Susceptibility of Grape-vine Rootstocks to Strains of *Agrobacterium tumefaciens* biovar 3.

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Forty grape-vine rootstocks were inoculated with five strains of *Agrobacterium tumefaciens* biovar 3 in a field trial. Grape-vine material like Paulsen 775 was immune to all strains whereas Kober 125 AA, Freedom, 3309 C, Harmony, Solonis, 101-14 Mgt., Berlandieri 13/5 and Kober 5BB appeared to be resistant. Jacquez, US 2-1, US 24-23, Muscat d'Alexandrie and US 24-10 were the most susceptible. Host range differences occurred among the five strains of *A. tumefaciens* on the different rootstock cultivars. Five different host range patterns were evident. *A. tumefaciens* biovar 3 strain 1771 was the most virulent and strain 2221 was the least virulent of the strains used.

Crown gall on grape-vine caused by *Agrobacterium tumefaciens* Conn. biovar 3 is an economically important disease in various countries (Panagopoulos & Psallidas, 1973; Süle, 1978) including the Republic of South Africa (Matthee, Thomas & Du Plessis, 1977). Biovar 3 strains of *A. tumefaciens* appear to be pathogenic on grape-vine only (Panagopoulos, Psallidas & Alivizatos, 1978). Reduced yields occur in affected grape-vines and they tend to die back.

Dieback among grape-vines was reported from the Orange River irrigation area, Northern Cape, which led to the first identification of the causal organism, i.e. *A. tumefaciens* biovar 3, in South Africa (Loubser, 1978). Nearly 50% of vineyards in the same area were infected with crown gall which in some instances caused up to 50% reduction in yield (J. Louw, personal communication, 1984).

Limited information is available concerning susceptibility of grape-vine rootstock cultivars to *A. tumefaciens*. Preliminary observations indicated that Jacquez and scion cultivars Sultanina and Muscat d'Alexandrie were extremely susceptible to crown gall disease. It was necessary, therefore, to evaluate potential rootstock cultivars for the Orange River area for their susceptibility to crown gall.

MATERIALS AND METHODS

A trial was laid out at Upington in the Orange River area during August 1981 using a randomised block design. The forty-one different rootstocks tested for susceptibility are presented in Table 1. Rooted Muscat d'Alexandrie was included because it is susceptible to crown gall disease. Each cultivar was replicated eight times with each replicate consisting of five plants. Visually disease free material, rooted in mist beds, were kept in pots in a glasshouse for one year prior to planting.

During November 1983 all cultivars were artificially inoculated with five different strains of *A. tumefaciens* biovar 3. Local strains 2160 and 2164 were isolated from Muscat d'Alexandrie, 2221 from Sultanina and strain W7 was isolated from an unidentified vine. Strain 1771, isolated from grape-vine in Iran, was obtained from the National Collection of Plant Pathogenic Bacteria, Hertfordshire, England. Preliminary laboratory tests showed these five strains to be the most pathogenic on grape-vine (F.G.H. van Zyl, unpublished data).

Cultures were grown on nutrient agar slants in Mc-Cartney bottles for forty-eight hours. Ten millilitres of sterile distilled water was added to each of the cultures before use and shaken well to prepare a bacterial suspension (approx. 5×10^5 cells/ml).

Each plant was inoculated with a different bacterial strain. A cut (10 mm long and 5 mm deep) was made in the trunk 200 mm above ground level with a sterile knife (immersed in 70% ethanol for two minutes) to expose the xylem tissues. A drop (0,1 ml) of the bacterial suspension was applied to the cut with a syringe and covered with masking tape to prevent desiccation.

Four months after inoculation (March, 1984) gall diameters were measured. Tumour formation was again measured at the same time the following year (March, 1985). Susceptibility of the different cultivars was determined with Friedman's two way analysis of variance by ranks (Siegel, 1956). The greatest measurement from the two years was used for analyses because in many cases galls had dried out and fallen off during winter.

RESULTS AND DISCUSSION

Paulsen 775 was the only immune cultivar to the five strains tested, while Jacquez was the most susceptible (Table 1). Although significant differences occurred among rootstocks according to rank sums, these cultivars can be arbitrarily classified according to rank sums of the mean gall diameter as immune, resistant, moderately susceptible, and susceptible. Paulsen 775 is therefore classified as immune while Kober 125 AA, Freedom, 3309 C, Harmony, Solonis, 101-14 Mgt., Kober 5 BB and Berlandieri 13/5 are considered as resistant.

Significant differences were found in susceptibility of different rootstocks to the different *A. tumefaciens* strains (Fig. 1). *A. tumefaciens* strain 2221 affected the fewest and strain 1771 the most rootstocks. Different rootstock cultivars tend to differ in their response to the individual bacterial strains. For example, 110 Richter seems more susceptible to strain 2164 than to the other strains. Not all strains infected all rootstock cultivars. For example Teleki 5C was infected by strains 2164, 2221 and 1771 but not by W7 and 2160. Comparison of the virulence of the strains' rank sums, suggests that

strain 2221 was the least, and strain 2164 the most virulent.

