant differences between locations, isolates and potato progenies. All types of interaction: isolate × progeny, isolate × location, progeny × location, isolate × progeny × location, were highly significant. The main effects due to isolates and progenies, however, were the major source of total variation, similar to previous findings (Tung et al., 1990b & 1992).

Against isolate WP-17, there was a great variation among progenies at both locations (Tables 3 & 4). Though some differences between progenies belonging to different Groups were not statistically significant, Groups I, II and IV, which have a high level of resistance coupled with heat tolerance, tended to show the highest level of resistance in both % survival and wilt index (Tables 3 & 4, Fig. 1). In the absence of heat tolerance, resistance in progenies of Groups III and VI tended to diminish under hot conditions at Los Banos and recovered significantly under Sta. Lucia conditions. Heat tolerance alone did not help in resisting wilt indicated by the low performance of progenies of Groups VII and VIII (Tables 3 & 4, Fig. 1). Under very hot conditions at Los Banos, however, Group VII seemed to be better than Groups III and VI which have resistance from one (VI) or both (III) parents, but lack heat tolerance. In general, in progenies without any resistance and/or heat tolerance (Groups VI to IX) wilting was almost complete at 15 DAI (Tables 3 & 4, Fig. 1).

Against isolate 156, the resistance was almost lost at both locations. Trends in resistance expression among the F_1 progenies similar to that with WP-17 were also observed (Tables 3 & 4, Fig. 1). Although the differences between progenies were greatly reduced, especially under Los Banos conditions, it was those with higher levels of BW resistance and heat tolerance in their pedigrees which tended to show less wilting (Tables 3 & 4, Fig. 1).

Results obtained from this study clearly showed the general impact of genes for resistance and heat tolerance on expression of resistance to BW in potato.

Effects of resistance

The effects of resistance genes could be examined more closely by comparing the means of Resistance Categories (A, B and C, Table 2). These have a similar average degree of heat tolerance but differed from each other by their genetic levels of resistance (Table 2). A Mann-Whitney test (Steel & Torrie, 1960) revealed that there were significant differences in % survival between progenies of Categories A and B against both isolates at both Sta. Lucia and Los Banos (Table 5). The differences between them in wilt index were found to be significant at both locations against WP-156 but the general tendency was that progenies of A tended to

Table 3. Survival (%) of 30 F_1 progenies 15 days after inoculation with two isolates of *P. solanacearum* at two locations under screenhouse conditions. For Group characteristics, see Table 2

