Product Application

# Droplet Digital PCR Compatibility with Maxwell® RSC and HT DNA FFPE Purifications

*DNA was purified from FFPE human normal adjacent lung tissue using the Maxwell® RSC DNA FFPE Kit and the Maxwell® HT DNA FFPE Isolation System before quantitation via Droplet Digital PCR.*

**Kits:** Maxwell® RSC DNA FFPE Kit (Cat.# AS1450) or Maxwell® HT DNA FFPE Isolation System (Cat.# A6372)

**Analyses:** Droplet digital PCR

**Sample Type:** FFPE human normal adjacent lung tissue

**Input:** 10µm sections

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 Manuals TM437 and TM539, available at: **www.promega.com/protocols**

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

**Materials Required:**

- Maxwell® RSC DNA FFPE Kit (Cat.# AS1450) or Maxwell® HT DNA FFPE Isolation System (Cat.# A6372)
- Maxwell® RSC Instrument (Cat.# AS4500) or other automated HT platform
- Bio-Rad ddPCR™ Copy Number Variation Assays, Validated

**Protocol**:

1. Purify DNA from FFPE tissue samples using standard protocol for the Maxwell® RSC DNA FFPE Kit outlined in technical manual TM437 or the Maxwell® HT DNA FFPE Isolation System outlined in technical manual TM539.
2. Dilute DNA to a maximum of 10ng/µl based on QuantiFluor® ONE dsDNA System.
3. Set up ddPCR amplification reactions using the Bio-Rad ddPCR™ Copy Number Variation Assays, Validated protocol (10033173 Ver C), with a maximum of 50ng DNA input. Do not digest DNA prior to the reactions and do not use restriction enzymes in the reactions.

**Results:**

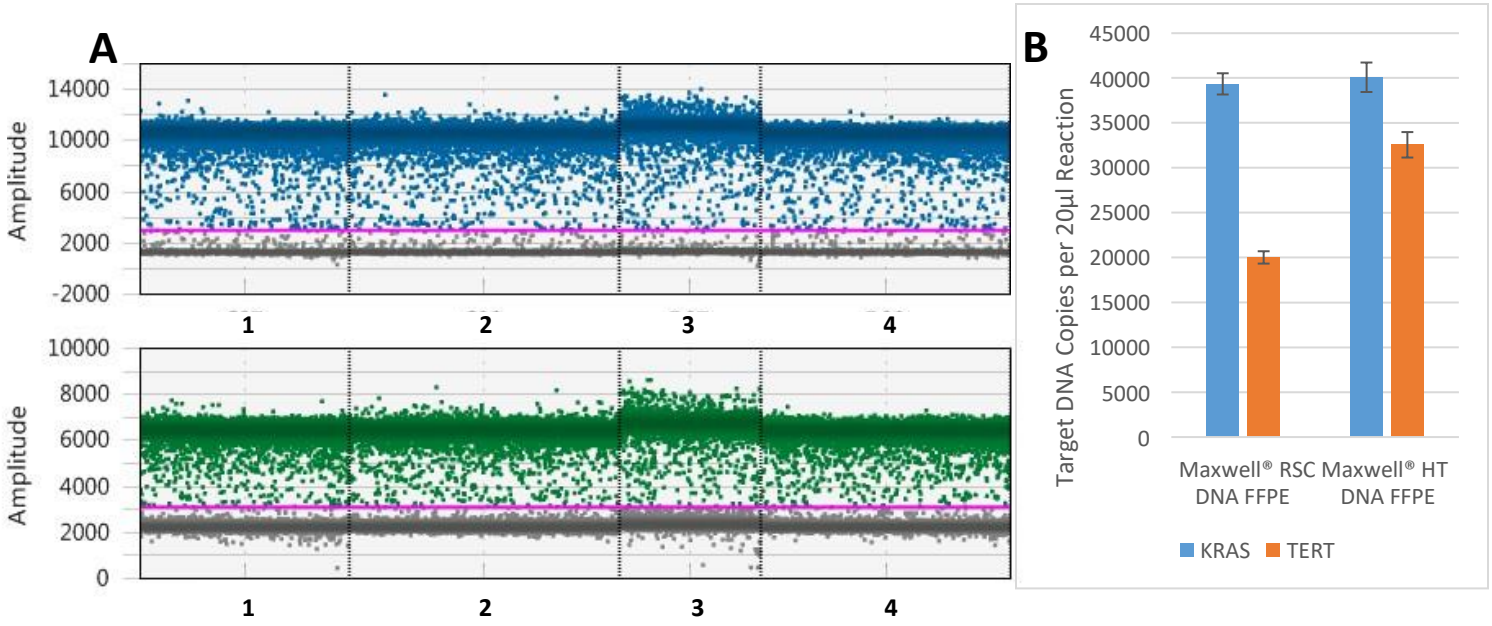


**Figure 1. Droplet digital PCR Quantitation of DNA purified from FFPE human normal lung tissue using the Maxwell® RSC DNA FFPE Kit (Cat.# AS1450) and the Maxwell® HT DNA FFPE Isolation System (Cat.# A6372) run on a Tecan Freedom EVO®.** Resulting DNA was diluted to 10ng/µl based on QuantiFluor® ONE dsDNA System (Cat.# E4870) and amplification reaction mixtures were set up based on the Bio-Rad ddPCR™ Copy Number Variation Assays, Validated protocol, with CNV assays for human KRAS (FAM) and TERT (HEX), using 50ng of DNA input. Panel A (left) displays the 1D droplet plot for ddPCR amplification of KRAS (blue) and TERT (green) targets. The threshold, above which droplets are considered positives (contain target DNA), is shown in pink. Each column (labeled 1-4 beneath each plot) represents a different sample replicate. The Y-axis is relative fluorescence. Droplets with intermediate fluorescence (rain) indicate inefficiently amplified target DNA. Rain is commonly increased in heavily fragmented samples such as FFPE DNA and is not specific to Maxwell® purified samples. Panel B (right) displays the average copies of KRAS and TERT DNA targets per ddPCR reaction calculated using QuantaSoft™ Analysis Pro Software.