

## Short communication

# Susceptibility of five *Prunus* rootstocks to *Agrobacterium tumefaciens*

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**Abstract** Five *Prunus* rootstocks—GF677 (*P. persica* × *P. amygdalus*), Antafuel (*P. amygdalus* × *P. persica*), St. Julien 655/2 (*P. insititia* × *P. domestica*), Peach Seedling (*P. persica*), and Gisela 5 (*P. cerasus* × *P. canescens*)—were evaluated for their susceptibility to *Agrobacterium tumefaciens*. The results showed that no rootstock tested was immune. The St. Julien 655/2 rootstock was the most resistant of all tested, whereas no significant difference was observed between the other rootstocks. The level of susceptibility did not vary according to the strains used.

**Keywords** *Agrobacterium tumefaciens*; *Prunus*; rootstocks; strains; susceptibility

## INTRODUCTION

Crown gall, which is caused by *Agrobacterium tumefaciens* E.F. Smith & Town, is distributed worldwide and is responsible for nursery and field losses among a large variety of plants, especially stone fruit trees (Pinochet et al. 2002; Bliss et al. 1999; Aysan et al. 2004). No chemical pesticide is efficient against this bacterium, which multiplies in injured tissues causing a gall, which can occur any time during the plant's life. If the K84 strain of *A. radiobacter* can be used to protect plants against *A. tumefaciens* more-or-less successfully, it may be

possible to use genetic resistance by exploiting the variability in the genus *Prunus* as an alternative approach. Differences in virulence between *A. tumefaciens* strains have been found (Peluso et al. 2003) and this parameter should be considered when rootstocks are evaluated for their susceptibility. The main aims of this study were to test the susceptibility of the *Prunus* rootstocks GF677 (*P. persica* × *P. amygdalus*), Antafuel (*P. amygdalus* × *P. persica*), St. Julien 655/2 (*P. insititia* × *P. domestica*), Peach Seedling (*P. persica*), and Gisela 5 (*P. cerasus* × *P. canescens*) to *A. tumefaciens* and the virulence of three different strains isolated from *Prunus* species.

## MATERIALS AND METHODS

Three strains of *A. tumefaciens* were kindly supplied by the Benaki Phytopathological Institute Collection (BPIC 142 and 149 isolated from *P. dulcis*; BPIC 997 isolated from *P. domestica*). All bacteria were grown on the Medium 523 described by Schaad et al. (2001).

One-year-old GF677, St. Julien 655/2, Antafuel, Peach Seedling, and Gisela 5 rootstocks were bought from a commercial tissue culture station (Bitro Hellas). The trunk, crown, and roots of the plants were washed with sterile water to remove soil. The crown was superficially wounded and 10 roots of each plant were cut with sterile scissors and inoculated with one of the bacterial strains tested by immersion of the crown and roots for 15 min in suspensions of late-exponential-phase cells adjusted turbidometrically to densities of c. 10<sup>7</sup> colony forming units (cfu)/ml as described by Schaad et al. (2001). Inoculated plants were planted, without being rinsed, in pots containing 2 litres of sterile sand in a greenhouse at 23–29°C. They were periodically irrigated with sterile Hoagland solution. Results were collected by recording the total number and size of tumours, 2 months after inoculation.

Thirty pots for each rootstock were used, 10 for each *A. tumefaciens* strain. Also, there were 10 pots for each rootstock containing non-inoculated plants

**Table 1** Susceptibility of five *Prunus* rootstocks on different strains of *Agrobacterium tumefaciens*. Values are the means of two experiments; results were similar according to the Bartlett's test of homogeneity of variance, so data were combined. Treatment means were separated by using Least Significant Differences ( $P > 0.05$ ).

