Promega

Product Application

DNA Purification from Chewing Gum for Noninvasive SNP Genotyping

Purify DNA from freshly chewed gum for downstream noninvasive SNP genotyping.

Kit: Maxwell® RSC PureFood GMO and Authentication Kit (Cat.# AS1600)

Analyses: qPCR, SNP genotyping

Sample Type(s): Chewing gum (Juicy Fruit Original and Bubble Yum Original recommended)

Input: ¼ piece of gum

Materials Required:

- Maxwell® RSC PureFood GMO and Authentication Kit (Cat.# AS1600)
- Maxwell® RSC Instrument (Cat.# AS4500)
- Vortexer
- Heat block set to 65°C
- Centrifuge

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 TM473, available at: **www.promega.com/protocols**

or contact Technical Services at: **techserv@promega.com**

Protocol:

Gum preparation.

1. Chew one stick of gum until flavor dissipates.
2. Place in saran wrap and cut into fourths.

DNA purification.

1. Place ¼ piece of gum in a 1.5ml tube with 300µl of CTAB.
2. Add 20µl of RNase A and 40µl of Proteinase K.
3. Tap, invert, and vigorously vortex.
4. Incubate at 65°C for 30 minutes.
5. Vortex tubes vigorously and centrifuge for 10 minutes at max speed (≥16,000 *x g*).
6. Meanwhile, prepare the cartridges and elution tubes as described in the Maxwell® RSC PureFood GMO and Authentication Kit Technical Manual (TM473).
7. Add 300µl of Lysis Buffer to well #1 of the cartridges.
8. Transfer the entire supernatant to well #1, taking care not to transfer any solid pieces of gum.
9. Process on the Maxwell® RSC following the PureFood GMO and Authentication protocol.

Results: DNA from chewing gum was purified with the Maxwell® PureFood GMO and Authentication Kit following the protocol outlined above. DNA yield was determined with the ProNex® DNA QC Assay ABI 7500/7500FAST (Cat.# NG1002, Figure 1), and 11ng DNA/sample was prepared for the TaqMan® SNP Genotyping System according to the technical manual (ThermoFisher). Genotyping was performed for the rs12913832 SNP near OCA2, one of many genes impacting eye color. G/G and A/G genotypes are more common in individuals with dark colored eyes (brown/hazel) and A/A genotypes are more common in those with light colored eyes (blue/gray). Allelic discrimination between the genotypes is shown in Figure 2.

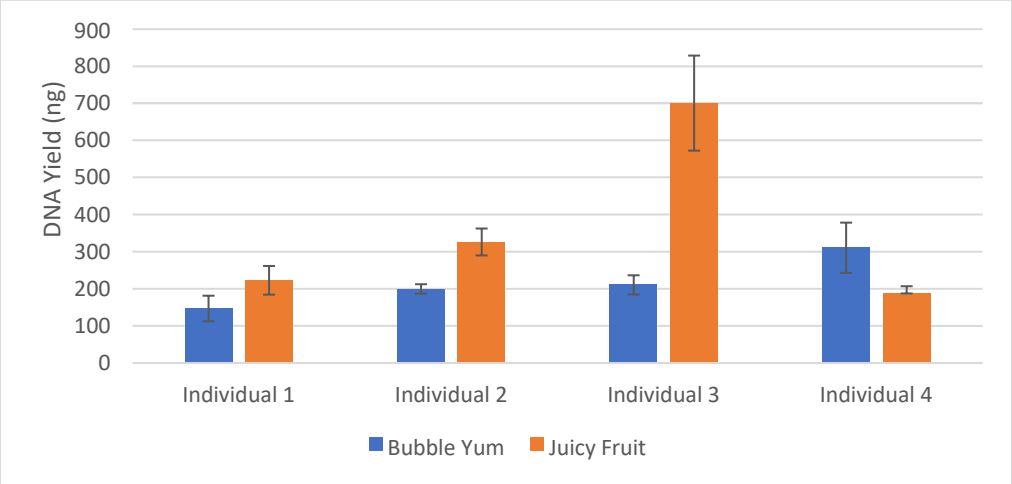

Figure 1. Yield of amplifiable DNA from chewing gum. DNA was purified from chewing gum from four individuals. DNA concentration was determined using amplification-based quantitation with the ProNex® DNA QC Assay ABI 7500/7500FAST. DNA yield was then calculated using the measured eluate volume. Mean ± SD of n=3 is shown.

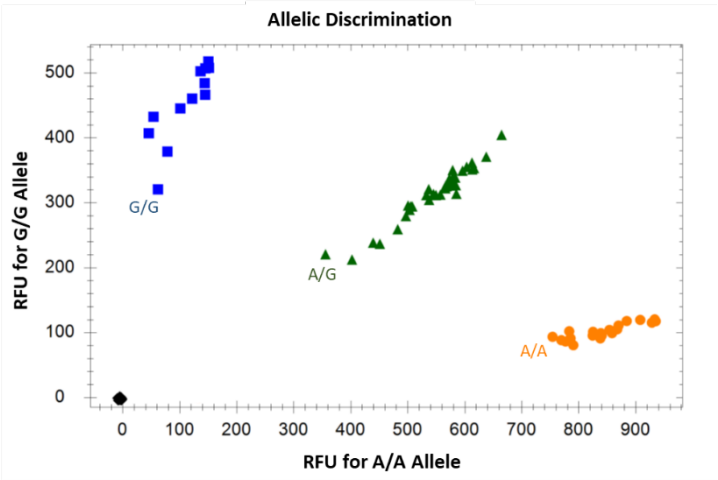

Figure 2. SNP genotyping of the eye color-related SNP rs12913832. DNA was purified from chewing gum from nine individuals. Samples were then amplified with the TaqMan® SNP Genotyping System with GoTaq® Probe qPCR Master Mix (Cat.# A6101) in duplicate. The rs12913832 SNP near OCA2, one of many genes impacting eye color, was analyzed by the assay. Individuals with a G/G genotype are shown by the blue squares. Individuals with a A/A genotype are shown by the orange circles. Heterozygous individuals are shown by the green triangles. NTCs are shown by the back diamonds.