Promega

Product Application

Viral RNA Purification and Detection from Environmental Surface Swabs

Purify viral RNA from environmental surface swabs stored in viral transport medium using the Maxwell® RSC PureFood GMO and Authentication Kit on the Maxwell® RSC Instrument.

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

Analyses: RT-qPCR for detection of spiked viral targets

Sample Type(s): Environmental swabs collected and stored in a viral transport medium (VTM)

Input: 200µl

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**

Materials Required:

- Maxwell® RSC PureFood GMO and Authentication Kit (Cat.# AS1600)
- Maxwell® RSC Instrument (Cat.# AS4500)
- Heat block set to 56°C
- GoTaq® Probe 1-Step RT-qPCR System (Cat.# A6121)

Protocol:

1. Transfer 200µl of inoculated viral transport medium to a 1.5ml tube.
2. Add 200µl of CTAB and 40µl of Proteinase K Solution to each sample.
3. Vortex to mix.
4. Incubate samples at 56°C for 10 minutes.
5. Meanwhile, prepare cartridges as indicated in the Technical Manual (TM473).
    a. Add 100µl of Elution Buffer to each elution tube.
6. Add 200µl of Lysis Buffer to well #1.
7. Transfer the entire lysate to well #1.
8. Select the Maxwell® RSC PureFood GMO and Authentication Kit run method, place the prepared deck tray in the Maxwell® RSC Instrument, and start the method.

Results:

The Maxwell® RSC PureFood GMO and Authentication kit can be used to purify viral RNA from multiple swab materials stored in different transport mediums. As expected, earlier Cq values were observed with swabs stored in 1ml VTM as compared to 3ml. The differences in Cq values between the two surfaces tested, cleaned lab bench and tile floor, indicate that the surface being swabbed can have a notable impact on Cq value and may introduce PCR inhibitors to the RT-qPCR reaction.

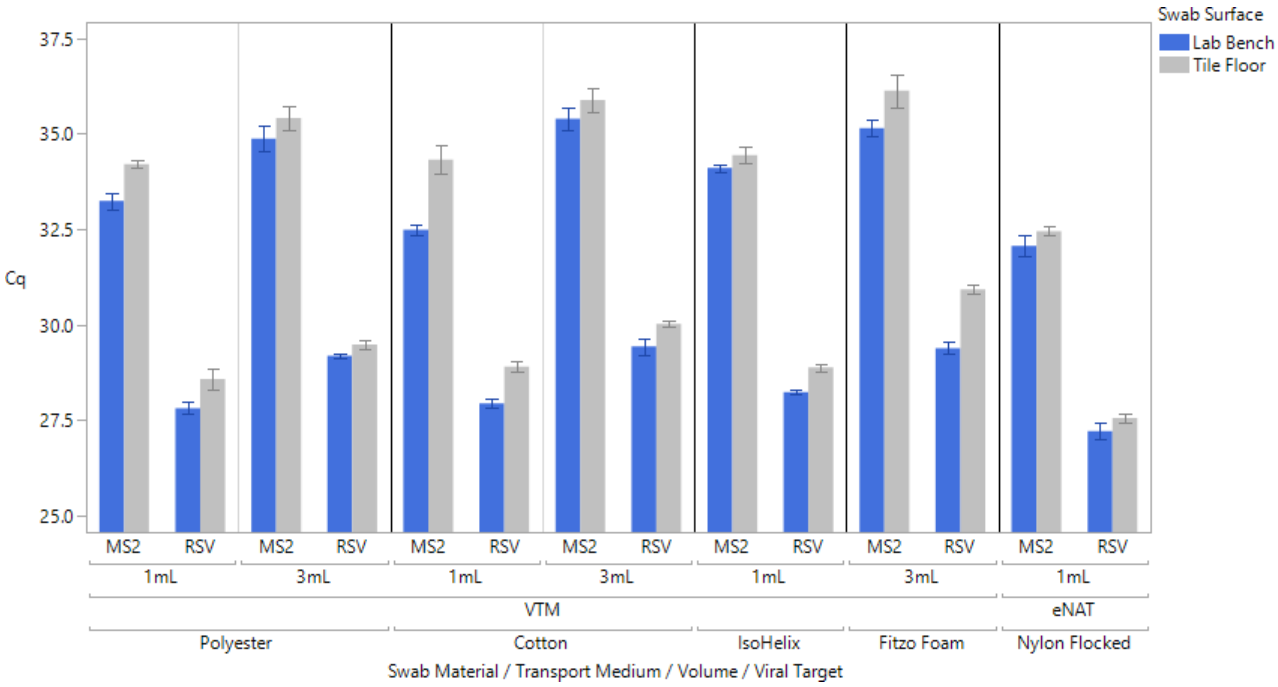

Figure 1. Amplification of MS2 and RSV viral RNA purified from environmental swabs stored in viral transport medium using the Maxwell® RSC PureFood GMO and Authentication Kit. Environmental swabs were pre-wet with 100µl of 1X PBS and collected[1] from a bleached lab bench or atrium tile floor using multiple swab materials. Swabs were placed immediately into Viral Transport Medium (VTM), unless a transport medium was provided as in the case of eNAT™ system. Each transport medium was spiked with ~1E5 copies of inactivated RSV pellet along with ~5E4 copies of MS2 bacteriophage. 200µl of inoculated sample was purified in triplicate using the Maxwell® RSC PureFood and Authentication Kit (Cat.# AS1600) on the Maxwell® RSC Instrument (Cat.# AS4500). The presence of viral RNA was detected using 5µl of eluate in a 20µl RT-qPCR reaction using GoTaq® Probe 1-Step RT-qPCR System (Cat.# A6121) and virus-specific primers. Data represent the average Cq of triplicate purifications amplified in duplicate. Error bars show standard deviation.

Reference:

1. World Health Organization. (2020). Surface sampling of coronavirus disease (COVID-19): a practical “how to” protocol for health care and public health professionals. Reference Number WHO/2019- nCoV/Environment_protocol/2020.1. Retrieved from **https://www.who.int/publications/i/item/surface-sampling-of-coronavirus-disease-(-covid-19)-a- practical-how-to-protocol-for-health-care-and-public-health-professionals**.