Group	Cross	Los Baños		Sta. Lucia	
		WP-17	WP-156	WP-17	WP-156
I	381064.3 × 378597.1	44.9 b–e	17.8 ab	55.3 a–c	23.8 a
(++/++)	381064.7 × AVRDC-1287.19	71.3 a	17.9 ab	65.4 a	12.0 a–c
	AVRDC-1287.19 × 378597.1	69.2 ab	10.4 a–c	61.4 ab	7.8 a–c
II	BR-63.5 × 381064.7	27.3 d–h	10.6 a–c	48.7 a–c	18.4 a–c
(++/−+)	BR-112.113 × AVRDC-1287.19	56.4 a–c	11.0 a–c	58.2 ab	19.4 ab
	BR-63.74 × 381064.3	51.8 a–d	20.3 a	60.7 ab	18.3 a–c
III	BR-63.74 × BR-63.76	16.2 g–i	8.1 a–d	28.7 b–g	7.9 a–c
(++/−−)	BR-63.74 × BR-112.113	27.0 d–h	12.2 a–c	30.0 b–f	17.7 a–c
	Cruza-148 × BR-63.5	8.2 ij	8.0 a–d	22.1 d–h	12.7 a–c
IV	381064.7 × DTO-33	42.3 c–f	6.0 a–d	46.1 a–c	9.3 a–c
(+−/++)	381064.3 × Atlantic	39.7 c–g	11.3 a–c	39.9 b–f	13.5 a–c
	AVRDC-1287.19 × LT-8	51.3 a–d	6.3 a–d	51.4 a–c	2.3 b–c
Va	378597.1 × I-1035	22.8 e–h	0.0 d	30.1 b–f	10.8 a–c
(+−/+−)	381064.7 × I-1039	41.2 c–f	9.0 a–c	55.4 a–c	13.6 a–c
	381064.3 × I-1035	40.0 c–f	7.7 a–d	53.1 a–c	9.8 a–c
Vb	BR-63.5 × DTO-33	19.0 f–i	11.7 a–c	31.4 b–f	10.9 a–c
(+−/−+)	BR-63.5 × 7XY.1	10.1 h–j	3.2 cd	17.2 f–h	7.3 a–c
	BR-63.74 × Atlantic	23.2 e–h	0.0 d	24.1 c–g	0.0 c
VI	BR-63.74 × I-1039	4.7 jk	1.5 cd	23.8 c–g	0.0 c
(+−/−−)	BR-63.5 × I-1035	8.2 ij	2.3 cd	31.4 b–f	7.1 a–c
	Cruza-148 × I-1039	0.0 k	0.0 d	18.8 e–h	4.8 bc
VII	DTO-28 × Atlantic	24.0 e–h	9.0 a–c	10.3 g–i	0.0 c
(−−/++)	DTO-33 × Atlantic	13.4 g–i	6.7 a–d	18.8 e–h	6.6 a–c
	LT-8 × DTO-33	29.9 d–g	0.0 d	23.4 c–g	5.0 a–c
VIII	I-1035 × DTO-33	9.3 ij	0.0 d	17.7 f–h	10.0 a–c
(−−/−+)	DTO-28 × I-1039	7.7 ij	2.9 cd	0.0 i	0.0 c
	LT-8 × I-1039	5.6 jk	0.0 d	6.3 hi	1.7 bc
IX	P-7 × I-1039	16.7 g–i	0.0 d	8.5 hi	4.7 bc
(−−/−−)	Serrana × I-1035	0.0 k	0.0 d	0.0 i	0.0 c
	Conchita × CFK-69.1	7.9 ij	1.8 cd	22.4 d–h	7.2 a–c
CV (i)%		18.76		19.09	
CV (p)%		19.94		15.66	

Note: In a column, values followed by a common letter are not significantly different at P = 0.01 by Duncan's multiple range test. CV(i) = coefficient of variation for isolates, CV(p) = coefficient of variation for progenies.

give a better average index than did those of B (Table 5, Fig. 1). The non-significance in other cases was probably due to large significant variation among Groups within Categories. It is clear that under milder temperatures at Sta. Lucia, effects of resistance level were expressed more consistently. The lack of difference between the two Categories against WP-17 indicates that under hot conditions, the effects of heat tolerance genes have greatly masked the difference in level of resistance of these Categories.

Except for wilt index for WP-156 at Los Banos,

Table 4. Disease index of 30 F_1 progenies 15 days after inoculation with two isolates of *P. solanacearum* at two locations under screenhouse conditions. For Group characteristics, see Table 2

