Maxwell® RSC Blood Kits: A Comparison to QIAcube® Methods

A Maxwell® RSC Blood DNA Kit and Maxwell® RSC simplyRNA Blood Kit Application Note

Materials Required:
- Fresh whole blood
- Maxwell® RSC Blood DNA Kit (Cat.# AS1400)
- Maxwell® RSC simplyRNA Blood Kit (Cat.# AS1380)
- GoTaq® Probe qPCR Master Mix (Cat.# A6101)
- GoTaq® Probe 1-Step RT-qPCR System (Cat.# A6120)
- QuantiFluor® ONE dsDNA System (Cat.# E4871)
- QuantiFluor® RNA System (Cat.# E3310)

Instrument Requirements:
- Maxwell® RSC Instrument (Cat.# AS4500)
- NanoDrop® Spectrophotometer
- Quantus™ Fluorometer (Cat.# E6150)
- Agilent Technologies Bioanalyzer Instrument 2100

Performance Comparison:
- QIAamp® DNA Blood Mini Kit automated on the QIAcube®
- QIAamp® RNA Blood Mini Kit automated on the QIAcube®

The Maxwell® RSC instrument purifies high-quality DNA and RNA from whole blood, providing equivalent or better performance than QIAcube® methods, and more flexibility in input sample volume.

Introduction

The Maxwell® Rapid Sample Concentrator (RSC) provides automated purification of DNA, RNA or total nucleic acids from up to 16 samples in a single run. Used with the prefilled reagent cartridges supplied in the Maxwell® purification kits, the Maxwell® RSC Instrument can purify DNA or RNA from a wide range of sample types. The intuitive graphical user interface makes the instrument easy to use, and the integrated Quantus™ Fluorometer lets you collect purification and quantification data in one report.

The Maxwell® RSC Blood DNA and simplyRNA Blood Kits provide a simple method for purifying DNA or RNA from whole blood. Here, we compare the performance of these Maxwell® RSC blood DNA and RNA purification methods with extraction methods using the QIAamp® DNA and RNA Blood Mini Kits on the QIAcube® instrument.

Methods

Freshly drawn whole blood was used for nucleic acid purification. RNA purification was performed using the Maxwell® RSC simplyRNA Blood Kit and the Maxwell® RSC Instrument, or the QIAamp® RNA Blood Mini Kit and the QIAcube® Instrument. DNA was purified using the Maxwell® RSC Blood DNA Kit or QIAamp® DNA Blood Mini Kit. The maximum recommended blood input values were used for each kit (2.5ml for Maxwell® RSC simplyRNA Blood, 1.5ml for QIAamp® RNA Blood, 300µl for Maxwell® RSC Blood DNA and 200µl for QIAamp® DNA Blood Mini Kit). All conditions were performed in quadruplicate for each donor.

