Promega

Product Application

Automated Purification of Viral DNA from Pig Blood

Purify high quality viral DNA from pig blood using the Maxwell® RSC Instrument and the Maxwell® RSC Blood DNA and Whole Blood DNA Kits.

Kits: Maxwell® RSC Blood DNA Kit (Cat.# AS1400) and Maxwell® RSC Whole Blood DNA Kit (Cat.# AS1520)

Analyses: UV-absorbance, qPCR

Sample Type(s): Pig Blood

Input: 200µl

Materials Required:

- Maxwell® RSC Instrument (Cat.# AS4500), or
- Maxwell® RSC 48 Instrument (Cat.# AS8500)
- Maxwell® RSC Blood DNA Kit (Cat.# AS1400)
- Maxwell® RSC Whole Blood DNA Kit (Cat.# AS1520)

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 TM419 and TM455, available at: **www.promega.com/protocols**

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

Maxwell® RSC Blood DNA Protocol:

1. Add 30µl of Proteinase K (PK) Solution to each incubation tube.
2. Add 200µl of liquid blood to each incubation tube.
3. Add 300µl of Lysis Buffer to each incubation tube.
4. Vortex each tube for 10 seconds.
5. Immediately transfer each blood lysate sample to well #1 of each cartridge (well #1 is the largest well). Note: Inclusion of the standard 56°C incubation step for 20 minutes has been shown to reduce the quality of the purified DNA from pig blood.
6. Place one plunger into well #8 of each cartridge.
7. Place an empty elution tube into the elution tube position for each cartridge in the deck tray. Add 50µl of Elution Buffer to the bottom of each elution tube.
8. Run the Blood DNA method on the Maxwell® RSC or Maxwell® RSC 48 Instrument.

Maxwell® RSC Whole Blood DNA Protocol:

1. Transfer 200µl of each blood sample to well #1 of each cartridge (well #1 is the largest well).
2. Tip-mix the blood sample in well #1 to ensure all blood has been transferred. Change pipette tips between samples.
3. Place one plunger into well #8 of each cartridge.
4. Place an empty elution tube into the elution tube position for each cartridge in the deck tray. Add 60µl of Elution Buffer to the bottom of each elution tube.
5. Run the Whole Blood DNA method on the Maxwell® RSC or Maxwell® RSC 48 Instrument.

Results:

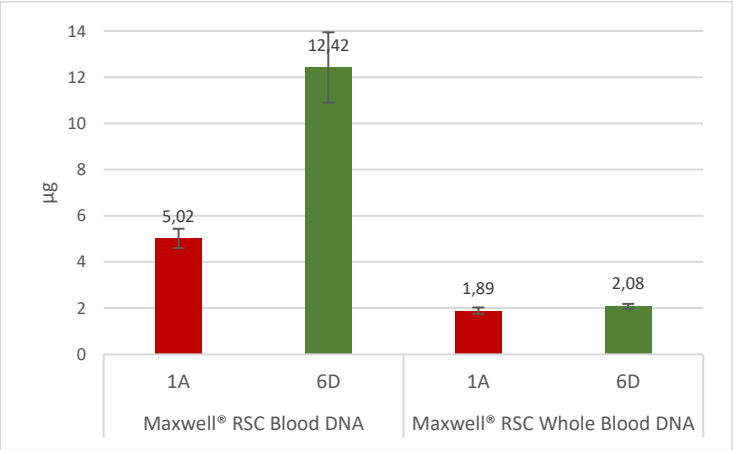

Figure 1. DNA yields from pig blood samples 1A and 6D purified using the Maxwell® RSC Blood DNA Kit (Cat.# AS1400) and Maxwell® RSC Whole Blood DNA Kit (Cat.# AS1520) on the Maxwell® RSC Instrument (Cat.# AS4500). Assessed by NanoDrop™ 8000 Spectrophotometer. Data represent the mean of N=3 ± SD.

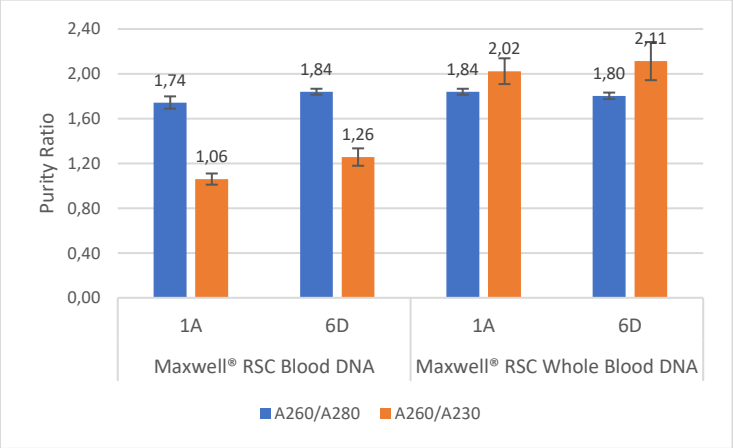

Figure 2. Purity of DNA (A_{260}/A_{280} and A_{260}/A_{230}) from pig blood samples 1A and 6D purified using the Maxwell® RSC Blood DNA Kit and Maxwell® RSC Whole Blood DNA Kit on the Maxwell® RSC Instrument. Assessed by NanoDrop™ 8000 Spectrophotometer. Data represent the mean of N=3 ± SD.

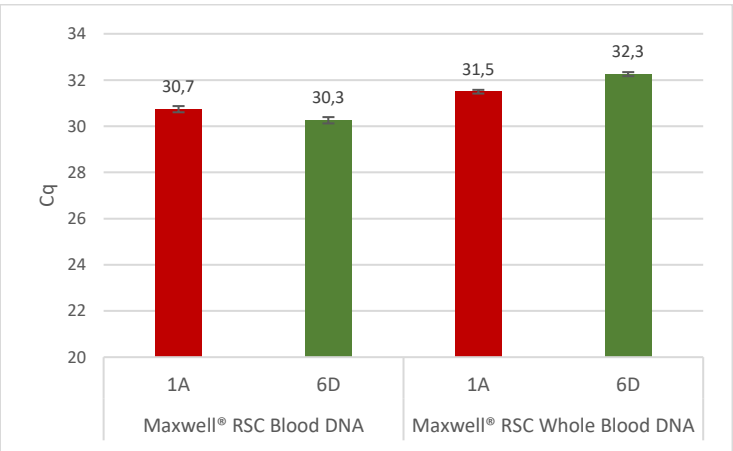

Figure 3. Amplifiability of CMV DNA purified from pig blood samples 1A and 6D spiked with 5,000 copies of CMV (200µl sample) using the Maxwell® RSC Blood DNA Kit and Maxwell® RSC Whole Blood DNA Kit on the Maxwell® RSC Instrument. DNA was analyzed on a Biorad CFX96™ Real-Time PCR System using the GoTaq® Probe qPCR Master Mix (Cat.# A6101) with CMV specific primers (104bp amplicon). 2µl of purified DNA was used in a 20µl qPCR reaction volume. Data represent the mean of N=3 ± SD.