Group	Cross	Los Baños		Sta. Lucia	
		WP-17	WP-156	WP-17	WP-156
I	381064.3 × 378597.1	2.5 d–f	4.1 b	2.4 e–i	3.3 gh
(++/++)	381064.7 × AVRDC-1287.19	2.0 ef	4.1 b	1.8 h–j	4.3 c–f
	AVRDC-1287.19 × 378597.1	1.9 f	4.7 ab	2.0 g–j	4.5 a–d
II	BR-63.5 × 381064.7	3.6 a–d	4.8 ab	1.9 g–j	3.8 fg
(++/−+)	BR-112.113 × AVRDC-1287.19	2.4 d–f	4.7 ab	1.4 j	3.8 fg
	BR-63.74 × 381064.3	2.7 d–f	4.3 ab	1.5 ij	3.2 h
III	BR-63.74 × BR-63.76	4.4 a–c	4.7 ab	2.4 e–i	4.8 a–c
(++/−−)	BR-63.74 × BR-112.113	3.3 b–e	4.7 ab	2.4 e–i	3.9 ef
	Cruza-148 × BR-63.5	4.6 a–c	4.9 a	3.0 d–f	4.3 c–f
IV	381064.7 × DTO-33	2.6 d–f	4.8 ab	1.9 g–j	4.7 a–c
(+−/++)	381064.3 × Atlantic	2.6 d–f	4.7 ab	2.1 f–j	4.4 b–e
	AVRDC-1287.19 × LT-8	2.3 ef	4.9 a	1.9 g–j	5.0 a
Va	378597.1 × I-1035	3.1 c–f	5.0 a	2.6 d–h	4.4 b–e
(+−/+−)	381064.7 × I-1039	2.6 d–f	4.9 a	2.5 e–h	4.0 d–f
	381064.3 × I-1035	2.5 d–f	4.9 a	2.4 e–i	4.4 b–i
Vb	BR-63.5 × DTO-33	3.2 c–f	4.8 ab	2.4 e–i	4.8 a–c
(+−/−+)	BR-63.5 × 7XY.1	2.9 d–f	4.9 a	3.9 a–c	4.9 a–b
	BR-63.74 × Atlantic	2.9 d–f	5.0 a	2.8 d–g	5.0 a
VI	BR-63.74 × I-1039	4.8 ab	4.9 a	3.2 de	5.0 a
(+−/−−)	BR-63.5 × I-1035	4.5 a–c	4.9 a	2.5 e–h	4.7 a–c
	Cruza-148 × I-1039	5.0 a	5.0 a	3.0 d–f	4.8 a–c
VII	DTO-28 × Atlantic	4.4 a–c	4.8 ab	4.4 a–c	5.0 a
(−−/++)	DTO-33 × Atlantic	4.6 a–c	4.8 ab	4.2 a–c	4.8 a–c
	LT-8 × DTO-33	3.5 a–d	5.0 a	3.5 cd	4.9 ab
VIII	I-1035 × DTO 33	4.7 ab	5.0 a	4.2 a–c	4.8 a–c
(−−/−+)	DTO-28 × I-1039	4.8 ab	4.9 a	5.0 a	5.0 a
	LT-8 × I-1039	4.8 ab	5.0 a	4.9 ab	4.9 ab
IX	P-7 × I-1039	4.6 a–c	5.0 a	4.7 ab	4.9 ab
(−−/−−)	Serrana × I-1035	5.0 a	5.0 a	5.0 a	5.0 a
	Conchita × CFK-69.1	4.9 a	4.9 a	3.1 de	4.8 a–c
CV (i)%		3.41		7.73	
CV (p)%		2.17		6.25	

Note: In a column, values followed by a common letter are not significantly different at P = 0.01 by Duncan's multiple range test. CV (i)% = coefficient of variation for isolates, CV (p)% = coefficient of variation for progenies.

