# illumina

## NūPCR™ Reagents for Gene Expression Analysis

NuPCR reagents provide gold standard specificity and simplified multiplexing at a budget-friendly price for superior results in your qPCR gene expression analysis studies.

#### Highlights

- Superior Sensitivity and Specificity Unique enzymatic probe produces a signal only in the presence of the exact target sequence
- Time-Saving Workflow State-of-the-art algorithm enables fast assay design and pre-optimized multiplexing studies
- Increased Productivity
  Economical prices enable you to perform more
  experiments within the same budget

#### Introduction

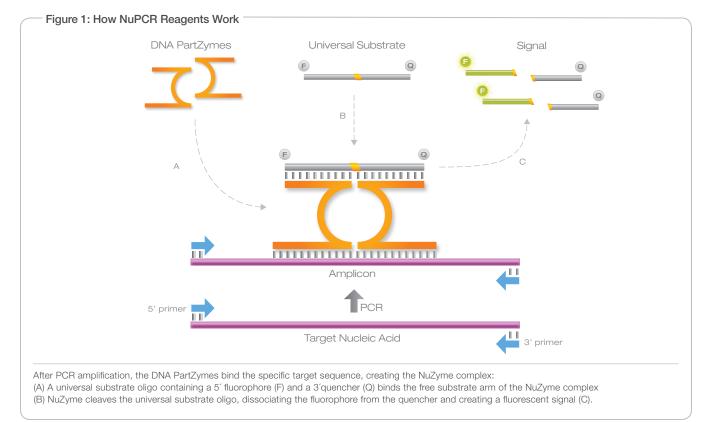
Quantitative real-time PCR (qPCR) is a widely used technique for extremely sensitive analysis of gene expression. Although effective, current probe–based assays can be limited in their performance ability due to tedious design and optimization procedures and expensive reagents. NuPCR qPCR reagents overcome these challenges, offering researchers superior results in less time at a lower cost.

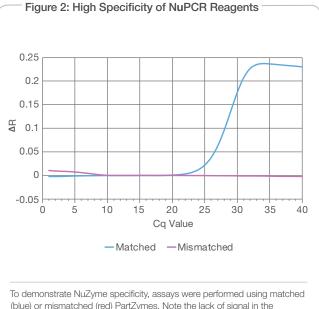
#### How It Works

The key to the NuPCR reaction is the unique NuZyme™ complex. This three-part probe consists of two PartZyme™ oligos (PartZyme A and PartZyme B) and universal substrate oligo containing a 5′ fluorophore and a 3′ quencher that binds to the PartZymes. Once the target is amplified via PCR, PartZymes A and B bind to the specific target sequence forming the NuZyme complex (Figure 1). The labeled universal substrate then binds the free substrate region of the NuZyme complex. The inherent catalytic activity of NuZyme cleaves the universal substrate, dissociating the fluorophore from the adjacent quencher, creating detectable fluorescence levels. NuZyme will continue to bind and cleave universal substrate oligos, leading to amplification of the fluorescent signal.

### **High Specificity**

Unlike other probes that can produce a fluorescent signal when partially bound, NuPCR reagents only produce fluorescence when all three parts of the probe combine. Since this occurs only in the presence of the specific target, all signal is due to





(blue) or mismatched (red) PartZymes. Note the lack of signal in the mismatched reaction.

the target DNA, ensuring the highest specificity of any PCR probe (Figure 2). For even higher data integrity, NuPCR reagents can be easily duplexed, enabling detection of the target and reference gene in the same well (Figure 3).

## **High Sensitivity**

The NuZyme complex acts as a multi-turnover enzyme, meaning that it can bind and release multiple universal probes, resulting in higher sensitivity (Figure 4) and a more efficient PCR experiment (Figure 5). NuPCR reagents can detect targets present at less than 10 copies with high confidence. In fact, targets are consistently detected by NuPCR earlier in the cycle count than other probes (Figure 6).

