

## Qualitative PCR diagnostics of *A. solani* and *A. alternata*

TUM, Wissenschaftszentrum Weihenstephan, Andrea Volz

June 2014

Genomic DNA of *A. solani* isolates was extracted from mycelia cultivated on V8 medium under near-UV light for 14 days at 21°C. Mycelium and spores were carefully scraped off with a spatula and ground in liquid nitrogen. Genomic DNA extraction was carried out using CTAB. The DNA was resuspended in sterile H<sub>2</sub>O.

Cycling conditions, qualitative PCR, *A. solani* + *A. alternata*

Denaturation	94°C	3 min	
Denaturation	94°C	10 s	} 35x
Annealing	60°C	30 s	
Elongation	72°C	30 s	
	72°C	5 min	

Mastermix, qualitative PCR, *A. solani* + *A. alternata*

	µl
H <sub>2</sub> O	15,3
10x buffer with 15mM MgCl <sub>2</sub>	2,5
25mM MgCl <sub>2</sub>	2
10 µM Primer F	1
10 µM Primer R	1
10 mM dNTPs	1
Taq	0,16
sample	2
total	25

Primer sets for qualitative PCR

*A. solani*

<i>A. solani</i> F	CAC CAC AAG GAC CAA CCC A	355	
<i>A. solani</i> R	TGG GGC TGG AAG AGA GCG		

---

A. alternata

Aalt F1	GCG GGC TGG AAC CTC TC		
Aalt R1,1	AGA CCT TTG CTG ATA GAG AGT	443 bp	

Technische Universität München  
Lehrstuhl für Phytopathologie  
M. Sc. Andrea Volz  
Emil-Ramann-Str. 2  
85350 Freising  
Germany  
Tel.: 00498161713737  
[<a.backhaus@wzw.tum.de>](mailto:a.backhaus@wzw.tum.de)