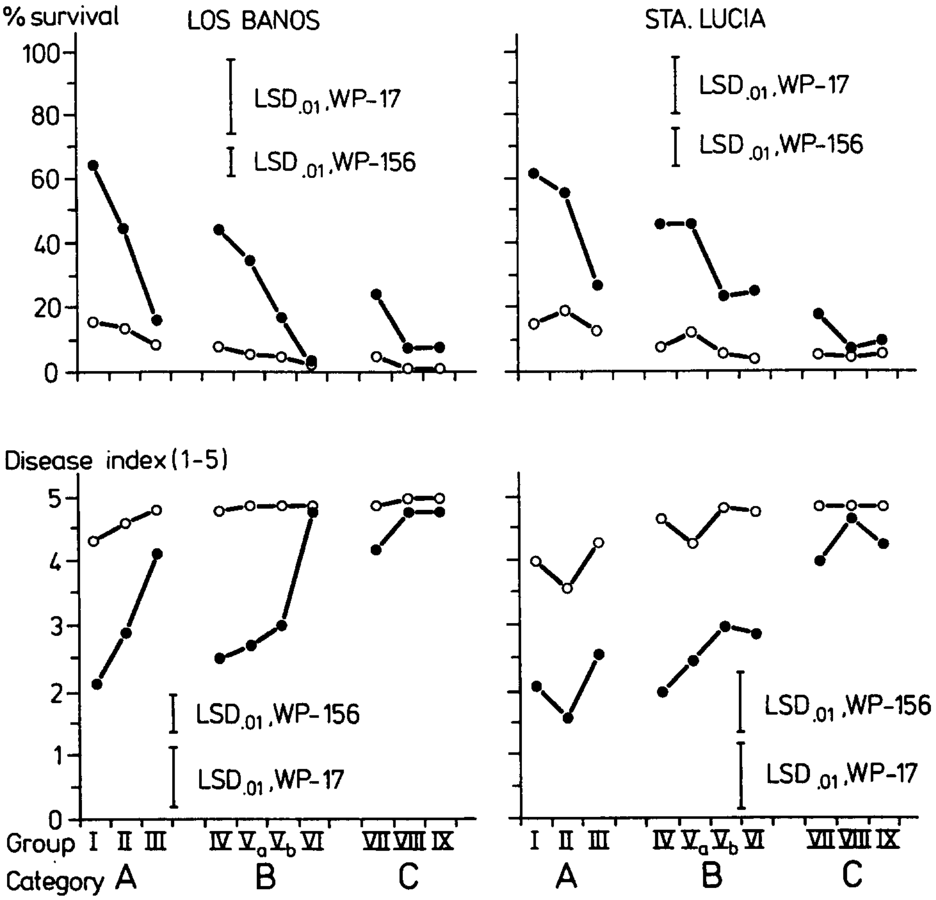

Fig. 1. Group averages for survival (0–100%) and wilt index (1–5) of 10 groups of F_1 progenies (of Tables 3 & 4), differing in level of BW resistance and heat tolerance, inoculated with isolates WP-17 (●) and WP-156 (○) of *P. solanacearum* at Los Banos (left) and Sta. Lucia (right).
Categories: A = resistance comes from both parents; B = resistance from one parent and C = resistance from neither parent.
Groups: I, IV & VII, heat tolerance from both parents; II, Va, Vb & VIII, heat tolerance from one parent, and III, VI & IX, heat tolerance from neither parent.

the differences between Categories A and C were significant in all cases (Table 5). An explanation for these differences is straightforward since these two Categories were the extreme situations: A had the highest level of parental resistance and C had no resistance at all. The differences in % survival between Categories B and C were statistically significant at both locations against isolate WP-17 and diminished under very hot conditions at Los Banos and/or against WP-156, in which situations resistance seemed to be lost (Table 5 and Fig. 1). The general tendency was that B tended to show higher levels of resistance than C (Fig. 1, Table 5).

As it was suggested earlier (Tung et al., 1992) that there is no effective gene(s) against isolate WP-156 in the population of potato clones used, the resistance to BW in this study also seemed to be broken down by this isolate. It is likely that the presence of some level of resistance against WP-156 in the progenies (Tables 3–5) was due to certain residual effects of the resistance which is effective against WP-17. The differences between Categories A and B in Los Banos might thus indicate that the residual resistance was to some extent correlated with the genetic level of resistance of the progenies as A always tended to resist wilt better (Table 5, Fig. 1).

Table 5. Effects of resistance (R)/susceptibility (S) of parents (P_1 and P_2) on % survival (upper part) and disease index (lower part) in F_1 progenies. Roman figures refer to the groups in Tables 3 and 4

F_1 groups pooled (Category)	Parental characteristics		Los Baños		Sta. Lucia		Overall category mean
	P_1	P_2	WP-17	WP-156	WP-17	WP-156	
% survival (*)							
I + II + III (A)	R	R	41.4 a	12.9 a	47.8 a	15.2 a	29.3 a
IV + V + VI (B)	R	S	25.2 b	4.9 b	35.4 b	7.2 b	18.2 ab
VII + VIII + IX (C)	S	S	12.7 c	2.2 b	11.9 c	3.8 b	7.6 b
	Mean		26.4	6.6	31.7	8.7	18.3
Disease index (1–5)(*)							
I + II + III (A)	R	R	3.0 a	4.6 a	2.1 a	4.0 a	3.4 a
IV + V + VI (B)	R	S	3.4 a	4.9 a	2.6 a	4.7 b	3.9 a
VII + VIII + IX (C)	S	S	4.6 b	4.9 a	4.3 b	4.9 b	4.7 b
	Mean		3.7	4.8	3.0	4.5	4.0

* In a column, values followed by a common letter are not significantly different at P = 0.05 by Mann-Whitney test.

