

Isolation of Genomic DNA from Mouse Tails on the Maxwell® RSC

Purify genomic DNA from mouse tail on Maxwell® RSC Instrument with Maxwell® Tissue DNA Kit using enzymatic or manual homogenization.

Kit: Maxwell® RSC Tissue DNA Kit (Cat.# AS1610)

Sample Type(s): Mouse tail clips

Input: Up to 10mg mouse tail tissue

Materials Required:

- Maxwell® RSC Instrument (Cat.# AS4500)
- Maxwell® RSC Tissue DNA Kit (Cat.# AS1610)
- Enzymatic Protocol
 - Tissue Lysis Buffer (Cat.# A5091)
 - Proteinase K, (Cat.# MC5005)
 - RNase A (Cat.# A7973) Optional
- Manual Homogenization Protocol
 - pestle homogenizer
 - RNase A (Cat.# A7973) Optional

This protocol was developed by Promega Applications Scientists and is intended for research use only.

Users are responsible for determining suitability of the protocol for their application.

For further information, see Technical Manual TM476, available at:
www.promega.com/protocols

or contact Technical Services at:
techserv@promega.com

Protocol:

Enzymatic Protocol:

1. Place mouse tail snips in a 1.5ml tube.
2. Add 200µl of Tissue Lysis Buffer, 20µl of Proteinase K and 20µl of RNase A (optional).
3. Incubate at 56°C for 1 hour.
4. Spin at room temperature for 2 minutes at $\geq 16,000 \times g$
5. Prepare cartridges as described in Section 4.B of the Technical Manual (TM476).
6. Carefully transfer only the supernatant to well #1 of the prepared Maxwell® cartridge and mix into the lysis buffer in well #1 by pipetting 10 times.
7. Run samples on the Maxwell® RSC using the Tissue DNA protocol.

Manual Homogenization Protocol:

1. Place mouse tail snips in a 1.5ml tube.
2. Add 80µl of 1X TE and 20µl of RNase A (optional).
3. Homogenize tail snips using a pestle.
4. Prepare cartridges as described in Section 4.B of the Technical Manual (TM476).
5. Transfer homogenized sample (as well as non-homogenized tissue) to well #1 of the prepared Maxwell® cartridge and mix into the lysis buffer in well #1 by pipetting 10 times.
6. Run samples on the Maxwell® RSC using the Tissue DNA protocol.

Results:

DNA was isolated from mouse tail clippings using either the enzymatic or manual homogenization methods listed above. Performance of these two protocols is shown below.

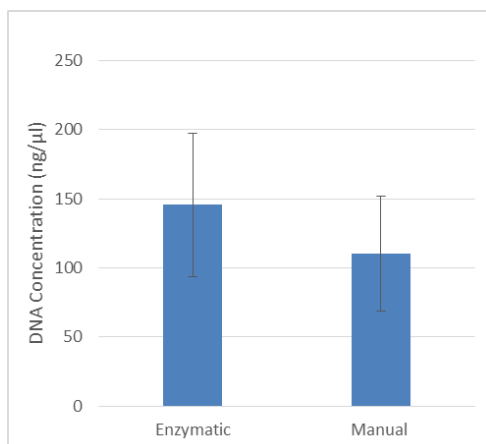


Figure 1. Concentration of DNA isolated from mouse tails. DNA concentration was measured using QuantiFluor® ONE dsDNA System (Cat.# E4871) on a Quantus™ Fluorometer (Cat.# E6150). Shown is the average \pm standard deviation for at least 7 replicates for each condition.

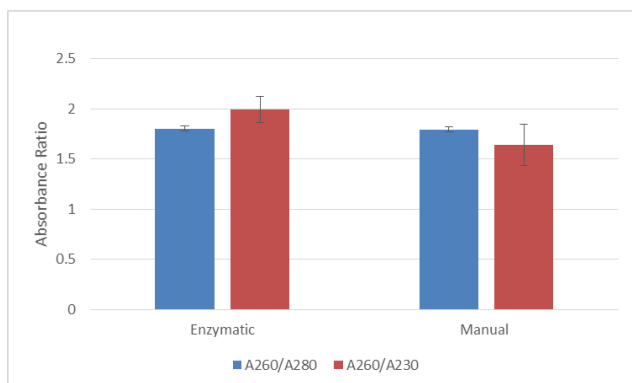


Figure 2. Purity of DNA isolated from mouse tails. Absorbance was read at 230, 260 and 280nm on a NanoDrop Spectrophotometer, and ratios were calculated to assess purity.

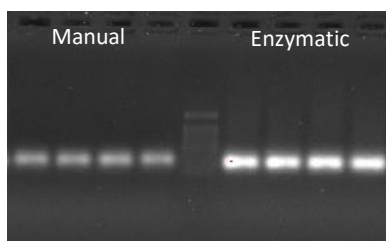


Figure 3. Amplification of DNA isolated from mouse tissues. DNA was amplified from mouse tissue isolated using the manual or enzymatic protocol using a GAPDH primer set, and then the product was run on a 1% agarose gel. Four biological replicates are shown for manual (first four lanes) or enzymatic (last four lanes) with 1kb Ladder in the center.