



Product Application

RNA purification from bacterial culture using Maxwell® RSC simplyRNA Blood Kit

High quality RNA was purified from bacterial culture using Maxwell® RSC simplyRNA Blood Kit (Cat.# AS1380)

Kit: Maxwell® RSC simplyRNA Blood Kit (Cat.# AS1380)

Analyses: UV absorbance, dye-based quantitation, TapeStation analysis.

Sample Type(s): Bacterial culture

Input: from 250µl to 500µl

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.

Further information can be found in Technical Manual TM#417, available at: www.promega.com/protocols

or by e-mailing technical services at techserv@promega.com

Materials Required:

- Maxwell® RSC Instrument (Cat.# AS4500 or Cat.# AS8500)
- Maxwell® RSC simplyRNA Blood Kit (Cat.# AS1380)

Protocol:

1. Before starting, prepare DNase I Solution and 1-Thioglycerol/Homogenization Solution as indicated in TM#417.
2. Aliquot bacterial culture in 1.5ml microtubes.
3. Pellet at 8000rpm for 5min and remove medium.
4. Resuspend pellets in 200µl of 1-Thioglycerol/Homogenization Solution by vortexing or pipetting.
5. Add 200µl of Lysis Buffer and 25µl of Proteinase K on resuspended pellets, invert 6 times and incubate for 10min at room temperature.
6. During the incubation prepare cartridges as indicated in TM#417:
7. Start the simplyRNA method on the Maxwell® RSC Instrument.



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Results:

Sample	input	NanoDrop™ One				QuantiFluor® RNA System		TapeStation
		Concentration (ng/μl)	Yield (μg)	A260/A280	A260/A230	Concentration (ng/μl)	Yield (μg)	
KRX (N=3)	250μl	90.87±9.63	4.54±0.48	1.75±0.01	1.54±0.01	93.84±9.27	4.69±0.46	9.53±0.06
	500μl	223.63±38.32	11.18±1.92	1.77±0.01	1.56±0.06	230.20±49.87	11.51±2.49	9.60±0.00
	1ml	405.47±81.12	20.27±4.06	1.78±0.02	1.58±0.01	402.74±32.13	20.14±1.61	9.63±0.06
JM109 (N=3)	250μl	68.26±8.65	3.41±0.43	1.76±0.01	1.57±0.07	73.96±7.34	3.70±0.37	9.53±0.06
	500μl	209.28±16.21	10.46±0.81	1.78±0.00	1.69±0.00	234.83±28.92	11.74±1.45	9.43±0.06
	1ml	448.24±10.80	22.41±0.54	1.86±0.02	1.75±0.02	440.61±13.78	22.03±0.69	9.43±0.06

Table 1: Concentrations, yields, purity ratios and RIN scores of RNA purified from bacterial cultures (KRX Cat.#L3002 and JM109, Cat.# L2005) with Maxwell® RSC simplyRNA Blood kit. Yields were calculated for 50μl elution volume. Shown is the average yields for N=3 ± standard deviation.

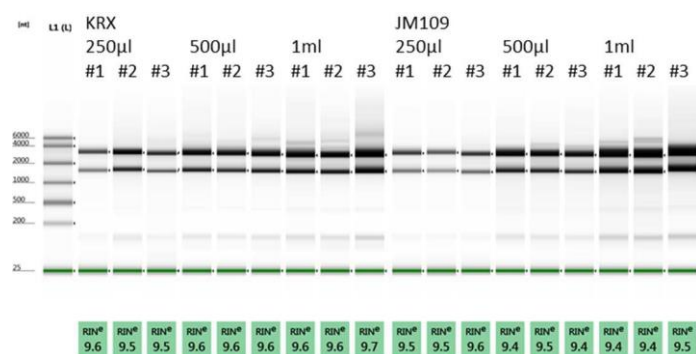


Figure 1: TapeStation 4200 gel image and RIN scores obtained for RNA purified from bacterial cultures with Maxwell® RSC simplyRNA Blood kit.

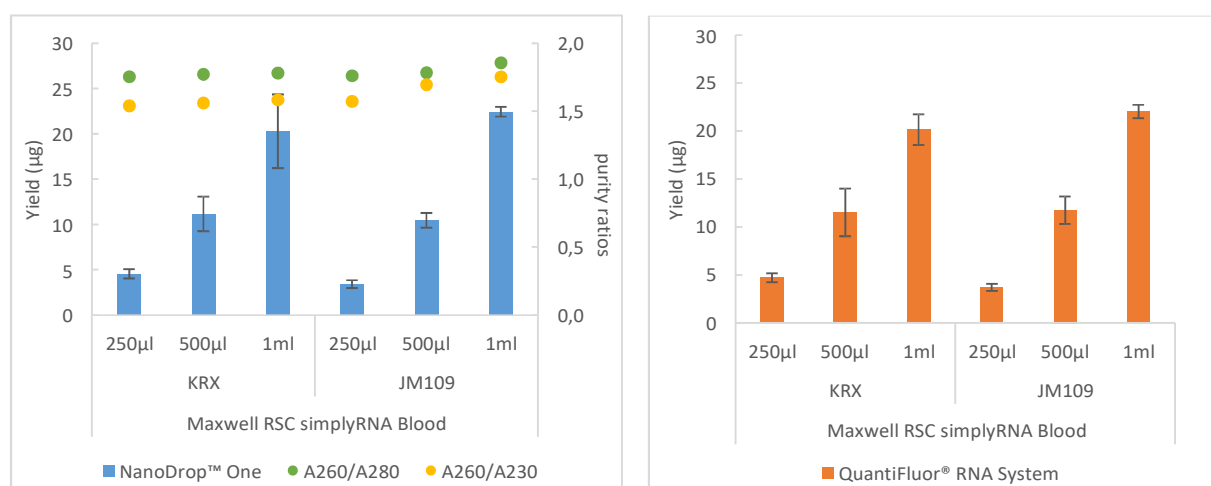


Figure 2. Average yields and purity ratios of RNA purified from bacterial cultures using Maxwell® RSC simplyRNA Blood kit. Yields were calculated for 50μl elution volume. Left. Yields and purity of RNAs measured by absorbance using a NanoDrop™ One Spectrophotometer. Right. Yields measured by fluorescence with the QuantiFluor® RNA System (Cat.# E3310). Shown is the average N=3 ± standard deviation.