

Resistance of rose rootstocks to crown gall (*Agrobacterium tumefaciens*)

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Abstract

Various types of rose rootstocks were tested for their resistance to crown gall. The rootstock 'Iowa State University (ISU) 60-5' was the most resistant, followed by 'Brooks 48', 'Clarke 1957' and 'Welch'. *Rosa multiflora*, *R. noisettiana* ('Manetti') and 'Basye No. 3' were very susceptible. The inoculations were made with four isolates of *Agrobacterium tumefaciens* (Smith et Townsend) Conn, respectively from a *Dahlia* sp., *Rosa* spp. and *Prunus persica*. It was found that the isolate from *Dahlia* was a different race to the isolates from *Rosa* and *Prunus* spp.

Introduction

The rose has long been listed as a plant subject to crown gall (*Agrobacterium tumefaciens* (Smith et Townsend) Conn); references in literature, published in the United States of America, began to appear about 1892 (Massey, 1950). Munnecke et al. (1963) made a comparative study of hairy root (*A. rhizogenes*) and crown gall on roses inter alia, including rootstocks, but apart from this, relatively little experimental work has been done on this disease. The present study is part of a project, which was undertaken to find types of rose rootstocks which would be more suitable than the thorny *Rosa multiflora*, which was generally used as rootstock when the investigations were started. Some of the rootstocks that were tested for resistance to crown gall fulfilled the horticultural requirements for rooting, budding capacity, vigour and thornlessness.

Material and methods

Rootstocks. The following rootstocks were included: *Rosa noisettiana* ('Manetti'), *Rosa multiflora* (type) and various forms: 'Clarke 1957', 'Brooks 48', 'Welch', 'Iowa State University (ISU) 60-5' and 'Basye No. 3'. Cuttings about 8 inches long, were planted in winter in sterilized soil in 8" pots; they were grown in a shaded glasshouse. The number of cuttings per treatment varied from 8 to 14 in each test, except in the final tests with 'Brooks' and 'ISU 60-5', when between 18 and 20 cuttings per treatment were used and also in the case of crown inoculations of *R. multiflora* (type), when only 3 cuttings were used.

Tomatoes, cv 'Pearson', that were 5 weeks old, were used as test plants.

Isolates of *A. tumefaciens*. Isolations were made according to methods described by Dowson (1957). The pathogenicity of bacterial cultures isolated in this way was tested

on sunflower (*Helianthus annuus*, cv 'Jupiter'), by introducing the cultures into the hypocotyls or epicotyls of young seedlings by means of a needle prick. Four isolates were included in the tests on rose rootstocks and tomatoes, respectively from (a) dahlia (KGD), (b) a hybrid tea rose (KGR), (c) rose rootstock Basye No. 3 (KGB) and (d) peach (KGPe). The cultures were isolated and grown at 25° or 27°C on one of the following Difco media: Bacto-Nutrient Agar, Bacto-Yeast Dextrose Agar or Bacto-Tryptone Glucose Extract Agar.

Inoculation methods

Rose rootstocks. Two methods of inoculation were used, viz. T-cut and crown inoculation.

T-cut inoculation. T-cuts were made in the bark of rootstocks in the same way as for budding, about four inches above the ground. Crown gall bacteria were then introduced under the bark by means of a camel hair brush dipped in a suspension of one of the isolates of *A. tumefaciens* in sterile water. The wound was then bound with "Plastrip". This was removed when tumours started to develop or when the wounds had healed. Control plants were treated with sterile water.

Crown inoculation. Rooted cuttings were taken out of the soil and a slanted cut was made at the base of the rootstock. The wounded end of the rootstocks with the remaining roots was then dipped in a suspension of *A. tumefaciens* in sterile water. After the branches had been trimmed the rootstocks were planted again. The control rootstocks were treated in the same way, but dipped in sterile water.

Inoculations were made either in October, November or December. The number and size of the tumours were determined in the autumn, i.e. March, April or May.

Tomatoes. The plants were inoculated in the fourth internode above the cotyledons by means of a needle prick. Control plants were pricked with a sterile needle. The number of plants per treatment was eleven or twelve. Observations were made after a month. The inoculation experiments, both on rootstocks and tomatoes, were done in a glasshouse at relatively high temperatures. The tomato plants were inoculated in February-March when the temperatures varied between 19.5° and 35.5°C. These values were measured in a Stevenson screen, which was placed in the same glasshouse compartment.

Results and discussion

The results of the rootstock experiments are presented in Table 1.

The results of the inoculation experiments with tomato plants were as follows. No tumours were observed on the control plants nor on those inoculated with KGR. Small, medium and large tumours developed on tomato plants inoculated respectively with KGB, KGPe and KGD.

