# ipsogen® NPM1 MutaQuant® Standards

### Standards for real-time quantitative RT-PCR and PCR of the mutated NPM1 gene

The *ipsogen* NPM1 MutaQuant Standards are intended to be used in research studies for the accurate quantification of mutated NPM1 genes in RNA or genomic DNA preparations from biological samples.

Genomic DNA preparation and RNA extraction should be performed with recommended procedures (e.g., QIAGEN® QIAamp® DNA Mini Kit, cat. no.51304; RNeasy® Mini Kit, cat. no. 74104 or RNeasy Midi Kit, cat. no. 75144; or Life Technologies TRIzol®, cat. nos. 15596-026 and 15596-018). The performance of the RT-PCR assay is dependent on the concentration and quality of input RNA. We therefore recommend qualifying purified RNA, prior to downstream analysis, by agarose\* gel electrophoresis, Agilent® BioAnalyzer®, or spectrophotometry.

Using standards with a known number of molecules can establish a standard curve to determine the precise amount of target transcript present in the test sample.

### **Product description**

- The *ipsogen* NPM1 MutaQuant Standards are intended to provide calibration for the quantification of individual single polymorphisms
- Each standard offers 5 dilutions of the cloned mutated gene to establish standard curves
- The ipsogen NPM1 MutaQuant Standards are available for specific detection of polymorphisms from RNA and genomic DNA (see Table 1)
- Volume of standard per tube is 50 µl, sufficient for 8 reactions

Table 1. ipsogen MutaQuant Standards

Cat. no.	Number of tubes	Dilutions (copies in 5 µl)
677491	5	10 <sup>1</sup> , 10 <sup>2</sup> , 10 <sup>3</sup> , 10 <sup>5</sup> , 10 <sup>6</sup>
677591	5	10 <sup>1</sup> , 10 <sup>2</sup> , 10 <sup>3</sup> , 10 <sup>5</sup> , 10 <sup>6</sup>
677691	10	10 <sup>1</sup> , 10 <sup>2</sup> , 10 <sup>3</sup> , 10 <sup>5</sup> , 10 <sup>6</sup>
677791	10	10 <sup>1</sup> , 10 <sup>2</sup> , 10 <sup>3</sup> , 10 <sup>5</sup> , 10 <sup>6</sup>
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### Shipping and Storage

The standards are shipped on dry ice and must be stored at -30°C to -15°C upon receipt. Vortex and centrifuge the tubes before opening. For frequent use, we recommend preparing aliquots of more convenient volumes.

<sup>\*</sup> When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles



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### **Safety Information**

When working with chemicals, always wear a suitable laboratory coat, disposable gloves, and protective goggles. For more information, please consult the appropriate safety data sheets (SDSs). These are available online in convenient and compact PDF format at <a href="https://www.qiagen.com/safety">www.qiagen.com/safety</a> where you can find, view, and print the SDS for each QIAGEN kit and kit component. Discard sample and assay waste according to your local safety regulations.

### **General precautions**

Use of quantitative RT-PCR tests to determine transcript levels requires both the reverse transcription of mRNA and amplification of the generated cDNA by PCR. Therefore, use extreme caution to prevent:

- RNase and DNase contamination
- Carryover contamination by RNA, DNA, control reagents, or PCR resulting in false positive signals

We therefore recommend the following:

- Use nuclease-free labware (e.g., pipets, pipet tips, reaction vials) and wear gloves when performing assays
- Use fresh aerosol-resistant pipet tips for all pipetting steps to avoid cross-contamination of the samples and reagents
- To avoid cross-contamination from carryover, transfer solutions for an experiment into fresh tubes instead of directly pipetting from a stock solution

The ipsogen NPM1 MutaQuant Standards are intended for research use only. Not for use in diagnostic procedures.

### **Quality Control**

Quality control of the *ipsogen* NPM1 MutaQuant Standards was performed on a LightCycler 480 instrument, quantitatively with vector-specific primers, and qualitatively with insert-specific primers. The standards are manufactured according to ISO 13485 standard. Certificates of Analysis are available upon request at <a href="https://www.qiagen.com/support/">www.qiagen.com/support/</a>.

#### Procedure

To obtain a standard curve to calibrate the mutated gene in sample RNA or genomic DNA, use the corresponding gene standard with the dedicated primers and probes (extra material not supplied).

We recommend that each dilution of a standard be used in duplicate (10 reactions) to establish the standard curve.

For information on primers and probes in *ipsogen* NPM1 mut A cDNA MutaQuant Standards (cat. no. 677591) and *ipsogen* NPM1 mut B&D cDNA MutaQuant Standards (cat. no. 677791) please refer to:

Gorrello, P. et al. (2006) Quantitative assessment of minimal residual disease in acute myeloid leukemia carrying nucleophosmin (NPM1) gene mutations. Leukemia **20**, 1103.

Trademarks: QIAGEN®, ipsogen®, MutaQuant®, QIAamp®, RNeasy® (QIAGEN Group); Agilent® BioAnalyzer® (Agilent Technologies); TRIzol® (Molecular Research Center, Inc.).
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