| Strains                             | Antafuel             |       |          | GF677            |          |      | Seedling            |     |     | St. Julien 655/2 |     |     | Gisela 5                |     |                              | Mean                    | LSD <sub>0.05</sub>          |
|-------------------------------------|----------------------|-------|----------|------------------|----------|------|---------------------|-----|-----|------------------|-----|-----|-------------------------|-----|------------------------------|-------------------------|------------------------------|
|                                     | <2                   | 2-4   | 4<       | <2               | 2-4      | 4<   | <2                  | 2-4 | 4<  | <2               | 2-4 | 4<  | <2                      | 2-4 | 4<                           |                         |                              |
| <b>Size of tumors (diam. in cm)</b> |                      |       |          |                  |          |      |                     |     |     |                  |     |     |                         |     |                              |                         |                              |
| BPIC 142                            | 1.3                  | 1.4   | 0        | 0.4              | 0.8      | 0.1  | 0.7                 | 0.3 | 0.5 | 0.2              | 0.3 | 0.6 | 1.2                     | 0.9 | 1.6                          | 0.7                     | 0.65 (for bacterial strains) |
| BPIC 149                            | 1.1                  | 0.5   | 3.6      | 1.2              | 1.8      | 1    | 0.2                 | 1.5 | 3.8 | 0.4              | 0.1 | 0.3 | 1.1                     | 1.2 | 0.9                          | 1.2                     |                              |
| BPIC 997                            | 0.8                  | 0.8   | 1.8      | 0.8              | 1.8      | 1.5  | 1                   | 2.6 | 1   | 0.1              | 0.5 | 0.5 | 1.2                     | 0.7 | 1                            | 1.1                     |                              |
| Mean                                | 1.1                  | 0.9   | 1.8      | 0.8              | 1.5      | 0.9  | 0.6                 | 1.5 | 1.8 | 0.2              | 0.3 | 0.5 | 1.2                     | 0.9 | 1.2                          | 0.75 (for interactions) |                              |
| LSD <sub>0.05</sub>                 | 0.6 (for rootstocks) |       |          |                  |          |      |                     |     |     |                  |     |     |                         |     |                              |                         |                              |
| <b>Total no. tumors</b>             |                      |       |          |                  |          |      |                     |     |     |                  |     |     |                         |     |                              |                         |                              |
| BPIC142                             | 2.7                  |       | 1.3      |                  | 1.5      |      | 1.1                 |     | 1.1 |                  | 3.7 |     | 2.1                     |     | 1.65 (for bacterial strains) |                         |                              |
| BPIC 149                            | 5.2                  |       | 4        |                  | 5.5      |      | 0.8                 |     | 0.8 |                  | 3.2 |     | 3.7                     |     |                              |                         |                              |
| BPIC 997                            | 3.4                  |       | 4.1      |                  | 4.6      |      | 1.1                 |     | 1.1 |                  | 2.9 |     | 3.2                     |     |                              |                         |                              |
| Mean                                | 3.8                  |       | 3.1      |                  | 3.9      |      | 1.0                 |     | 1.0 |                  | 3.3 |     | 1.35 (for interactions) |     |                              |                         |                              |
| LSD <sub>0.05</sub>                 | 1.1 (for rootstocks) |       |          |                  |          |      |                     |     |     |                  |     |     |                         |     |                              |                         |                              |
| Strains                             | Antafuel             | GF677 | Seedling | St. Julien 655/2 | Gisela 5 | Mean | LSD <sub>0.95</sub> |     |     |                  |     |     |                         |     |                              |                         |                              |

on which no tumours developed. All experiments were conducted twice. Data were analysed by using Analysis of Variance. Experiments were similar according to Bartlett's test and data were combined. Treatment means were separated by using Least Significant Difference ( $P > 0.05$ ).

## RESULTS AND DISCUSSION

All *A. tumefaciens* strains used were pathogenic to the rootstocks tested (Table 1). The rootstocks GF677, Peach Seedling, Antafuel, and Gisela 5 challenged against *A. tumefaciens* were susceptible. The results are in good agreement with previous work. These showed that clones of *P. cerasifera* (Pierronnet & Salesses 1996) and the peach rootstocks GF677 and Peach Seedling (Zoina & Raio 1999) were susceptible to *A. tumefaciens*. The rootstock St. Julien 655/2, a hybrid of *P. insititia* × *P. domestica*, was moderately resistant to *A. tumefaciens* as in previous work with other plum rootstocks such as M.r.S.2/6 (Zoina & Raio 1999) and Torinel (Pinochet et al. 2002). Bliss et al. (2003) evaluated 20 *Prunus* species for their susceptibility to *A. tumefaciens* and found that resistant plants were identified in *P. insititia*. The level of susceptibility was statistically similar for all strains used although interactions showed different virulence of strains for some of the rootstocks individually (GF677, Peach Seedling, Antafuel) (Table 1). According to Benjama et al. (2002) the level of susceptibility of stone fruit rootstocks varied according to the strains used.

The virulence of *A. tumefaciens* strains tested suggests that this pathogen is a serious threat to *Prunus* trees grafted on GF677, St. Julien 655/2, Gisela 5, Peach Seedling, and Antafuel rootstocks. Although none of the rootstocks tested is immune or resistant to *A. tumefaciens*, the rootstock St. Julien 655/2 should be used in fields with high disease pressure.

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