Breeding for Resistance in *Zantedeschia* spp. (Araceae) against Soft Rot Caused by *Erwinia carotovora* ssp. *carotovora*

Ronald C. Snijder and Jaap M. van Tuyl
Plant Research International
P.O. Box 16
6700 AA Wageningen
The Netherlands
Tel: +31-317-477329
Fax: +31-317-418094.
E-mail: R.C.Snijder@plant.wag-ur.nl

**Keywords:** Breeding, disease-resistance, *Erwinia, Zantedeschia*

**Abstract**

A huge problem in growing *Zantedeschia* (calla or arum lily) is the occurrence of soft rot caused by *Erwinia carotovora* ssp. *carotovora*. At Plant Research International, a research programme was started for finding and developing *Erwinia*-resistant genotypes of *Zantedeschia*. Hereto, a disease test was developed with which the level of resistance of clones, and therefore the variation in resistance within the genus, can be screened. The results show clearly, what is already known from practice, that *Z. aethiopica* is more resistant to the soft rot than species of section *Aestivae*. Interspecific hybrids between genotypes of the *Aethiopica* and the *Aestivae* section obtained using embryo rescue techniques deliver up till now only albino plants. An in vitro disease test was developed for screening these heterotrophic albino hybrids. There is variation for susceptibility/resistance within section *Aestivae*. Finding the genetics behind the resistance within this section is the next step in the programme.

**INTRODUCTION**

*Zantedeschia* Sprengel (Araceae), also 'calla lily' or 'arum lily', is a genus of about eight species, all from southern Africa (Letty, 1973; Singh et al., 1996). *Zantedeschia*, is growing as a crop in the Netherlands. In 1998, a 68% and in 1999, a 29% growth to 35 million guilders was experienced for cut flowers (VBN, 2000). 50% of flowering size calla tubers are produced in the United States, The Netherlands and New Zealand account for 45%. In the United States, more than 95% of the tubers are grown for flowering potted plants, in Europe, 80-85% are used for cut flowers (Kuehny, 2000).

Two groups of *Zantedeschia* can be distinguished. The first group is made up of cultivars of *Z. aethiopica* (also white calla), further in this paper called 'Aethiopica-group'. The second is made up of cultivars of the so-called 'coloured callas'. These are cultivars descending from crosses between species of the botanical section *Aestivae* (Singh et al., 1996), further called the 'Aestivae-group'.

Bacterial soft rot caused by *Erwinia carotovora* subsp. *carotovora* is a major problem in growing and storage of *Zantedeschia*. This anaerobic pathogen causes maceration and rotting of parenchymatous tissue of the affected organ, resulting in loss of the whole plant (Perombelon and Kelman, 1980). Cultural practices can partly decrease the problem (Wright, 1994). The use of resistant cultivars is another obvious approach in overcoming problems caused by soft rot.

No publications are known on the variation in resistance against soft rot caused by *Erwinia carotovora subsp. carotovora*, present in cultivars and wild accessions of *Zantedeschia*. But it is generally accepted that *Z. aethiopica* is much less susceptible than genotypes from section *Aestivae*. Therefore, hybridisation of genotypes from the Aethiopica-group and the Aestivae-group seems an obvious method for obtaining resistant genotypes. However, huge breeding barriers exist between the two groups (Yao et al., 1994; 1995). Therefore, besides hybridising the two groups, exploitation of
resistance that is already present in the Aestivae-group, could be a possibility. However, no studies are known on the variation within the Aestivae-group and the genetics behind the resistance mechanism in Zantedeschia.

This paper describes the research that has been started at Plant Research International (the result of the merger of CPRO, AB and IPO) for creating genotypes of Zantedeschia, resistant to soft rot caused by Erwinia carotovora subsp. carotovora.

MATERIALS AND METHODS

Material
All plant material (see Table 1) was obtained from commercial sources. Aestivae genotypes were T2-material. The used strain of Erwinia carotovora subsp. carotovora, PD1784, was isolated by the Dutch Plant health service (Plantenziektenkundige Dienst).

