ViTEST

Microneedle DNA Extraction 8/10-8/20

Extraction method testing

EPFL iGEM

Saturday, 10 August 2019

Title: Amplifying different The CE using different amounts of TE buffer to extract the DNA extracted with the MN patch and using the Gblock as reference

Aim: Find the optimal concentration of EB for the extraction of DNA.

Participants: Hana, Stefania

Protocols: Gel electrophoresis, PCR

Notes:

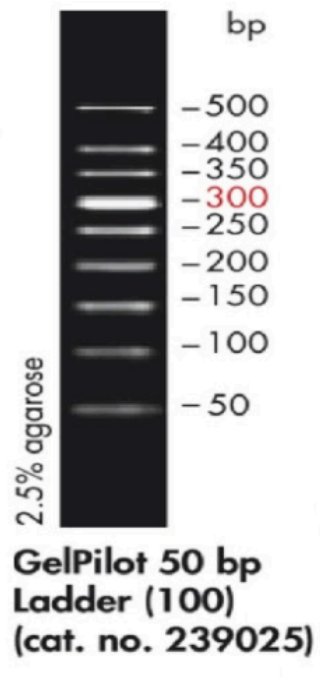
Gel electrophoresis

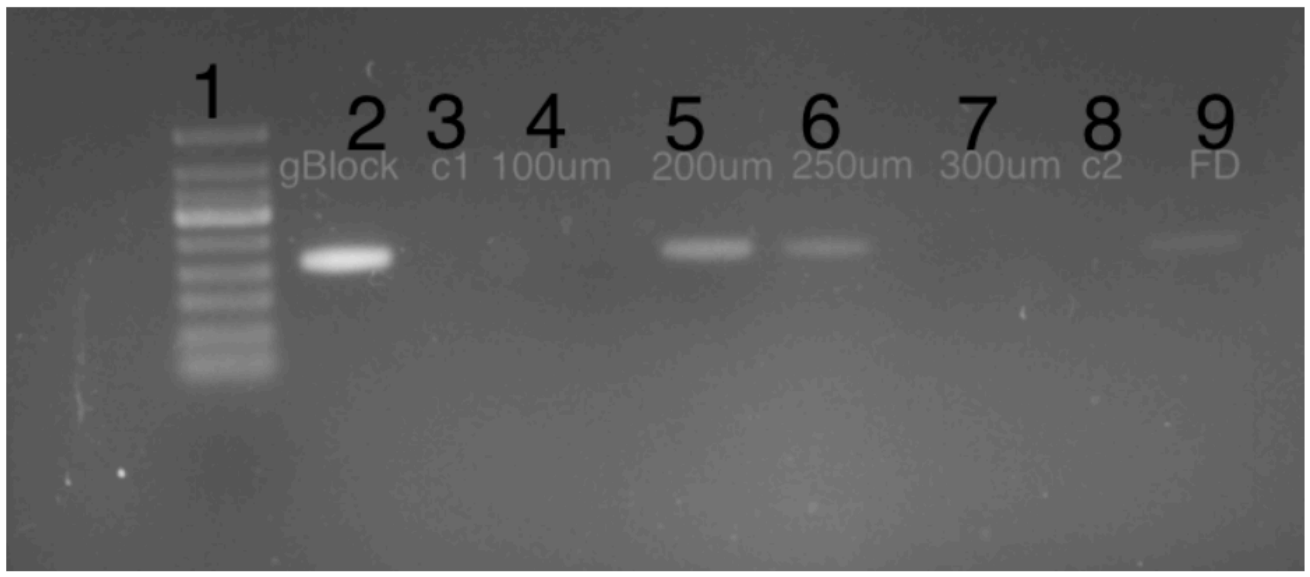
2% agarose gel, midori green gel stain and gel pilot 6X loading dye

5 µL GelPilot 50 bp ladder (Qiagen)

All amplifications were performed using 1 µL of DNA , 25 µL of Q5 master mix and 2,5 µL of each primer in a 50 µL PCR reaction.

Results:





Lane	Fwd primer	Rev primer	DNA	Comment
1	X	X	X	Ladder
2	FD_T7_F1	FD_R1	gBlock	control
3	X	X	gBlock	control
4	FD_T7_F1	FD_R1	DNA + TE buffer standard volume (100um)	We must have messed something up here cause it the highest concentration and we don't have a signal.
5	FD_T7_F1	FD_R1	DNA + TE buffer larger volume (200um)	we can detect the CE using 200 µLof TE buffer
6	FD_T7_F1	FD_R1	DNA + TE buffer larger volume (250um)	We can still detect the CE using 200 µLof TE buffer though we have a lower signal
7	FD_T7_F1	FD_R1	DNA + TE buffer larger volume (300um)	We can no longer detect the CE using 300 µLof TE buffer though we have a lower signal
8	X	X	DNA 001 (diagnosed with FD)	control
9	FD_T7_F1	FD_R1	DNA 001 (Diagnosed with FD)	we can also detect CE from the FD affected sample that we have extracted from the field

Type to enter text

Conclusion:

We can keep a volume of up to 250 um of TE buffer during extraction.

We successfully extracted DNA from contaminated leaves.

Tuesday, 20 August 2019

Title: Nanodrop

Aim: Verify correct amplification of DNA using microneedles

Participants: Hana, Stefania

Protocols: Nanodrop UV absorption spectra, with gBlock amplificatiom, MN patch amplification, and control

Notes: gBlock: green,
MN: red,
control: blue