Promega

Product Application

Automated Purification of DNA from Beef Broth Powder, Beef Broth Jelly and Pork Gelatin

Purify DNA from broth and gelatin food products using the Maxwell® RSC Instrument and the Maxwell® RSC PureFood GMO and Authentication Kit.

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

Analyses: Dye-based quantification and qPCR

Sample Type(s): Beef Broth Powder (Migros - Rindsbouillon)
Beef Broth Jelly (Knorr – Bouillon Töpfli)
Pork Gelatin (Patissier – Blattgelatine)

Input: 20-100mg

Materials Required:

- Maxwell® RSC PureFood GMO and Authentication Kit (Cat.# AS1600)
- Maxwell® RSC Instrument (Cat.# AS4500) or Maxwell® RSC 48 Instrument (Cat.# AS8500)
- Thermoblock (preferably with shaking)
- 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:

1. Add 1ml of CTAB buffer to each sample containing up to 100mg of sample.
2. Add 20µl of RNase A Solution and 40µl of Proteinase K (PK) Solution to each sample.
3. Vortex tubes until the samples are resuspended.
4. Incubate the samples at 65°C for 90 minutes in a thermoblock (preferably while shaking at 750rpm).
5. Prepare the Maxwell® RSC PureFood GMO and Authentication Kit cartridges according to TM473. Add 300µl of Lysis Buffer to well #1, add 100µl of Elution Buffer to the Elution Tube, and place a plunger into well #8.
6. Briefly vortex samples and load 300µl of the lysate into well #1 of the Maxwell® cartridge.
7. Process samples on the Maxwell® RSC Instrument using the PureFood GMO and Authentication method.

Results:

DNA was purified from either 20, 50 or 100mg of three different food samples. DNA concentrations were determined using the QuantiFluor® ssDNA System (Cat.# E3190). Since these food samples are highly processed, most of the DNA is single stranded. DNA amplifiability and detection of bovine or porcine DNA was assessed using RapidFinder™ ID Beef and Pork kits (5µl of DNA eluate was used in each amplification reaction).

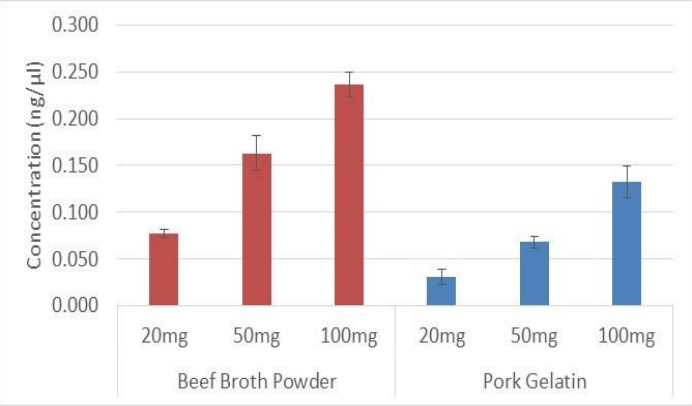

Figure 1: Concentration of DNA extracted from either 20, 50 or 100mg of Beef Broth Powder or Pork Gelatin using the Maxwell® RSC PureFood GMO and Authentication Kit (Cat.# AS1600). Samples were eluted in 100µl. DNA concentrations were determined using a fluorescent based method (QuantiFluor® ssDNA System, Cat.# E3190). Data are shown as mean ± STD of n=3.

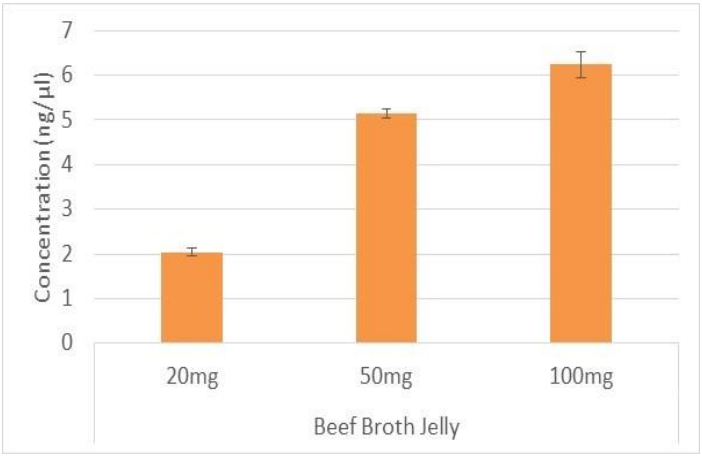

Figure 2: Concentration of DNA extracted from either 20, 50 or 100mg of Beef Broth Jelly using the Maxwell® RSC PureFood GMO and Authentication Kit (Cat.# AS1600). Samples were eluted in 100µl. DNA concentrations were determined using a fluorescent based method (QuantiFluor® ssDNA System, Cat.# E3190). Data are shown as mean ± STD of n=3.

Table 1: RealTime PCR analysis of DNA purified from either 20, 50 or 100mg of Beef Broth Powder, Pork Gelatin or Beef Broth Jelly. Cq values were determined based on 5µl DNA input into each RapidFinder ID detection assay (Thermo Fisher Scientific). Beef samples were only amplified using the Beef specific kit and Pork samples were only amplified using the Pork specific kit. Data are shown as mean ± STD of n=3. * denotes that only one of the three samples amplified.

Sample	Input	Lysate Volume	Average Cq ± StDev
Beef Broth Powder	20mg	300µl	34.4 ± 1.4
		600µl	33.4 ± 1.2
	50mg	300µl	33.8 ± 0.7
		600µl	32.9 ± 0.4
	100mg	300µl	33.8 ± 0.9
		600µl	32.4 ± 0.5
Beef Broth Jelly	20mg	300µl	35.5*
		600µl	39.3 ± 0.4
	50mg	300µl	35.3 ± 0.8
		600µl	36.8 ± 1.8
	100mg	300µl	37.9 ± 1.8*
		600µl	33.8 ± 1.3
Pork Gelatin	20mg	300µl	30.4 ± 0.2
		600µl	29.6 ± 0.0
	50mg	300µl	29.2 ± 0.1
		600µl	28.2 ± 0.1
	100mg	300µl	28.5 ± 0.1
		600µl	27.7 ± 0.2