Generation of DNA fragments by sonication

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Abstract

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Protocol

Step 1.
Generation of DNA fragments by sonication is performed by placing a microcentrifuge tube containing the buffered DNA sample into an ice-water bath in a cup-horn sonicator.

Step 2.
Sonication is conducted for a varying number of 10 s bursts using maximum output and continuous power.

Step 3.
Exact conditions for sonication should be empirically determined for a given DNA sample before a preparative sonication is performed.

Step 4.
Typically, 100 µg DNA in TE buffer is split into 10 aliquots of 35 µL.

Step 5.
5 aliquots are subjected to sonication for increasing numbers of 10 s bursts.

Step 6.
Aliquots from each time point are run on an agarose gel to determine optimal-sized DNA fragments (1–4 kb).

Step 7.
Once optimal sonication conditions are determined, the remaining 5 aliquots (approximately 8 µg) are sonicated according to those predetermined conditions.

NOTES
Declan Schroeder 12 Oct 2015
DNA can be blunt-ended and size-selected prior to downstream cloning.