

## Quick Guide: Shearing Genomic DNA with PIXUL™

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This Quick Guide provides instructions for users fragmenting gDNA with a PIXUL Multi-Sample Sonicator using the PIXUL gDNA Shearing Kit (cat. no. 53131). PIXUL sonication is compatible with a wide range of purified genomic DNA (500 ng – 20 µg) or cells (100,000 – 1,000,000) with no changes in sonication efficiency or processing time.

**IMPORTANT:** PIXUL sonication requires using PIXUL 96-well round bottom plates (cat. no. 53139). Using any other plates may result in inefficient sonication, may damage the instrument transducers, and will void the instrument warranty.

### Buffer Preparation Before Starting

80% Ethanol: 4.8 mL 100% ethanol + 1.2 mL nuclease-free water

### Sample Preparation Notes

- If sonicating purified gDNA in water, add concentrated Tris-HCl pH 8.0 to a final concentration of 10 mM Tris-HCl.
- If sonicating cells growing directly on the PIXUL 96-well plate, aspirate media, wash with 1X PBS, and add 100 µL Cell Shearing Buffer.
- If sonicating cells grown in a different plate or dish, resuspend the desired number of cells in 100 µL Cell Shearing Buffer and add to wells in the PIXUL 96-well plate (cat. no. 53139).

### PIXUL Sonication of gDNA

1. Add 100 µL of purified genomic DNA or cells in Cell Shearing Buffer to wells in the PIXUL 96-well plate (cat. no. 53139) and seal the plate with a PIXUL plate seal.  
**NOTE:** All wells lacking sample in the columns being sonicated **MUST** be filled with liquid (water, buffer, etc.) prior to starting the run.
2. Turn on and run the instrument as specified in the PIXUL Multi-Sample Sonicator User Manual or Quick Guide.
3. We recommend using the following parameter specifications as a starting point, and adjusting Process Time to optimize for your specific sample and application:

Sonication Parameter	Setting
Pulse [N]	50
PRF [kHz]	1.00
Process Time [min]	36:00
Burst Rate [Hz]	20.00

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### DNA Purification Using SPRI Beads

1. Add 0.5 µL RNase A to each well and incubate for 30 minutes at 37 °C.
2. Add 5 µL Proteinase K to each well and incubate for 30 minutes at 55 °C.
3. Add 120 µL (1.2 volumes) of SPRI beads to each well, pipette the bead-lysate mixture 10 times to mix, and incubate for 5 minutes at room temperature.
4. Place plate on a magnet for 5 minutes to collect beads, and then aspirate supernatant.
5. Keeping the plate on the magnet, add 180 µL of 80% ethanol and incubate for 30 seconds at room temperature.
6. Aspirate supernatant and perform a second wash with 180 µL of 80% ethanol.
7. Completely aspirate the supernatant and air dry the beads until they are no longer shiny (2-5 minutes).
8. Remove plate from the magnet and elute DNA by adding 50 µL of Low EDTA TE.
9. Pipette the beads 10 times and incubate for 3 minutes at room temperature.
10. Place tube or plate back on the magnet for 5 minutes to collect beads.
11. Carefully remove eluted DNA in the supernatant and transfer to a new tube or plate.

### Analyze Sonicated DNA

Analyze gDNA yield using a spectrophotometric (Nanodrop) or fluorometric (Qubit) method of your choice and analyze fragmentation efficiency using either agarose gel electrophoresis, Agilent TapeStation, or Agilent Fragment Analyzer. For most applications, we recommend shearing DNA to 200-600 bp.

### Technical Support

If you need assistance at any time, please contact Active Motif Technical Support at [tech\\_service@activemotif.com](mailto:tech_service@activemotif.com).