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Studies on the inheritance of resistance to crown gall disease of grapevine

by

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Untersuchungen zur Vererbung der Mauke-Resistenz bei der Rebe

Zusammenfassung. — Annähernd 1800 Genotypen aus 27 Kreuzungsfamilien wurden auf ihre Anfälligkeit gegen den Stamm AT-1 des Biotyps 3 von *Agrobacterium tumefaciens* geprüft. Die untersuchten Kreuzungsfamilien umfaßten 12 Kreuzungskombinationen resistenter und anfälliger Phänotypen und 15 Selbstungen resistenter und anfälliger Eltern. Die Quelle der Mauke-Resistenz ist *Vitis amurensis*. Das Aufspaltungsverhältnis zwischen resistenten und anfälligen Sämlingen beträgt bei Fremdbefruchtung 1 : 1 und bei Selbstung der resistenten Eltern 3 : 1; bei Selbstung der anfälligen Eltern bildete jeder Sämling Tumoren aus. Aufgrund der Ergebnisse kann eine dominante Vererbung der Resistenz gegen *A. tumefaciens* AT-1 angenommen werden.

Introduction

Resistance of *Vitis amurensis* to *Agrobacterium tumefaciens* (synonymous name: *A. radiobacter* subsp. *tumefaciens*) was first described by TAMM in 1954. Four of the five plants of *V. amurensis* tested failed to form tumors after infection by *A. tumefaciens* strain AT-1 belonging to biotype 3. The resistance was expressed in the *V. vinifera* × *V. amurensis* F_1 hybrids, in a *V. amurensis* × *V. vinifera* F_2 hybrid (28/19) and in the BC_1 and BC_2 progenies of 28/19 (SZEGEDI *et al.* 1984).

Further resistant hybrids were found among the selected offsprings of A2/11, which was obtained from a *V. amurensis* × *V. vinifera* BC_1 family. The resistance to *A. tumefaciens* AT-1 was manifested in the C-26 and C-43 (A2/11 × Seyve-Villard 12375) and in the D-7, D-8 and D-9 [A2/11 × (Chaus × Pearl of Csaba)] hybrids, too (KRISZTEN and SZEGEDI, unpublished results).

This report deals with the inheritance of resistance to crown gall disease of grapevine. Experiments were carried out on hybrids and varieties having *V. amurensis* in their parentage crossed in Hungary for frost and downy mildew resistance (KOLEDA 1975). The purpose of our studies was to obtain a workable genetic model for breeding programmes.

Materials and methods

Plant material

The *V. amurensis* clones 34 and 115, the hybrids A2/11 and C-43 and the variety Kunbarát were used for crosses as resistance sources. C-43 was obtained from A2/11 × Seyve-Villard 12375, Kunbarát from 28/19 × Italia crosses. The resistant D-7, D-8 and D-9 obtained from the cross A2/11 × (Chaus × Pearl of Csaba) were also tested.

Among the sensitive parents tested, Favorit, Pearl of Csaba and Sultanina are European varieties, Seyve-Villard 12375, Pearl of Zala and R-10 are of Franco-American, B-45 and Kunleány of *V. amurensis* origin.

Seeds obtained from crosses and self-pollinations were disinfected with 0.5 % (w/v) Solvochin Extra (8-hydroxyquinolinsulphate) for 2 h and washed twice with autoclaved tap water. Disinfected seeds were layered into moist perlite and germinated in steam-sterilized garden soil at the end of March. The young seedlings having 4—6 leaves were infected after wounding with a heavy bacterial inoculum on the stem at two sites. Inoculated plants were kept in greenhouse at 23—28 °C. Results were scored after 6-week incubation.

The following combinations were tested for genetical studies: resistant × sensitive, sensitive × resistant crosses, and self-pollinated seedlings both of sensitive and of resistant parents.

Bacterial strain

A. tumefaciens strain AT-1 belonging to biotype 3 was used for genetical studies because of its virulence on a wide range of grapevine varieties (SZEGEDI 1981) and the dominance of biotype 3 strains on grapevine (LOUBSER 1978, PANAGOPOULOS *et. al.* 1978, SÜLE 1978, PERRY and KADO 1982, BURR and KATZ 1983). Bacterial cultures were grown on YE agar (10 g glucose, 5 g yeast extract and 15 g agar dissolved in 1,000 ml of distilled water) at 25 °C for 48 h prior to infection.

Results and discussion

Tumors became visible at the sites of infection within 3 weeks. After 6-week incubation, the susceptible seedlings formed relatively large tumors (6—8 mm in diameter), while on the resistant plants only wound healing was observed.

The average segregation of resistant × sensitive crosses was 56.25 % resistant to 43.75 % sensitive. All plants of the *V. amurensis* 34 × Favorit family proved to be resistant. In the case of sensitive × resistant combinations 42.2 % of progenies failed to form tumors and 57.8 % were susceptible (Tables 1 and 2). Among the selfings from resistant parents (hybrids) 74.3 % proved to be resistant and 25.7 were sensitive. Each of the sensitive parents' self-pollinated seedlings was susceptible (Tables 3 and 4).

The segregation ratio in the crosses corresponded to 1 : 1 and was 3 : 1 (resistant to sensitive) in the selfings of resistant parents. Deviations of R-10 × A2/11 and R-10 × C-43 families could be caused by self-pollination of the sensitive parent during the crossing procedure (Table 2).

The resistance of BC_1 hybrids obtained from 28/19 (SZEGEDI *et al.* 1984) was determined by 28/19 considering that each of the selfings of Afuz Ali, Italia and of Muscat of Thalloczy Lajos were sensitive. Similarly, the resistance of C and D signed hybrids could be assigned to A2/11, because the selfings of Seyve-Villard 12375 and of Chaus × Pearl of Csaba were uniformly susceptible, while the self-pollinated seedlings of A2/11 showed 3 : 1 segregation ratio.

