

Maxwell® RSC DNA FFPE Kit

INSTRUCTIONS FOR USE OF PRODUCT AS1450.

Preprocessing FFPE Section Samples

1. Place the FFPE tissue section into a 1.5ml microcentrifuge tube. If using slide-mounted tissue sections, scrape the section off the slide using a clean razor blade. Tap or centrifuge tube briefly to collect the sample at the bottom of the tube.

Note: 5–10 micron-thick tissue sections ranging in size from 20mm² to 200mm² for a total of up to 2.0mm³ of tissue can be used.

- 2. Add 300µl of Mineral Oil to the sample tubes. Vortex for 10 seconds.
- 3. Heat the samples at 80°C for 2 minutes, then place samples at room temperature while the master mix is prepared.
- 4. Prepare a master mix of the Lysis Buffer, Proteinase K Solution and Blue Dye as shown below:

	Reactions		
Reagent	Amount/Reaction	(number to be run + 2)	Total
Lysis Buffer	224µl	n + 2	224 × (n + 2)μl
Proteinase K	25μΙ	n + 2	$25 \times (n+2)\mu I$
Blue Dye	1μΙ	n + 2	1 × (n + 2)µl

For fewer than six samples, prepare enough master mix for n + 1 samples.

Note: Use the master mix within 1 hour of preparation. Master mix cannot be stored for later use.

- 5. Add 250µl of master mix to each sample tube, and vortex for 5 seconds.
- 6. Centrifuge sample tubes at $10,000 \times g$ for 20 seconds to separate layers. If a pellet is present in the aqueous layer (lower, blue layer), gently mix the aqueous phase with a pipet to resuspend the pellet.
- 7. Transfer the sample tubes to a 56°C heat block and incubate for 30 minutes.
- 8. Transfer the sample tubes to an 80°C heat block and incubate for 4 hours.
- 9. Remove the sample tubes from the heat block, and allow the samples to cool to room temperature for 5 minutes.
- 10. Add 10µl RNase A Solution to the aqueous (blue) phase in each sample tube. Mix by pipetting.
- 11. Incubate for 5 minutes at room temperature (15–30°C). During the incubation, begin cartridge preparation.
- 13. Centrifuge the sample tubes at full speed in a microcentrifuge for 5 minutes.
- 14. Immediately transfer the blue, aqueous phase containing the DNA to well #1 of a Maxwell® FFPE Cartridge.



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Method Setup and Cartridge Preparation

Maxwell® RSC Method Setup

Before using the Maxwell® RSC DNA FFPE Kit for the first time, the FFPE DNA method must be installed on your instrument. The method is available at: **www.promega.com/resources/tools/maxwellrscmethod**. See the *Maxwell® RSC Methods Installation Technical Manual* #TM435 for instructions.

Cartridge Preparation

1. Place the cartridges to be used in the deck tray with well #1 (the largest well in the cartridge) farthest away from the Elution Tubes. Press down on the cartridge to snap it into position. Ensure both cartridge ends are fully seated in the deck tray. Carefully peel back the seal so that the entire seal is removed from the top of the cartridge. Ensure that all sealing tape and any residual adhesive are removed from the cartridges. **Caution:** Handle cartridges with care. Seal edges may be sharp.

Note: If you are processing fewer than 16 samples, center the cartridges on the deck tray.

- 2. Place one plunger into well #8 of each cartridge.
- 3. Place an empty Elution Tube into the Elution Tube position for each cartridge in the deck tray.
- Add 50µl of Nuclease-Free Water to the bottom of each Elution Tube. The Elution Tubes must stay open during the RNA purification.

Note: Use only the CSC/RSC Plungers, Elution Tubes and Nuclease-Free-Water supplied with the Maxwell® RSC DNA FFPE Kit. Plungers for Maxwell® 16 LEV kits are not compatible with the Maxwell® RSC Instrument. Other elution tubes may not be compatible with the Maxwell® RSC Instrument and may affect performance. Use of other elution buffers may impact DNA purification performance or downstream use.



Figure 1. Setup and configuration of the deck tray.

Instrument Run on the Maxwell® RSC Instrument (Cat.# AS4500)

- 1. Follow the instrument run instructions in the Maxwell® RSC DNA FFPE Kit Technical Manual #TM437.
- 2. Refer to the *Maxwell*[®] *RSC Instrument Operating Manual* #TM411 for detailed information.

Additional protocol information in Technical Manual #TM437 available online at: www.promega.com/protocols/