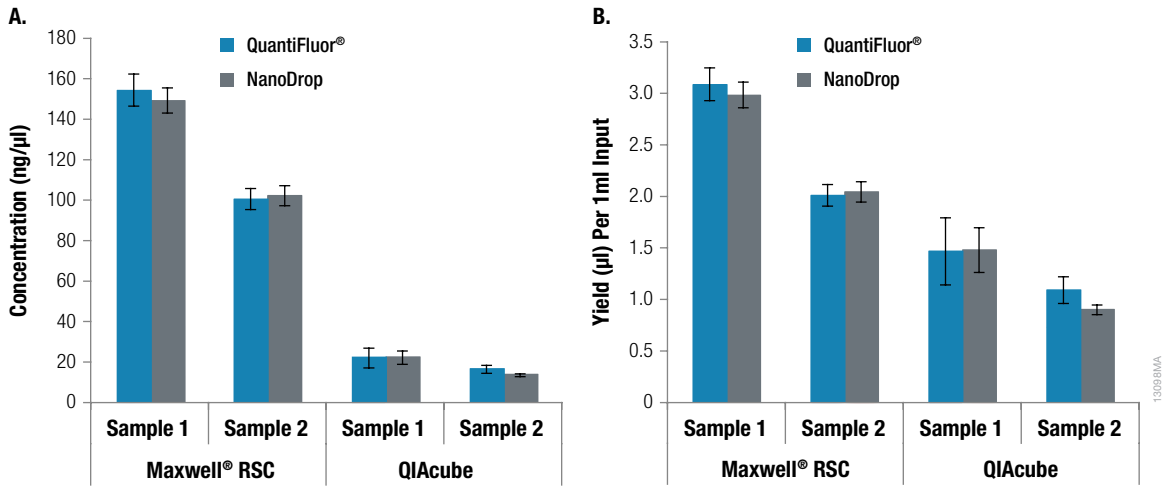

Figure 1. Concentration and yield of DNA isolated using different extraction methods. Panel A. Concentration of DNA isolated from whole blood. The maximum input volumes were used for each kit (300µl for the Maxwell® RSC Blood DNA Kit, and 200µl for the QIAamp® method). The isolated DNA was eluted in 200µl (QIAamp®) or 50µl (Maxwell® RSC). **Panel B.** DNA yield per 100µl of blood input. Data show the mean and standard deviation for four replicate samples from each donor and condition. DNA quantitation was performed using two methods: the NanoDrop® 1000 Instrument and the QuantiFluor® ONE dsDNA System with the Quantus™ Fluorometer.

DNA Purification Results

DNA Yield and Purity: Figure 1 shows the concentration and yield of DNA purified from whole blood. DNA samples purified with the Maxwell® RSC DNA Blood Kit were consistently higher in concentration than those purified using the QIAamp® kit. To correct for differences in sample input (300µl for Maxwell® RSC and 200µl for QIAamp®) and elution volumes (50µl for Maxwell® RSC and 200µl for QIAamp®), we calculated the DNA yield per 100µl input. The Maxwell® RSC consistently recovered more DNA per volume of blood when compared to the QIAamp® method. The purity of DNA isolated with the Maxwell® RSC method was equivalent or better than that of samples purified with the QIAamp®/QIAcube® method, as indicated by absorbance ratio (Figure 2).

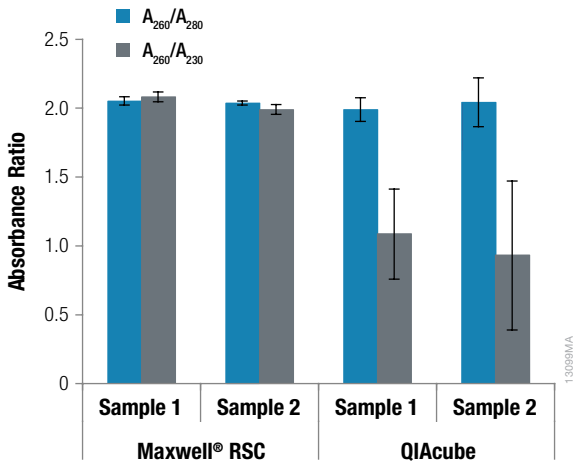

Figure 2. Purity of DNA isolated from blood using the QIAamp®/ QIAcube® and Maxwell® RSC Blood DNA methods. Data show the mean and standard deviation for four replicate samples from each donor and condition.

qPCR Performance: To determine the performance of the purified DNA in qPCR, we amplified 1µl of the purifed DNA in a 20µl GoTaq® Probe qPCR System reaction with DNA-specific CAPZA3 primers (Figure 3). Lower C_t values were observed with DNA purified on the Maxwell® RSC with the RSC Blood DNA Kit, indicating that the Maxwell® method generated a higher concentration of amplifiable DNA.

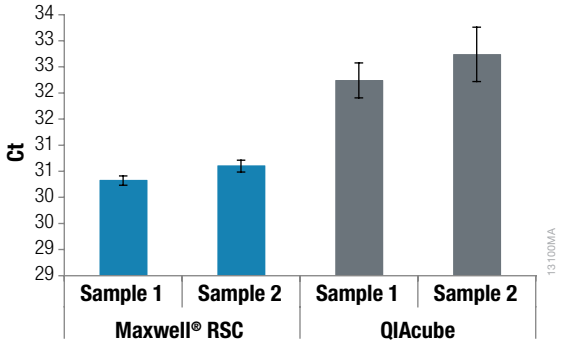

Figure 3. C_t values for purified DNA samples. qPCR was performed using 1µl of purifed DNA in a 20µl GoTaq® Probe qPCR System reaction with DNA-specific CAPZA3 primers. C_t values for each purification method are shown. Data show the mean and standard deviation for four replicate samples from each donor and condition.

RNA Purification Results

RNA Yield and Purity: Figure 4 shows the concentration and yield of RNA isolated from whole blood using each method. RNA samples purified with the Maxwell® RSC simplyRNA Kit were consistently higher in concentration compared to RNA purified using the QIAamp® method. To correct for differences in sample input volume (2.5ml for Maxwell® RSC and 1.5ml for QIAamp®) and elution volumes (50µl for Maxwell® RSC and 100µl for QIAamp®), we calculated the RNA yield per 1ml of input. The normalized results in Figure 4B show that the Maxwell® RSC method consistently recovered more RNA per volume of blood compared to the QIAamp® method. RNA purified using the Maxwell® RSC simplyRNA method was equivalent in purity to RNA from the QIAcube® method (Figure 5).

