

Hunziker et al. Supplementary Figures S1-S4

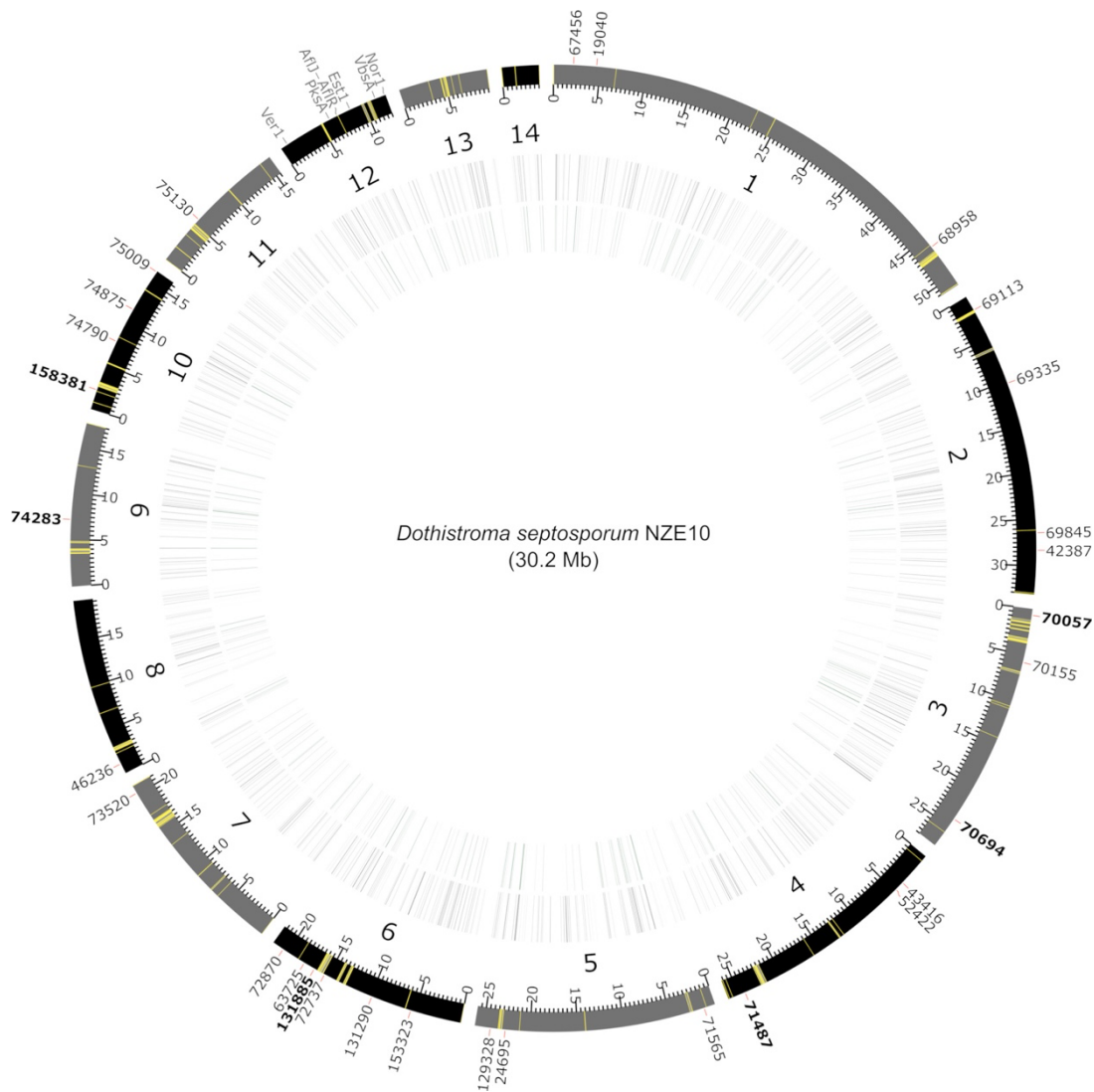


Figure S1: Locations of the 30 cloned *Dothistroma septosporum* candidate effector (DsCE) genes in the NZE10 genome. The 14 chromosome-level scaffolds of *D. septosporum* NZE10¹ are represented by the outer bars. Each minor tick represents 5,000 bp from the start of the scaffold; yellow stripes indicate the location of curated repetitive elements >200 bp in length². Outer numbers are protein IDs corresponding to the 30 *DsCEs*, with cell death inducing *DsCEs* in bold font. For reference, the positions of the dothistromin biosynthesis genes¹ are also shown (chromosome 12, grey labels). Within the inner rings, grey bars represent the 875 genes encoding putatively secreted proteins, and green bars (innermost) represent the 397 *in planta*-expressed (>50 Reads Per Million per Kilobase) secreted proteins. The figure was created using CIRCOS (<http://circos.ca/software/>;³).

References for Figure S1:

- 1 de Wit, P. J. G. M. *et al.* The genomes of the fungal plant pathogens *Cladosporium fulvum* and *Dothistroma septosporum* reveal adaptation to different hosts and lifestyles but also signatures of common ancestry. *PLoS Genetics* **8**, e1003088 (2012).
- 2 Ohm, R. A. *et al.* Diverse lifestyles and strategies of plant pathogenesis encoded in the genomes of eighteen Dothideomycetes fungi. *PLoS Pathogens* **8**, e1003037-e1003037 (2012).
- 3 Krzywinski, M. *et al.* Circos: an information aesthetic for comparative genomics. *Genome Research* **19**, 1639-1645 (2009).

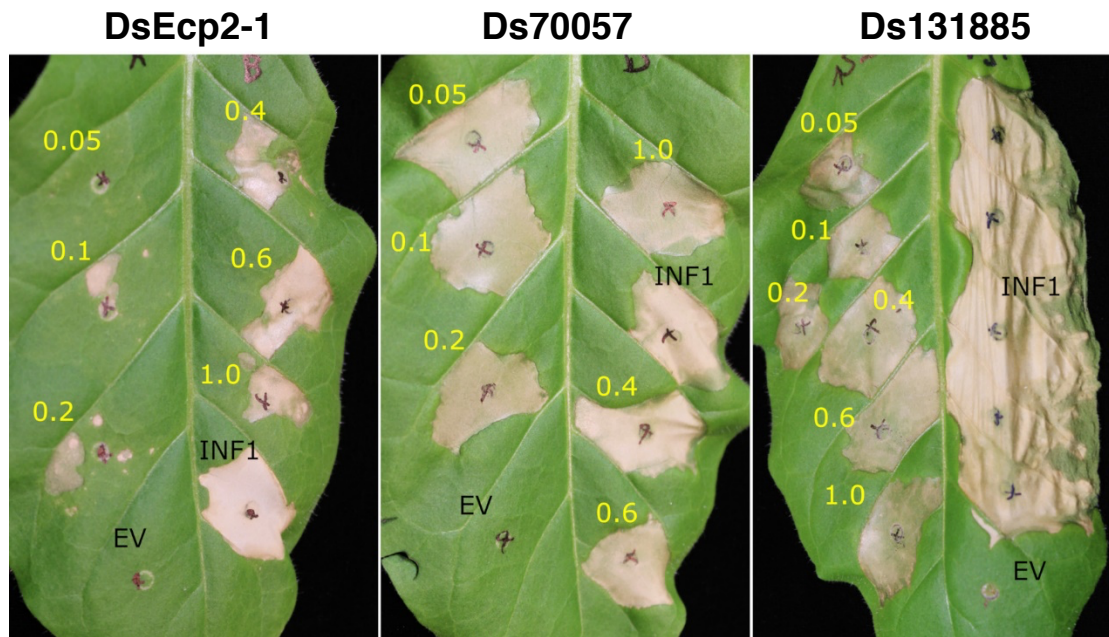


Figure S2: Optical density range trial of *Agrobacterium tumefaciens* cultures. Concentration thresholds are indicated for cell death triggering by DsEcp2-1, but not Ds70057 and Ds131885. The yellow numbers show the used culture OD₆₀₀; the positive (INF1) and negative (EV) controls were infiltrated at an OD₆₀₀ of 0.6. In the Ds131885 panel, INF1 was trialled at 0.05 to 0.6 (top to bottom).