Effects of heat tolerance

As mentioned in the foregoing section, there was a large variation among the Groups within Resistance Categories (Fig. 1). Groups within these Categories differed in their genetic levels of heat tolerance (Table 2). Although some of the between Group differences were not statistically significant, there was a general tendency that within a Category, Groups with higher levels of heat tolerance tended to show higher levels of resistance to wilt caused by both isolates at both locations, for both

Table 6. Effects of heat tolerance (T)/sensitivity (S) of parents (P_1 and P_2) on % survival (upper part) and disease index (lower part) in F_1 progenies. Roman figures refer to the groups in Tables 3 and 4

F_1 groups pooled (Category)	Parental characteristics		Los Baños		Sta. Lucia		Overall category mean
	P_1	P2	WP-17	WP-156	WP-17	WP-156	
			% survival (*)				
I + IV + VII (P)	T	T	42.9 a	9.5 a	41.2 a	8.9 a	25.6 a
II + V + VIII (Q)	T	S	26.3 b	6.4 a	33.4 a	10.2 a	18.7 a
III + VI + IX (R)	S	S	9.9 c	3.8 a	20.5 b	6.9 a	10.3 a
		Mean	26.4	6.6	31.7	8.7	18.3
			Disease index (1–5) (*)				
I + IV + VII (P)	T	T	2.9 a	4.7 a	2.7 a	4.5 a	3.7 a
II + V + VIII (Q)	T	S	3.4 b	4.8 a	3.0 a	4.4 a	3.9 a
III + VI + IX (R)	S	S	4.6 c	4.9 a	3.3 a	4.7 a	4.4 a
		Mean	3.6	4.8	3.0	4.5	4.0

(*) In a column, values followed a common letter are not significantly different at P = 0.05 by Mann-Whitney test.

% survival and indices (Fig. 1). The effect of heat tolerance genes was especially dramatic under Los Banos conditions and seemed to reduce at Sta. Lucia, where progenies of Group II and Va, having heat tolerance from only one parent, tended to be slightly better, against WP-156, than Group I which had twice the degree of heat tolerance. The differences were, however, not statistically significant.

When Groups with similar degree of heat tolerance were pooled in another way into Heat tolerance Categories (P, Q and R, Table 6), the effects of heat tolerance became even more clear. These three Categories differed significantly from each other in resisting wilt caused by WP-17 at Los Banos. At Sta. Lucia, these differences were levelled off and significance was found only for % survival and between P and R, and between Q and R (Table 6). With WP-156, no significant difference was found in any case as resistance was overcome by the isolate's extreme virulence.

Interaction

Large variation among Groups within Categories (Fig. 1) resulted in certain significant differences between them, on the one hand, and insignificant differences between Groups belonging to different Categories, on the other hand. While significant differences between Groups within Resistance Categories were attributable to the effects of heat tolerance genes, the lack of differences between Groups of different Categories indicated that there was a large amount of interaction between genes for resistance and genes for heat tolerance. This interaction effect also seemed to decrease considerably under milder temperatures at Sta. Lucia, where effects of heat tolerance became less pronounced and resistance was thus more clearly expressed. The significant differences between progenies within Groups (Tables 3 & 4) might be associated with the combining ability of the particular parent clones, as reported elsewhere (Tung et al., 1990a).

Whereas significant differences in overall means were detected between Resistance Categories (Table 5, last column), none was found between Heat tolerance Categories (Table 6, last column). This strongly suggests that though heat tolerance can greatly improve expression of resistance under heat stress conditions, tolerance alone would not make up any resistance to wilt and that resistance

Table 7. Average % survival and disease index of five reciprocal sets of F_1 progenies 15 days after inoculation with a race 1 isolate of *P. solanacearum*. For symbols BW, H, + and −, see caption of Table 1

Reciprocal set	Cross	Characteristics BW/H of		% survival	Disease index (1–5)
		Female	F_1		
1	BR-63.5 × DTO-33	+/−	+ −/− +	41.1 abc	3.4 abc
	DTO-33 × BR-63.5	−/+	− +/+ −	34.8 abc	3.7 abc
2	BR-112.113 × DTO-33	+/−	+ −/− +	47.7 ab	2.8 bc
	DTO-33 × BR-112.113	−/+	− +/+ −	52.3 a	2.6 c
3	BR-63.5 × I-1039	+/−	+ −/− −	10.5 cd	4.1 ab
	I-1039 × BR-63.5	−/−	− +/− −	13.6 cd	3.9 abc
4	BR-112.113 × I-1039	+/−	+ −/− −	36.5 abc	3.9 abc
	I-1039 × BR-112.113	−/−	− +/− −	48.5 ab	3.2 bc
5	DTO-33 × I-1039	−/+	− −/+ −	6.8 d	4.8 a
	I-1039 × DTO-33	−/−	− −/− +	4.1 d	4.9 a
CV (%)				15.02	10.04

Note: In a column, values followed by a common letter are not significantly different at P = 0.05 by Duncan's multiple range test.