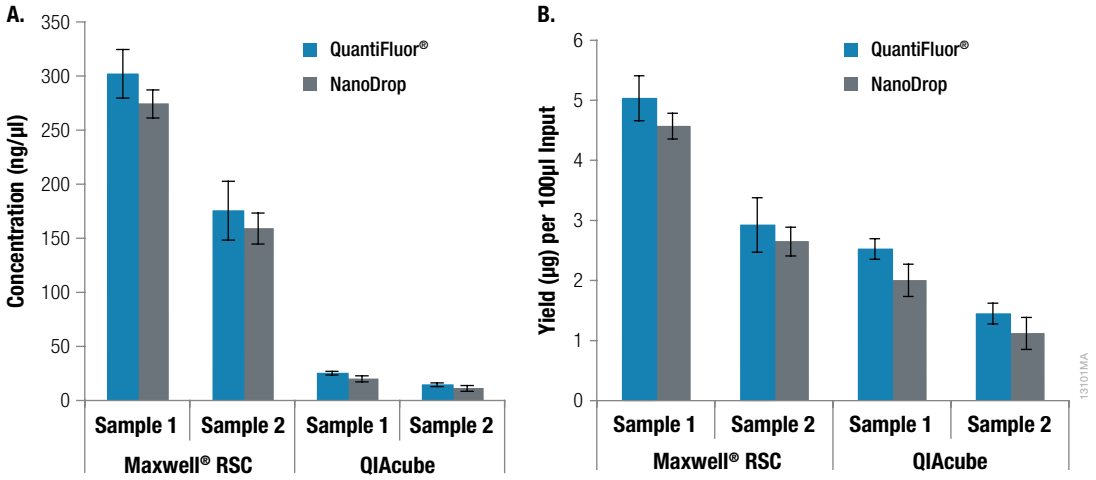

Figure 4. Concentration and yield of RNA isolated using Maxwell® RSC and QIAcube® methods. Panel A. Concentration of RNA isolated from whole blood. The maximum input volumes were used for each kit (2.5ml for RSC simplyRNA® and 1.5ml for the QIAamp® method). Isolated RNA was eluted in 100µl (QIAamp® method) or 50µl (Maxwell® method). **Panel B.** RNA yield per milliliter of blood. Data show the mean and standard deviation for four replicate samples from each donor and condition. RNA quantitation was performed using two methods: the NanoDrop® 1000 Instrument, and the QuantiFluor® RNA System with the Quantus™ Fluorometer.

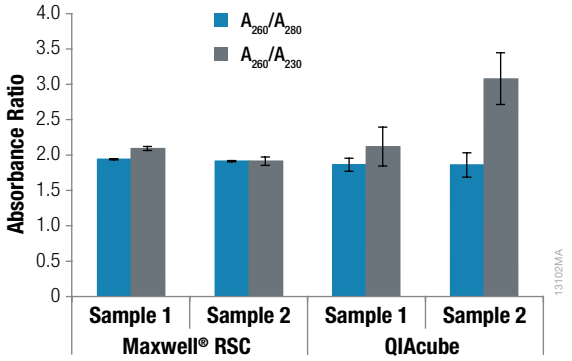

Figure 5. Purity of RNA isolated from blood using Maxwell® RSC and QIAamp®/QIAcube® methods. Data show the mean and standard deviation for four replicate samples from each donor and condition.

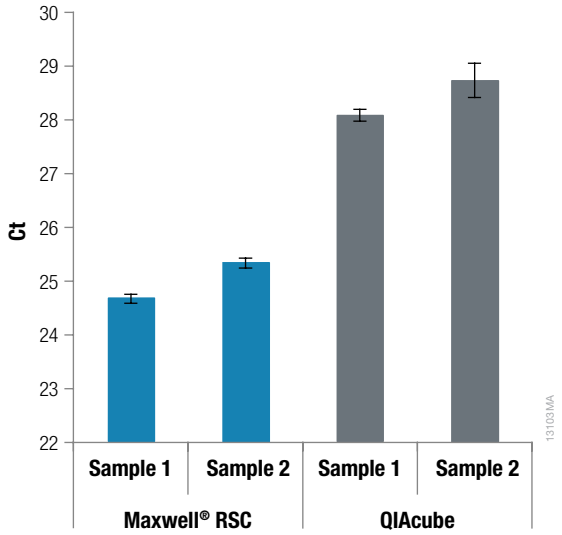

Figure 6. C_t values for RNA samples. RT-qPCR was performed using 1µl of purified RNA in a 25µl GoTaq® Probe 1-Step RT-qPCR System reaction with RNA-specific HPRT1 primers. C_t values for each purification method are shown. Data show the mean and standard deviation for four replicate samples from each donor and condition.