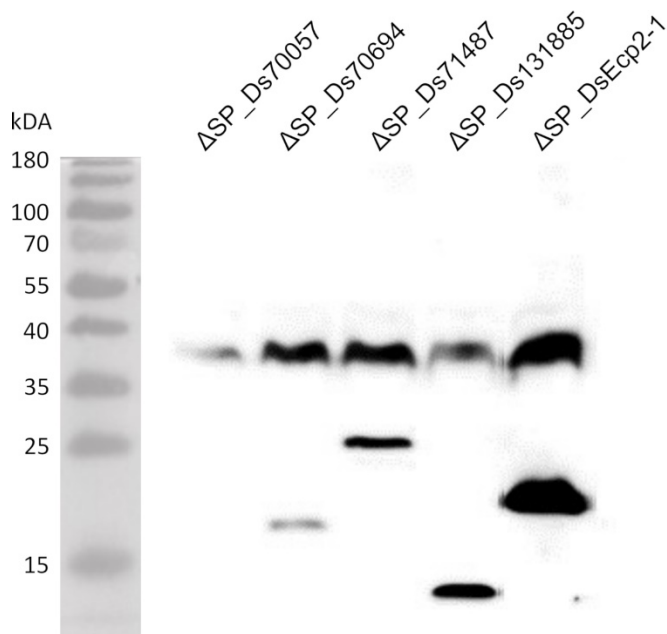
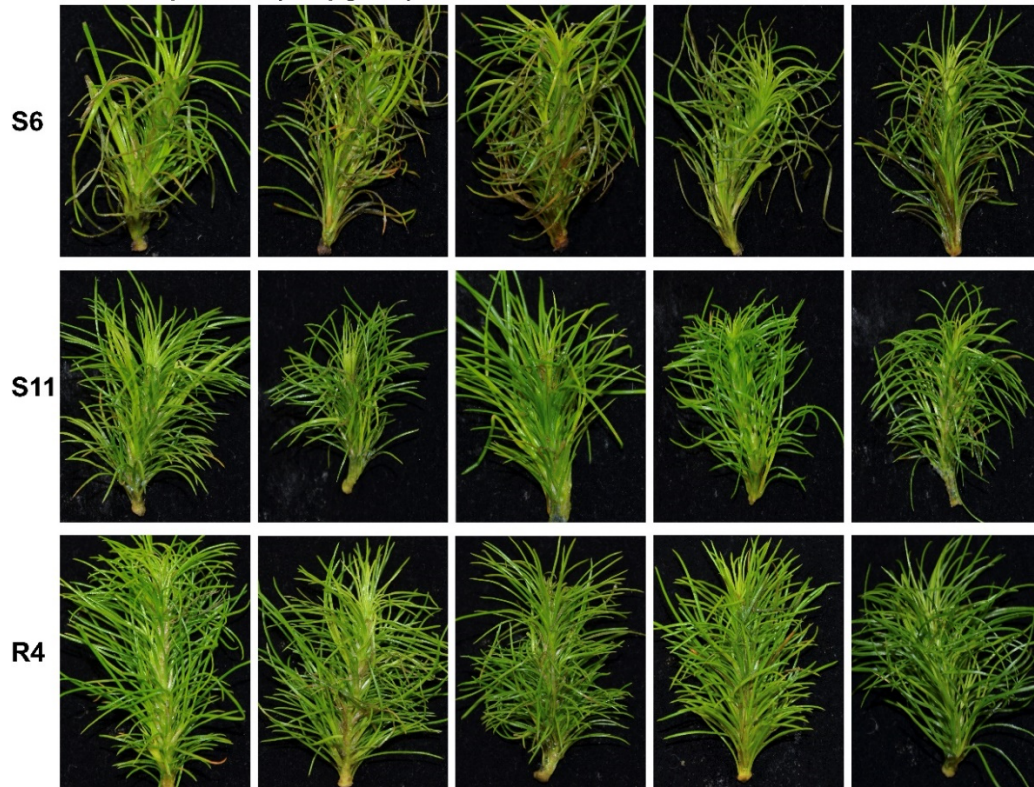


