Promega

Product Application

Isolation of Genomic DNA from Mouse Ear Punches using the Maxwell® RSC Tissue DNA Kit

Isolate genomic DNA from mouse ear using the Maxwell® RSC Tissue DNA Kit on the Maxwell® RSC Instrument using enzymatic or manual homogenization.

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

Analyses: UV absorbance, dye-based quantitation, gel electrophoresis

Sample Type(s): Mouse ear punches

Input: Up to 10mg mouse ear tissue

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**

Materials Required:

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

Protocol:

Enzymatic Protocol:

1. Place mouse ear punches in a 1.5ml tube.
2. Add 200µl Tail Lysis Buffer, 20µl Proteinase K and 20µl RNase A Solution (optional).
3. Incubate at 56°C for 1 hr.
4. Spin at room temperature for 2 minutes at ≥16,000 × *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 the Maxwell® RSC Tissue DNA method on the Maxwell® RSC Instrument.

Manual Homogenization Protocol:

1. Place mouse ear punches in a 1.5ml tube.
2. Add 80µl 1x TE and 20µl RNase A Solution (optional).
3. Homogenize ear punches 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 the Maxwell® RSC Tissue DNA method on the Maxwell® RSC Instrument.

Results:

DNA was isolated from mouse ear punches (single punch per extraction) using either the enzymatic or manual homogenization methods listed above. Performance of these two protocols is shown below.

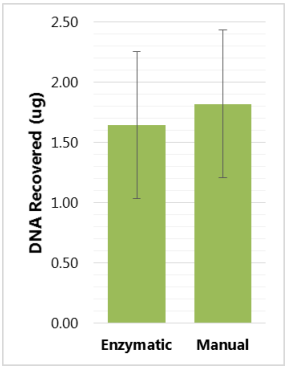

Figure 1. Yield of DNA isolated from mouse ear punches using the Maxwell® RSC Tissue DNA Kit (Cat.# AS1610) on the Maxwell® RSC Instrument (Cat.# AS4500). Following isolation, DNA concentration was measured using QuantiFluor® ONE dsDNA System (Cat.# E4871) on a Quantus™ Fluorometer (Cat.# E6150). Bars represent the average ± standard deviation (n=5).

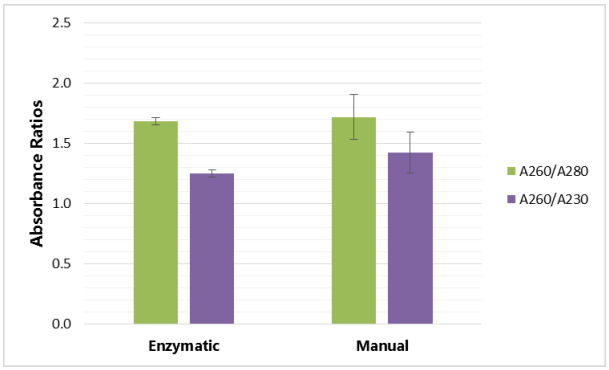

Figure 2. Purity of DNA isolated from mouse ear punches using the Maxwell® RSC Tissue DNA Kit (Cat.# AS1610) on the Maxwell® RSC Instrument (Cat.# AS4500). Absorbance was read at 230, 260, and 280nm on a NanoDrop ™ Spectrophotometer and ratios were calculated to access purity. Bars represent the average ± standard deviation (n=5).

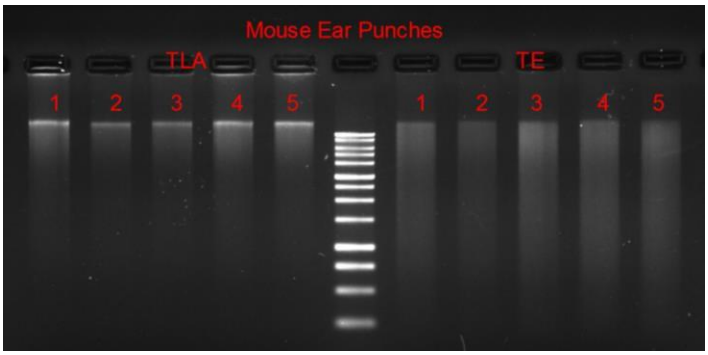

Figure 3. Agarose gel electrophoresis of DNA purified from mouse ear punches using the Maxwell® RSC Tissue DNA Kit (Cat.# AS1610) on the Maxwell® RSC Instrument (Cat.# AS4500). Approximately 100ng of DNA isolated from mouse ear using the manual (1X TE) or enzymatic (TLA) protocol was loaded per lane (n=5).