

Development of two biotests for the identification of *Aphanomyces cochlioides* resistance in sugar beet

Introduction

Aphanomyces cochlioides belongs to the economically most important pathogens in sugar beet production worldwide. The soil borne oomycete causes damping-off in seedlings as well as scab and *Aphanomyces* root rot in mature beets. Fungicidal control is only possible in relation to damping-off by seed coating but cannot prevent later infection and symptom development on older plants. Thus, the use of resistant varieties is currently the only control method to avoid severe losses. However, the selection under natural conditions is subject to high variation because the infection depends on weather conditions and varies between different years. Therefore, two different biotests for the detection of genotypic differences in the susceptibility to damping-off as well as *Aphanomyces* root rot were developed in the greenhouse.

Plant Material and Inoculum Preparation

Non-treated seed of breeding lines from Strube Research GmbH & Co. KG (Söllingen, Germany) differing in their susceptibility to *A. cochlioides* were sown in the greenhouse and inoculated either in the seedling stage or at 5-6 weeks. Five-day-old *A. cochlioides* cultures grown on PDA were used for the inoculation of oat meal broth. Flasks were incubated at 20°C in the dark without shaking. After six weeks, mycelial mats were homogenized in a blender and the oospore concentration in the solution was determined and adjusted with sterile water. Clay granules were mixed in a relation of 1:1 (w/v) with the oospore solution and dried under a sterile bench for three days. Inoculum bound to clay granules can be stored up to three month at room temperature without loss of pathogenicity.

Biotest for Damping-off Resistance

Seven-day-old seedlings were transplanted in multipot trays and kept in the greenhouse at 24/18°C (day/night). Each pot of the inoculated treatment contained 70 g of standard potting soil and clay granules corresponding to 300 oospores/g soil. Clay granules and soil were thoroughly mixed before planting. Control plants were transplanted in separate trays and mock-inoculated with non-treated clay granules. After inoculation, plants were subirrigated once daily. At 21 days post inoculation (dpi), symptoms were rated according to diseased root and hypocotyl surface.

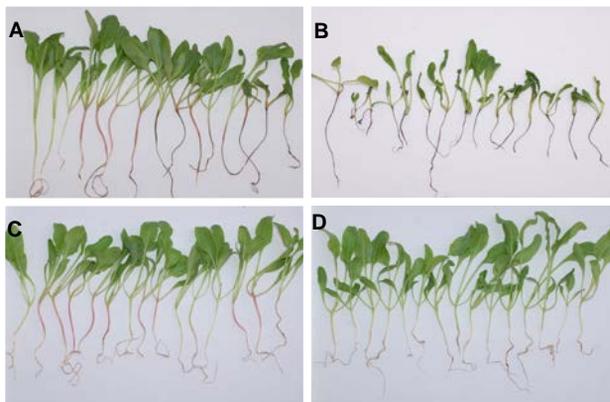


Figure 1: Damping-off symptoms in sugar beet seedlings of genotype BL1 (A, C) and BL2 (B, D). Top: inoculated with 300 oospores/g soil, bottom: control.

Conclusions

Both tests allowed the differentiation between more and less resistant genotypes. However, as *Aphanomyces* resistance in sugar beet is quantitative and in some genotypes a huge variation was observed, environmental conditions have to be standardized further in order to reduce the number of outliers. Subirrigation proved to be the method of choice as much more homogenous and reliable results were obtained.

Biotest for *Aphanomyces* Root Rot Resistance

Five- to 6-week-old plants were transplanted in pots containing 1 kg of standard potting soil and well incorporated clay granules corresponding to 600 oospores/g soil. Control plants were mock-inoculated with non-treated clay granules. Plants were kept in the greenhouse at 24/18°C and placed in water filled saucers for the first week after inoculation. Subsequently, plants were subirrigated once daily. At 30 dpi, beets were harvested, topped and rated for surface symptoms. Depending on the genotype, beets displayed either more scab or rot symptoms.



Figure 2: *Aphanomyces* root symptoms after inoculation of 6-week-old sugar beet in genotype BL2 (A) and BL1 (B) at 30 dpi.

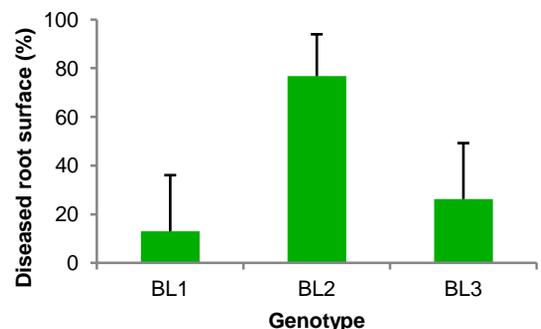


Figure 3: *Aphanomyces* root symptoms after inoculation of 6-week-old sugar beet in three different genotypes at 30 dpi (n=30).