Promega

Product Application

Automated Purification of DNA from *C. elegans*

Purify gDNA from C. elegans nematodes using the Maxwell® RSC Instrument and Maxwell® RSC PureFood GMO and Authentication Kit.

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

Analyses:

- UV absorbance
- Dye-based quantitation
- qPCR
- TapeStation

Sample Type(s): *C. elegans* worms

Input: 30 adult worms or a full plates of worms (60mm diameter)

Materials Required:

- Maxwell® RSC PureFood GMO and Authentication Kit (Cat.# AS1600)
- Maxwell® RSC Instrument (Cat.# AS4500)
- M9 Buffer[1]
- Thermomixer or heat block
- Microcentrifuge

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. Grow and collect *C. elegans* worms following standard method[1].
2. Centrifuge at 200rpm for 1 minute and remove supernatant. Optional: freeze worm pellet.
3. Add 300µl of CTAB Buffer and 30µl of Proteinase K to the worm pellet and vortex.
4. Incubate for 1 hour at 65°C with shaking at 800rpm.
5. During incubation, prepare purification cartridges as described in Maxwell® RSC PureFood GMO and Authentication Kit Technical Manual (TM473) section 5.1 and:
    a. Add 300µl of Lysis Buffer to Well #1.
    b. Add 10µl of RNAse A to Well #3.
    c. Place a plunger in Well #8.
    d. Place empty Elution Tubes into the elution position and add 100µl of Elution Buffer to each.
6. After incubation, vortex and spin down. Transfer the entire worm lysates to Well #1 of the cartridge.
7. Place the prepared cartridges in the Maxwell® RSC Instrument and run the Maxwell® RSC PureFood GMO and Authentication Kit method.

Results:

DNA was purified from *C. elegans* worms using the Maxwell® RSC PureFood GMO and Authentication Kit (Table 1). TapeStation analysis (Agilent) showed a DIN score of 8.9 for DNA purified from a whole worm plate (data not shown). Purified DNA was amplifiable by qPCR (Table 1) without inhibition (data not shown).

Table 1: Concentration, yield, absorbance ratios and Cq values of DNA purified from *C. elegans* using the Maxwell® RSC PureFood GMO and Authentication Kit. DNA was purified from 30 adult N2 worms* or the progeny of 2 worms grown for 5 days at 20°C in a 60mm diameter NGM (Nematode Growth Media[1]) plate. DNA concentration and purity ratios were measured by absorbance using a NanoDrop™ One Spectrophotometer. DNA concentration was also measured using QuantiFluor® ONE dsDNA System (Cat.# E4871) on a GloMax® Discover Microplate Reader (Cat.# GM3000). Average yields were calculated based on concentrations and the recovered volume in elution tubes (~90µl). Mean values are shown for n=3. DNA was amplified with *tba-1* gene-specific primers[2] using GoTaq® qPCR Master Mix (Cat.# A6001) following TM318 on a QuantStudio™ 5 Real-Time PCR System (Applied Biosystems™). Shown are the average values for N=6.

	Absorbance		Fluorescence		qPCR
Input	A260/A280	A260/A230	Concentration (ng/µl)	Yield (µg)	Cq Value
30 worms	1.97	1.17	3.58	0.32	22.95
1 plate (60mm diameter)	2.11	2.43	79.77	7.18	13.79

***C. elegans* worms were kindly provided by Dr Florence Solari from the Genetics and Neurobiology of *C. elegans* research team at Institut NeuroMyoGène, University of Lyon, France.**

References:

1. Stiernagle, T. Maintenance of *C. elegans* (February 11, 2006), WormBook, ed. The *C. elegans* Research Community, WormBook, doi/10.1895/wormbook.1.101.1, http://www.wormbook.org.

2. Mergoud dit Lamarche, A. et al., (2018) UNC-120/SRF independently controls muscle aging and lifespan in Caenorhabditis elegans. *Aging Cell*. Apr;17(2):e12713.