

Cutting DNA protocol

Aims: to cut DNA into smaller fragments. The cut is nonspecific, being an excellent example of contrast with the specificity of cas9/crispr.

Materials

- DNA solution (100ng/ul)
- Nuclease free water
- DNase I Reaction Buffer (10X; NEB#M0303S)
- DNAse I (2U/ul; NEB#M0303S)

Mix for Dna digestion:

2ul of DNA solution (100ng/ul) 1ul of DNase I Reaction Buffer 0,5ul of DNA 6,5ul Nuclease free water

Mix for Undigested Control:

2ul of DNA solution (100ng/ul) 1ul of DNase I Reaction Buffer 7ul Nuclease free water

Incubate at 37°C for 15 minutes.

Run the solution in an agarose gel.

Results

You should see a clear band in the Mix for Undigested Control (the molecular weight depends on the DNA fragment size) and a smear with a very low molecular weight in the Mix for DNA digestion, that represents the very small fragments of randomly cut DNA. Note: The smear might be very faint and hard to visualize in the gel.

