

## Exosome Reference Standards

Monitor the performance of your exosome-based diagnostic workflow

### ExoRef-EGFR-T790M

Cat. N. EXO-REF-EGFR-T790M

#### PRODUCT INFORMATION

<b>Category</b>	Exosome-based reference standards, containing cancer-relevant mutations
<b>Format</b>	Dry exosomes
<b>Mutation</b>	65% Allelic Frequency EGFR T790M

#### PROPERTIES

<b>Protein Content</b>	30 $\mu\text{g} \pm 10\%$ [based on Bicinchoninic acid assay (BCA)]
<b>Nanoparticle Tracking Analysis (NTA)</b>	$\geq 1.0\text{E}+11$ particles/ml
<b>Expected Mutation Allelic Frequency</b>	65% $\pm 10\%$
<b>Storage Condition</b>	+ 4°C for 24 months (sealed dry exosomes)

Request your Certificate of Analysis at [tech@exosomics.eu](mailto:tech@exosomics.eu) by citing ExoRef catalogue number and lot n°.

#### EXOSOME PURITY AND INTEGRITY

##### TRANSMISSION ELECTRON MICROSCOPY (TEM)

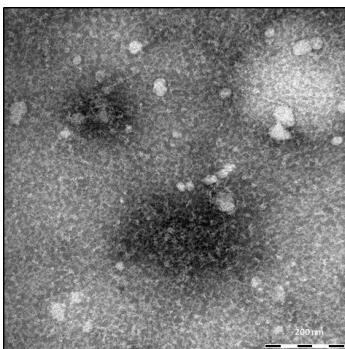


Fig 1. TEM of resuspended ExoRef-EGFR-T790M.

Dried ExoRef-EGFR-T790M were reconstituted in Resuspension Buffer (EXO-REF-RB) and analysed by TEM (Figure 1). Sample purification and drying guarantee exosome stability and integrity. Reference bar is 200 nm.

Exosome purity is determined by measuring nanoparticle concentration (estimated by NTA) and total protein concentration (estimated by BCA), as reported in Webber et al., Journal of Extracellular vesicles, 2013.

## MUTATION LOAD

### DROPLET DIGITAL™ PCR

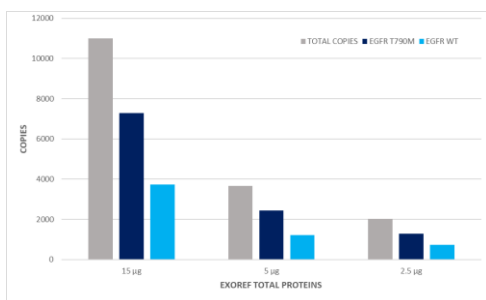


Fig 2. Serial dilutions of resuspended ExoRef-EGFR-T790M.

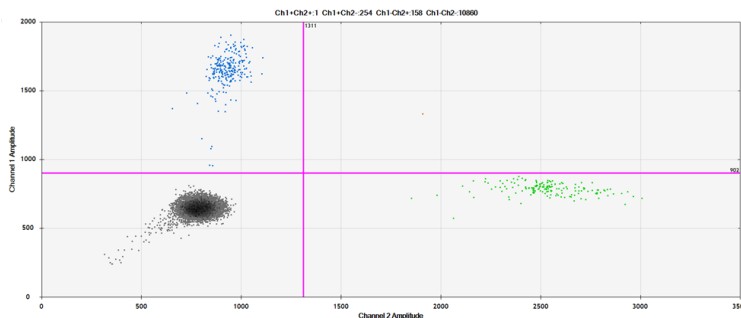


Fig 3. 2D Amplitude of DNA extracted from 5 µg of ExoRef-EGFR-T790M.

EGFR T790M total copies detected by ddPCR in three different serial dilutions (15, 5 and 2.5 µg of total proteins estimated by BCA). Figure 2 confirms high dilutions consistency and sets a detection threshold for EGFR T790M mutated copies at 5 µg of total protein assessed by BCA (estimated range of total mutated copies 2500 ± 10%). Figure 3 shows a ddPCR 2D Amplitude plot in which 5 µL of DNA extracted from 5 µg of ExoRef-EGFR-T790M has been loaded.

## ExoRef RESUSPENSION PROCEDURE

### IMPORTANT: Centrifuge tube prior to opening to prevent loss of product

ExoRef- Exosome Standards are dried and shipped at room temperature.

1. We recommend to spin down (5000 g for 30 sec) ExoRef tube before reconstitution with Resuspension Buffer (EXO-REF-RB). The dry pellet may dislodge during shipping.
2. Resuspend the pellet of ExoRef by adding 30 µL of Resuspension Buffer (EXO-REF-RB).
3. Vortex vigorously for 1 min and centrifuge at 5000 g for 30 seconds.
4. Immediately before use, mix the entire volume by slowly pipetting for at least ten times.

Storage: Properly stored dried exosomes are stable at 4°C for 24 months. Once resuspended, ExoRef can be stored at -20°C for 3 months. Aliquoting is recommended since freeze-and-thaw cycles reduce the quality of the sample.

### CUSTOMER SUPPORT

CoA Request	tech@exosomics.eu
Technical support	support@exosomics.eu
Orders	orders@exosomics.eu