From the results it is clear that the *A. tumefaciens* biovar 3 strains showed five different host range patterns.

A. tumefaciens strain 1771 infected the highest percentage (90,2%) of rootstock cultivars tested followed by 2164 (80,5%), W7 (73,2%) and 2160 (60,9%). A. tumefaciens strain 2221 infected the lowest number of rootstock cultivars (46,3%). This suggests that the five strains are host specific.

TABLE 1

Susceptibility of different grape-vine rootstock cultivars and crosses to crown gall.

Rootstock cultivars and crosses	Cross or species	Mean gall diameter (m	Rank m) sums
Paulsen 775	Immune V. berlandieri x V. rupestris	0,00	54,5
	Resistant		
Kober 125 AA	V. berlandieri x V. riparia	0,2	66,5
Freedom	1613 x Dogridge	0,2	68,5
3309 C	V. riparia x V. rupestris	0,2	68,5
Harmony	Dogridge x 1613 C	0,4	75,5
Solonis	V. candicans x V. riparia x V. arizonica	0,6	79,5
101-14 Mgt	V. riparia x V. rupestris	0,8	86,0
Kober 5BB	V. berlandieri x V. riparia	0,8	95,0
Berlandieri 13.5	Selection V. berlandieri	1,1	94,5
	Moderately susceptible		
Selection Oppenheim 4	V. berlandieri x V. riparia	1,4	105,5
216/3 Castel	Solonis x V. rupestris	1,5	105,5
Paulsen 1045	V. berlandieri x A. x RG II	1,8	87,0
161-Couderc	V. berlandieri x V. riparia	1,8	117,5
Teleki 5C	V. berlandieri x V. riparia	1,9	123,5
333 Em	V. vinifera x V. berlandieri	1,9	126,5
US 24-41	Ramsey x 99 Richt	er2,1	120,5
A x RG II	V. vinifera x V. rupestris	2,5	139,0
US 16-13-23	1202 x 99 Richter	2,5	144,0
US 24-26	1202 x 99 Richter	2,6	149,0
26 C	V. vinifera x V. riparia	3,4	160,5
LSD ($P = 0.05$) ran	k sums 190,0		

Rootstock cultivars and crosses	Cross or species	Mean gall diameter (mm)	Reank sums
Grezot-1	1616 x V. rupestris	3,5	167,0
Paulsen 1103	V. berlandieri x V. rupestris	3,8	162,0
Prosperi Super R99	V. berlandieri x V. rupestris	4,1	168,5
4401 Coudert	V. vinifera x V. riparia	4,3	169,5
1202 C	V. vinifera x V. rupestris	4,4	179,0
	Susceptible		
US 1-6	Jacquez x 99 Richter	7,4	191,0
110 Richter	V. berlandieri x V. rupestris	5,8	179,0
99 Richter	V. berlandieri x V. rupestris	6,0	194,5
Constantia Metallica	V. rupestris var. martin	6,3	207,5
14D Ruggeri	V. berlandieri x V. rupestris	6,6	204,0
Ramsey (5/18/10)	V. champini	8,1	241,0
143-B Mgt	V. vinifera x V. riparia	8,3	238,0
US 12-6-8	Jacquez x 99 Richter	8,4	237,7
US 3-6	Jacquez x 99 Richter	8,6	249,0
Ramsey (5/19/5)	V. champini	9,8	264,0
US 4-4	Jacquez x 99 Richter	10,7	269,5
US 24-10	Ramsey x 99 Richter	12,3	281,0
Muscat d'Alexandrie		12,3	282,5
US 24-23	Ramsey x 99 Richter	15,4	304,0
US 2-1	Jacquez x 99 Richter	16,6	304,0
Jacquez	V sestivalis x V. cinerea x V. cinifera	17,1	316,0

CONCLUSIONS

Rootstock cultivars varied in their susceptibility to crown gall. All five biovar 3 strains of *A. tumefaciens* infected the susceptible rootstock cultivars. Paulsen 775 was the only immune cultivar.

The host range of *A. tumefaciens* biovar 3 strains isolated from grape-vine varies considerably. Both strains 2160 and 2164 were isolated from the same host species in the same geographical area. Both these strains possessed their own different specific host range. Strain 1771 had the widest host range and strain 2221 the narrowest. There was a difference in virulence between strains which was reflected by gall size observed on the same cultivars in the field trial.

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(mm)

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Susceptibility of different rootstock cultivars to five A. tumefaciens biovar 3 strains according to mean tumour diameter.

The long term effect of crown gall infested rootstock material on yield and longevity of different scion cultivars has not been determined. However, growers are advised to use resistant or tolerant rootstock material when replanting in problem areas. It is very important to breed or select for disease resistance as this is probably the most important factor in disease control. This is a first report on resistance selection of grape-vine rootstock cultivars and crosses to crown gall for a specific grape-vine cultivation area.

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