Figure S3: Western blots of *Dothistroma septosporum* candidate effector proteins. Western blots showed that DsCEs triggering cell death in *Nicotiana benthamiana* were expressed in the plant tissue regardless of the presence of a secretion signal peptide and absence of a cell death response (constructs with deleted signal peptide sequences are shown). Immuno-detection was based on primary anti-FLAG antibody.

(a)

BcSSP2 purified (1.3 $\mu\text{g/mL}$)



(b)

Ds70057 purified (21 $\mu\text{g/mL}$)

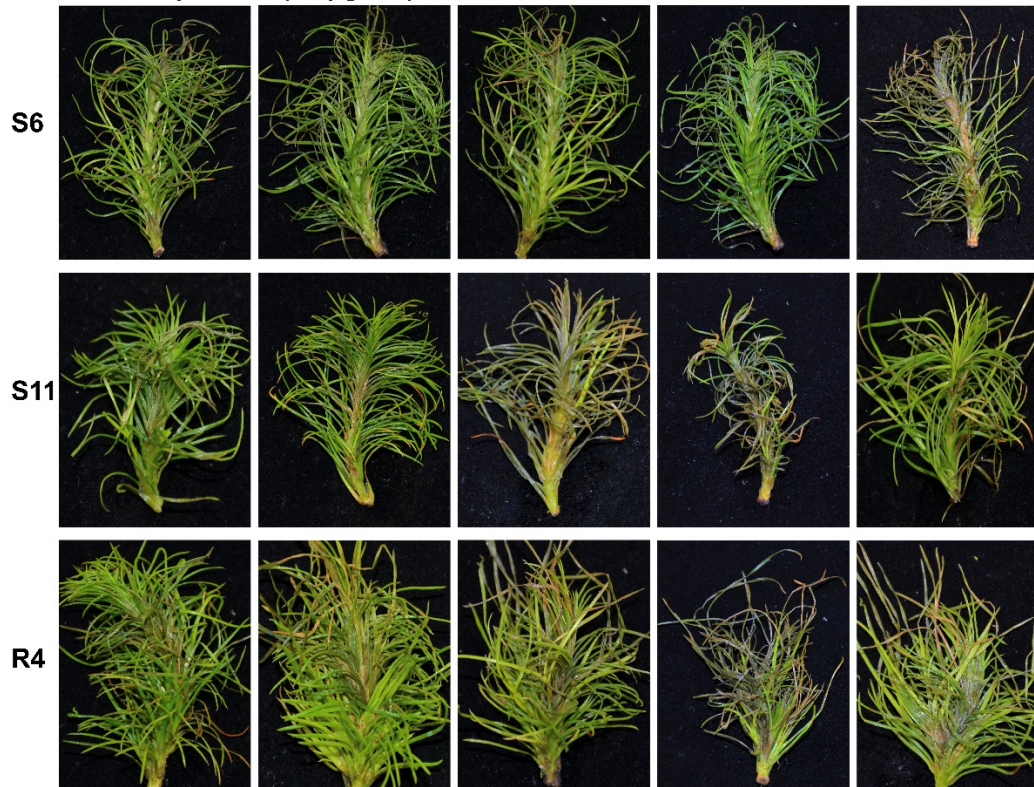


Figure S4: Replicates of *Pinus radiata* shoot tissue infiltrated with candidate effector proteins. BcSSP2 (a) and Ds70057 (b) were produced by heterologous expression in *Pichia pastoris*. Photos were taken 7 days after infiltration.