C-43 was backcrossed both with its resistant (A2/11) and sensitive (SV-12375) parents. The segregation corresponded to 3 : 1 in the case of the C-43 × A2/11 cross and to 1 : 1 in the C-43 × SV-12375 family. Selfings of the A2/11 showed a 3 : 1 ratio, while the SV-12375 selfings were uniformly susceptible (see Tables 3 and 4).

Table 1

The inheritance of resistance in resistant × sensitive crosses

Vererbung der Resistenz bei der Kreuzungskombination „resistent × anfällig"

Cross	No. of plants tested	Resistant	Sensitive	Expected ratio	χ^2
1. *V. amurensis* 34 × Favorit [1])	28	28	0	Uniform	—
2. *V. amurensis* 115 × Favorit	28	15	13	1 : 1	0.14
3. A2/11 × Sultanina	124	70	54	1 : 1	2.06
4. C-43 × Favorit	44	21	23	1 : 1	0.09
5. C-43 × Pearl of Csaba	85	51	34	1 : 1	3.40
6. C-43 × Seyve Villard 12375	23	14	9	1 : 1	1.08
All plants tested	304	171	133		
%	(100)	(56.25)	(43.75)		

[1]) These results were not considered in the summarised dates.

Table 2

The inheritance of resistance in sensitive × resistant crosses

Vererbung der Resistenz bei der Kreuzungskombination „anfällig × resistent"

Cross	No. of plants tested	Resistant	Sensitive	Expected ratio	χ^2
1. Pearl of Zala × Kunbarát	124	60	64	1 : 1	0.12
2. Kunleány × Kunbarát	29	15	14	1 : 1	0.03
3. R-10 × A2/11	97	33	64	1 : 1	10.86
4. R-10 × C-43	72	23	49	1 : 1	9.38
5. B-45 × C-43	73	31	42	1 : 1	1.64
6. Favorit × C-43	41	22	19	1 : 1	0.21
All plants tested	436	184	252		
%	(100)	(42.2)	(57.8)		

According to the results mentioned above the resistance to *A. tumefaciens* strain AT-1 is determined by a single dominant gene in the tested hybrids. *V. amurensis* 34 is probably homozygous for resistance, while the further sources *(V. amurensis* 115, Kunbarát, A2/11 and C-43) are heterozygous resistant genotypes.

Table 3

Segregation of self-pollinated seedlings from resistant parents

Aufspaltung bei den Selbstungen resistenter Eltern

Parent	No. of plants tested	Resistant	Sensitive	Expected ratio	χ^2
1. Kunbarát	90	68	22	3 : 1	0.01
2. A2/11	121	97	24	3 : 1	1.72
3. C-43	98	74	24	3 : 1	0.01
4. D-7 [1])	69	47	22	3 : 1	1.73
5. D-8 [1])	59	39	20	3 : 1	2.48
6. D-9 [1])	62	46	16	3 : 1	0.02
All plants tested	499	371	128		
%	(100)	(74.3)	(25.7)		

[1]) Families obtained from GY. KRISZTEN.

Table 4

Segregation of self-pollinated seedlings from sensitive parents

Aufspaltung bei den Selbstungen anfälliger Eltern

Parent	No. of plants tested	Resistant	Sensitive	Expected ratio
1. Favorit	33	0	33	Uniform
2. Muscat of Thallóczy Lajos	49	0	49	Uniform
3. Afuz Ali	68	0	68	Uniform
4. Italia	27	0	27	Uniform
5. Chaus × Pearl of Csaba [1])	13	0	13	Uniform
6. R-10	112	0	112	Uniform
7. Pearl of Zala	87	0	87	Uniform
8. Seyve-Villard 12375 [1])	96	0	96	Uniform
9. B-45 [1])	50	0	50	Uniform
All plants tested	535	0	535	
%	(100)		(100)	

[1]) Families obtained from GY. KRISZTEN (5. and 9.) and L. BEREZNAI (8.).

The proposed genetic model can be adopted for the studied host-pathogen system only, because the resistance (or susceptibility) of some grapevine varieties was strain-specific to biotype 1, while no differences were found between the tested biotype 2 and 3 strains (SZEGEDI *et. al.* 1984).

Greek grapevine varieties showed similar differences in their susceptibility to *Agrobacterium* strains having different Ti-plasmids and/or chromosomal background (KNAUF *et al.* 1982).

Crown gall disease attacks the woody parts of grapevine under field conditions, therefore it was reasonable to infect parallelly both the woody and the green stem. No differences were found in the susceptibility of woody and green parts of 4 resistant and 6 sensitive varieties (unpublished own results). These observations suggest that the screening method of young seedlings applied here could be reliable for early selection of resistant hybrids.

The sources used here were selected for frost and downy mildew resistance and for fruit and wine quality. The Mendelian dominant inheritance of resistance to *A. tumefaciens* allows to combine these important quality factors with resistance gene(s) in the same variety.

Summary

Nearly 1800 seedlings of 27 hybrid families were screened for resistance to *A. tumefaciens* strain AT-1 belonging to biotype 3. The tested families included 12 crosses of sensitive and of resistant phenotypes, and 15 self-pollinated families both of sensitive and of resistant parents. Resistance originated from *V. amurensis*. The segregation ratio corresponded to 1 : 1 among the crosses and was 3 : 1 (resistant to sensitive) among the selfings of resistant parents. All seedlings from selved susceptible parents formed tumors. According to our results, a Mendelian dominant inheritance can be proposed for resistance to *A. tumefaciens* strain AT-1.

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