The results presented in Table 1 showed considerable differences in the susceptibility of rose rootstocks to crown gall. *R. multiflora*, 'Basye No. 3' and 'Manetti' were very susceptible, whereas 'ISU 60-5', 'Brooks', 'Clarke' and 'Welch' were more resistant.

Table 1. Susceptibility of different rose rootstocks to four isolates of *Agrobacterium tumefaciens*

Isolate	Inoculation method (see text)	Rose rootstocks						
		<i>R. multiflora</i>	'Clarke 1957'	'Brooks 48'	'ISU 60-5'	'Welch'	'Basye no. 3'	'Manetti'
KGD	T-cut	+	—	—	—		±	—
(from dahlia)	Crown	++			—			
KGR	T-cut	+++	—	—			++	++
(from a hybrid tea rose)								
KGB	T-cut	+++	—	—	—	+	+++	+++
(from Basye No. 3)	Crown	+++			—			
KGPe	T-cut	+++	+	±	+	+	+++	+++
(from peach)	Crown	+++	++	++	+			
Control	T-cut	—	—	—	—	—	—	—
	Crown	—	—	—	—			

- Not susceptible
 ± Doubtful, callouslike growth on some rootstocks
 + Slightly susceptible, less than 25% infected, small tumours
 ++ Moderately susceptible
 +++ Very susceptible, more than 75% infected, mostly large tumours

Tabel 1. Vatbaarheid van verschillende roze-onderstammen voor vier isolaten van *Agrobacterium tumefaciens*

None of the rootstocks tested was immune. In the first tests with 'Brooks', when only the T-cut inoculation method was used, 'Brooks' appeared to be highly resistant. On a few rootstocks only, a callouslike growth developed, but proper tumours were not observed. However, when 'Brooks' was inoculated at the crown rather large galls developed; in one experiment tumours developed on more than half of the inoculated plants. Crown gall was also found on 'Brooks' planted outdoors; in one case 6% of the plants examined were found to be infected. 'ISU 60-5' was apparently the most resistant of the rootstocks tested. Even when inoculated at the crown with the most virulent isolate, less than a quarter of the inoculated rootstocks developed tumours.

Table 1 also shows differences between the four isolates of *A. tumefaciens*, used in these experiments. Isolates KGR, KGB and KGPe from species of Rosaceae, probably differed in virulence only. However, isolate KGD apparently belongs to a race, different from that of the other isolates. It was more virulent on tomato than KGPe, but unlike KGPe the dahlia isolate was not virulent on 'Manetti' and 'Basye No. 3'. Wormald (1945) described four races of *A. tumefaciens* but no attempt was made in the above investigations to compare the characteristics of the South African isolates with those of the races found in Britain. To distinguish the races Wormald inoculated inter alia tomatoes, apples and raspberries, but did not mention cultivars or clones.

The existence of different races of *A. tumefaciens* implies the possibility that a race might be found, which is virulent to the rose rootstocks that were found resistant in the above experiments.

Samenvatting

Resistentie van roze-onderstammen tegen wortelknobbel (Agrobacterium tumefaciens)

Bij onderstammen van rozen, kunstmatig geïnoculeerd met *Agrobacterium tumefaciens* (Smith et Townsend) Conn., werden verschillen in resistentie tegen wortelknobbel gevonden. Het meest resistent was 'Iowa State University (ISU) 60-5', gevolgd door 'Brooks 48', 'Clarke 1957' en 'Welch'. Zeer vatbaar waren *Rosa multiflora*, *R. noisettiana* ('Manetti') en 'Basye No. 3'. De vier isolaten van *A. tumefaciens*, gebruikt voor de inoculaties, waren respectievelijk afkomstig van een *Dahlia* sp., *Rosa* spp. en *Prunus persica*. Het isolaat van *Dahlia* en de isolaten van *Rosa* en *Prunus* spp. behoorden tot twee verschillende fysiologische rassen. De vorming van tumoren was in sommige gevallen afhankelijk van de methode van inoculatie; inoculaties bij de stambasis waren meer succesvol dan in het midden van de stam.

References

- Dowson, W. J., 1957. Plant diseases due to bacteria. University Press, Cambridge.
- Massey, L. M., 1950. Crown Gall. Am. Rose A. 35: 145-153.
- Munnecke, D. E., Chandler, P. A. and Starr, M. P., 1963. Hairy root (*Agrobacterium rhizogenes*) of field roses. Phytopathology 53: 788-799.
- Wormald, H., 1945. Physiologic races of the crown gall organism in Britain. Trans. Br. mycol. Soc. 28: 134-146.