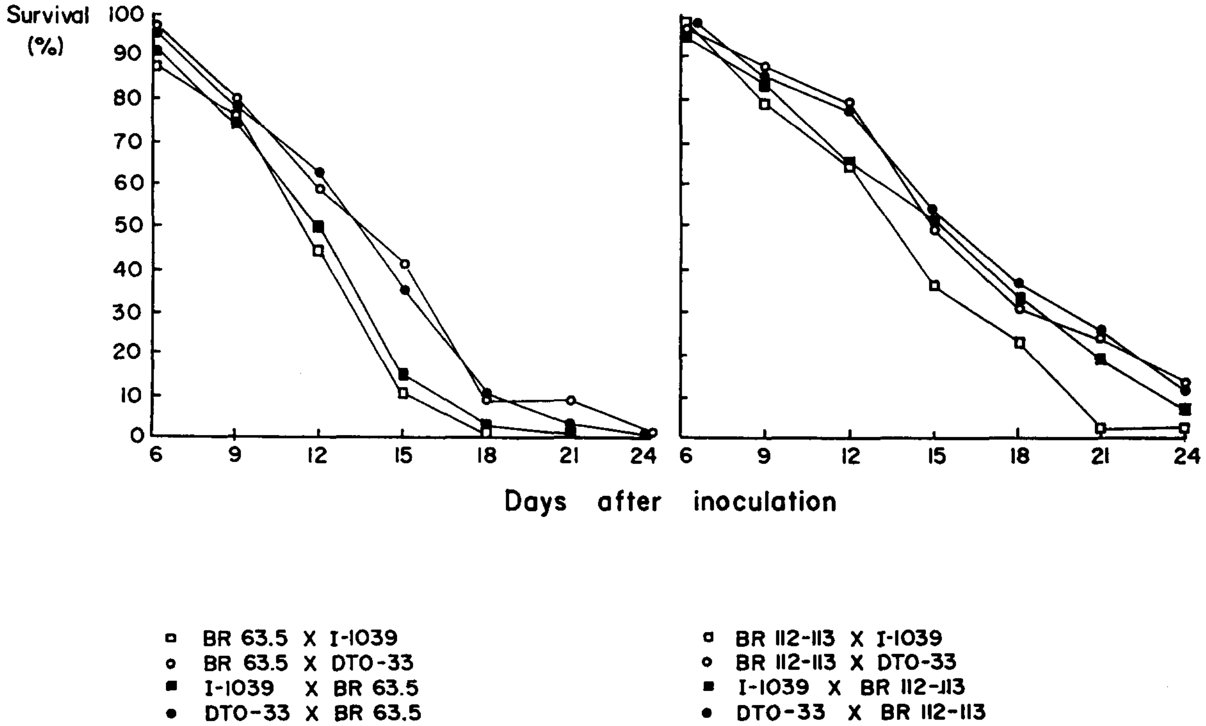

Fig. 2. Survival patterns of four pairs of reciprocal crosses after inoculation with isolate WP-17 of *P. solanacearum.*
In all cases, one parent (coded BR-) is BW resistant; □ & ■, both parents are heat sensitive; ○ & ●, one parent is heat tolerant; Squares vs circles exhibits effects of heat tolerance; Black vs white compares reciprocal crosses.

genes are indispensable despite the fact that their expression may be strongly affected by changes in environmental conditions.

Effect of reciprocal crosses

During the period of the test for effect of reciprocal crosses at Los Banos, the max/min temperatures in the screenhouse averaged 32.5/23.4° C with a mean of 27.9° C.