RT-qPCR Performance: To evaluate performance of the purified RNA in RT-qPCR, 1µl of RNA purifed with each method was used in a 25µl GoTaq® Probe 1-Step RT-qPCR System reaction with RNA-specific HPRT1 primers (Figure 6). Lower C_t values were observed with RNA purified with the Maxwell® RSC simplyRNA Blood Kit, indicating higher amplifiable RNA concentrations in these samples.

Bioanalyzer Analysis: RNA integrity was determined using an Agilent 2100 Bioanalyzer. RNA integrity number (RIN) values were determined for each purified sample, with 10 being the highest possible score (Figure 7). Maxwell® RSC simplyRNA-purified samples showed more consistent and higher RIN values compared to QIAcube®-purified samples.

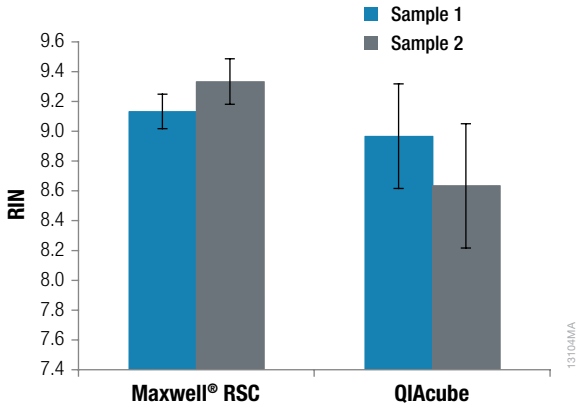

Figure 7. RIN values for RNA isolated from blood. Data show the mean and standard deviation for four purifications for each method using replicate samples from each donor.

Conclusions

The Maxwell® RSC Instrument provides consistent, high-quality DNA and RNA purification from whole blood samples.The data presented here show that DNA and RNA samples purifed using the Maxwell® RSC instrument and kits matched or outperformed the QIAamp®/QIAcube® samples in every parameter tested.

The concentration and yield data demonstrate the efficiency of nucleic acid purification with the Maxwell® RSC Blood DNA and simplyRNA Blood Kits. In addition to providing higher concentration and yield, the Maxwell® RSC Kits have a higher sample input capacity than the QIAamp® methods, providing greater protocol flexibility. More consistent purity ratios are another advantage of the Maxwell® kits compared to the QIAamp® methods.

The Maxwell® RSC-purified nucleic acids also gave more robust and consistent results in qPCR and RT-qPCR. Finally RIN values for RNA purified with the simplyRNA blood method were consistently higher than those for RNA.

Table 1. Summary of Results Comparing the Maxwell® RSC Blood DNA Kit, QIAamp® DNA Blood Mini Kit, Maxwell® RSC simplyRNA Blood Kit, and QIAamp® RNA Blood Mini Kit.

Purification Kit	Concentration by QuantiFluor®	Input	Elution Volume	Amplifiable Yield	Preprocessing Time
Maxwell® RSC Blood DNA Kit	175–302ng/µl	300µl	50µl	7.9–13.8µg	30 minutes
QIAamp® DNA Blood Mini Kit	14–25ng/µl	200µl	200µl	2.9–5.1µg	15–20 minutes
Maxwell® RSC simplyRNA Blood Kit	100–154ng/µl	2.5ml	50µl	543.2–650.5ng	35–40 minutes
QIAamp® RNA Blood Mini Kit	16–22ng/µl	1.5ml	100µl	323.6–433.5ng	55–60 minutes

Ordering Information

Product	Cat.#
Maxwell® RSC Blood DNA Kit	AS1400
Maxwell® RSC simplyRNA Blood Kit	AS1380
Maxwell® RSC Instrument	AS4500
QuantiFluor® ONE dsDNA System	E4870
QuantiFluor® RNA System	E3310
GoTaq® Probe qPCR Master Mix	A6101
GoTaq® Probe 1-Step RT-qPCR Master Mix	A6120