Disease Tests
In order to evaluate the level of resistance of clones and seedlings, proper disease tests are needed. The test to evaluate clones was modified from Allefs et al. (1993), the modifications being the inoculation of 8 whole Zantedeschia tubers with 10µl of bacterium-suspension (1*10^7 cfu/ml) and scoring 6 days after inoculation. The response variable for this tuber test, √{(W1-W2)/W1}, was adopted from Lojkowska and Kelman (1994). This test was done twice on all the genotypes mentioned in Table 1, except for 99103-1. Hybrids between the Aethiopica and Aestivae cultivars were obtained as described by Yao et al. (1995).

Vitro Propagation
Vitro plants were propagated using a method that is a slight modification of the method described by Cohen (1981), the modifications being the use of 8% bacteriological agar and 0.5 mg/l BA. After transferring to water agar medium, vitro plants were inoculated in vitro by dripping 6 µl of inoculum (1*10^5 cfu/ml) on the cut surface at the top of the biggest petiole. After three days of incubation in the dark at 20°C, the length of infected petiole tissue was measured.

RESULTS AND DISCUSSION
The tuber test revealed that the Aestivae genotypes mentioned in Table 1 have different levels of resistance. Figure 1 gives the results of the tuber test.

99008, An Aethiopica genotype, does not show any sign of infection after inoculation. Three genotypes score susceptible, namely 99015, 99016 and 99017. Genotypes 99011, 99012 and 99014 score less susceptible. These results prove that this Z. aethiopica is more resistant to soft rot caused by Erwinia carotovora subsp. carotovora than the Aestivae genotypes. There is quite some variation within the Aestivae group for susceptibility. Therefore, these Aestivae genotypes have been used to construct a diallel crossing population. Whether the difference in levels of resistance can be genetically transferred to their offspring and in which patterns, is the next step in the research programme. For this, besides the diallel population, a discriminative disease test is needed that is not destructive and uses only little tissue.

In order to determine the level of resistance of Aethiopica x Aestivae hybrids, a disease test in vitro was developed. These plants are namely albino and strictly grow heterotroph. Three days after inoculation it was clear that the albinos were highly susceptible, even more than vitro plants of 99016. Relative scores of 99008, 99014 and 99016 were similar to results of the tuber test. The high susceptible score of the albino was not expected, the resistant Aethiopica being its mother. This does not necessarily mean that Aethiopica cannot transfer its resistance to its offspring. It is also possible that their high susceptibility is due to the absence of leaf-green and the low vigour of the albino plants.
ACKNOWLEDGEMENTS

The work of Martijn van Paasen is appreciated by the authors. This research is co-financed by the Dutch Commodity Board for Horticultural Products (PT, Productschap Tuinbouw) and six Zantedeschia-related companies.

Literature Cited


Tables

Table 1. Plant material used in disease tests and as parents in constructing the Aestivae diallel population (only 99011-99017)

<table>
<thead>
<tr>
<th>PRI-Number</th>
<th>Genotype</th>
<th>Source</th>
<th>Test</th>
</tr>
</thead>
<tbody>
<tr>
<td>99008</td>
<td>Aethiopica</td>
<td>Commercial</td>
<td>Tuber</td>
</tr>
<tr>
<td>99011</td>
<td>Aestivae</td>
<td>Commercial</td>
<td>Tuber</td>
</tr>
<tr>
<td>99012</td>
<td>Aestivae</td>
<td>Commercial</td>
<td>Tuber</td>
</tr>
<tr>
<td>99014</td>
<td>Aestivae</td>
<td>Commercial</td>
<td>Tuber, vitro</td>
</tr>
<tr>
<td>99015</td>
<td>Aestivae</td>
<td>Commercial</td>
<td>Tuber</td>
</tr>
<tr>
<td>99016</td>
<td>Aestivae</td>
<td>Commercial</td>
<td>Tuber, vitro</td>
</tr>
<tr>
<td>99017</td>
<td>Aestivae</td>
<td>Commercial</td>
<td>Tuber</td>
</tr>
<tr>
<td>99103-1</td>
<td>Aethiopica x Aestivae</td>
<td>Plant Research International</td>
<td>Vitro</td>
</tr>
</tbody>
</table>

265
Fig. 1. The values of $\sqrt{\left(\frac{W1-W2}{W1}\right)}$ for six genotypes from the Aestivae group and one of the Aethiopica group. The black line represents the least significant difference (lsd; $\alpha=0.05$). The Y-axis starts at -0.1 to show that 99008 scores 0.