Under the conditions of this experiment, no significant effect of reciprocal crosses of any pair was detected in terms of both % survival and disease index (Table 7). It was the effect of a particular parent clone which gave the general impact of resistance expression (Table 7, Fig. 2). The clones BR-112.113 and BR-63.5 have been recognized for their resistance to BW under cool conditions. However, they usually succumb to the disease under hot conditions. In this experiment, BR-112.113 showed to be the better resistant parent compared to BR-63.5 at the 15 DAI (Table 7, Fig. 2), especially in the absence of heat tolerance. A similar comparision could be made between DTO-33 and I-1039, as the first was superior in giving more resistant offsprings (Table 7). The results obtained confirm our previous finding (Tung et al., 1990a) that combining ability is an important feature of BW resistance in potato and heat tolerance plays a crucial role in expression of resistance under hot conditions. The survival curves of the progenies showed that though the effect of parent clones was apparent during the first 15 DAI, the differences between them tended to level off at later dates. This was true for both resistant and susceptible parents, heat tolerant or sensitive. The resistance to BW thus seems to be the ability to slow down the bacterial multiplication and to delay wilt.

Discussion

Rowe and co-workers (Rowe & Sequeira, 1970; Rowe et al., 1972) reported on a system of three independent and dominant major genes that control specific resistance to two race 1 strains of *P. solanacearum* in the diploid *S. phureja*. Results from their later experiments, however, did not support their initial hypothesis (Sequeira, 1979). Since then, no further efforts have been dedicated to genetic analysis of BW resistance in potatoes, probably due to the very complex nature of the resistance: strain specificity and sensitivity to fluctuations in environmental conditions.

Results from this study confirm our previous finding that genes for adaptation are involved in conferring BW resistance (Tung et al., 1990a, b; 1992). This is indicated by the considerable effect of heat tolerance genes in conditioning resistance under heat stress conditions and their interaction with genes for resistance. Full susceptibility to isolate WP-156 further indicates absence of a gene(s) with a major effect in conditioning resistance to wilt caused by this isolate in the potato populations tested. There is obviously a strong gene(s) for resistance to WP-17 present in these populations. It is thus evident that the genetics of resistance to *P. solanacearum* in potatoes is very complex with both genes with major and minor effects involved. Strong interaction between genes for heat tolerance and those for resistance implies the presence of a large amount of favourable non-additive (epistatic) gene effects in a high resistance expression. In this study, there was further evidence that a widened genetic background for resistance and adaptation would provide a higher level of resistance which is likely more stable under diverse circumstances. This was exemplified by the performance of progenies of the c0lones 381064.3, 381064.7, and AVRDC-1287.19, which have undergone several cycles of selection for resistance and adaptation to warm conditions and combine resistance from at least two (AVRDC-1287.19) to three (381064.3, 381064.7) specific sources (Table 1).

All the aforementioned suggests that breeding at the population level by incorporating multiple sources of resistance and heat tolerance should be effective in producing superior genotypes suitable for potato production in the lowland tropics where high levels of BW resistance and heat tolerance are much needed. Recurrent selection in a population with a wide genetic background for resistance and adaptation should be a promising approach. An example is the great improvement in resistance to *Corynebacterium insidiosum* McCull Jens. (bacterial wilt) in alfalfa after just three cycles of recurrent selection (Barnes et al., 1971). If residual resistance to *P. solanacearum* is of any significance, incorporation of multiple sources of resistance in a genotype should also be an advantage in circumstances where mild compatible pathotypes/strains are prevalent, because residual effects seem to correlate with level of resistance. This advantage would be desirable especially under the conditions of subsistence agriculture in the developing world.

The foregoing discussion did not consider latent infection of tubers with *P. solanacearum* as it was shown by Ciampi & Sequeira (1980) that wilt symptoms are generally not correlated with latent infection. They suggested that latent infection of tubers is controlled by different genetic factors. Setting latent infection aside and taking into account the tremendous variation in pathotypes of *P. solanacearum* and involvement of genes for adaptation, one has to think of the BW resistance as quantitative and probably polygenic in nature.

The effect of reciprocal crosses, as tested in this study, on expression of resistance to BW did not show to be significant. The effect of a particular parental genotype appeared to be more important. Effects of reciprocal crosses need, however, further investigations, preferably in a controlled environment, since cytoplasmic effects are usually a sensitive objective demanding more sensitive test procedures.

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