### **Easy Probe Design**

To simplify primer design and ordering, Illumina offers the web-based DesignStudio<sup>™</sup> software. Users input information about their project, including species, genome source (NCBI or UCSC), and Genome Build. Target sequences can be input as RefSeq IDs, gene symbol, direct sequence, or uploaded from a .csv file. DesignStudio accesses the latest genome data to ensure probe integrity. Once designed, assays can be ordered directly from DesignStudio.

#### **Duplex Designs**

For faster, easier duplexing, DesignStudio bioinformatically validates the probes and assays, ensuring their compatibility. There's no need to spend time and resources optimizing each experiment.

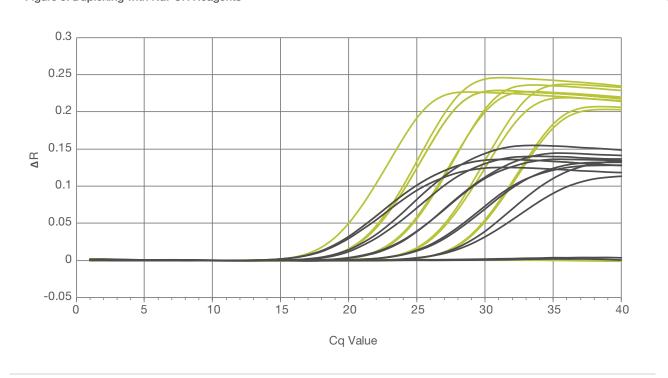
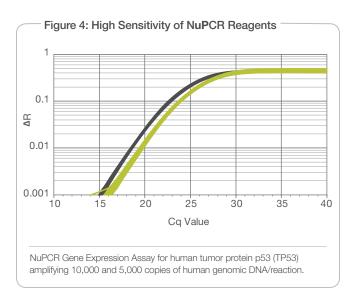
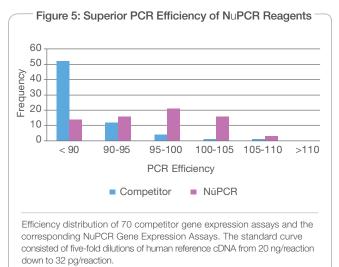


Figure 3: Duplexing with NuPCR Reagents

ROX-labeled (brown) NuPCR Gene Expression Assay for human profilin 1 (PFN1) multiplexed with Q670-labeled (pink) NuPCR Gene Expression Assay for human actin, beta (ACTB). Assays amplify a five-fold dilution series of cDNA prepared from Human Universal Reference RNA from 20 ng/reaction down to 32 pg/reaction. Real-time PCR was performed using NuPCR Gene Expression Master Mix.





#### Bench-Ready Workflow

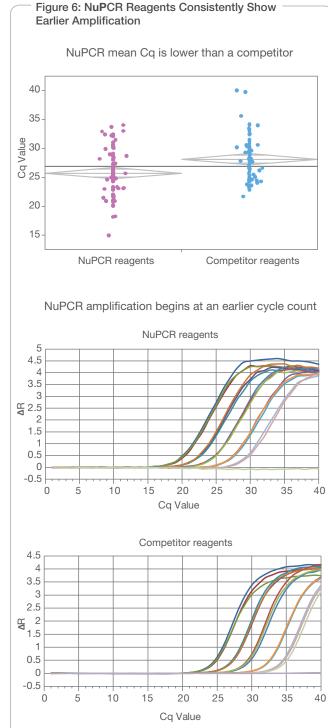
NuPCR assays are custom designed for each target sequence. Assays are supplied ready to use with your choice of FAM, HEX, ROX, or Quasar 670 (Q670) dye. In addition, the polymerase, nucleotides, and other amplification reagents are provided in a master mix solution. To use, simply load the PCR plates with the assay, master mix, and DNA and run.

#### **Cross-Platform Compatibility**

NuPCR assays produce superior results on any PCR platform. There's no need to purchase any additional equipment or learn new methods.

#### Cost

By eliminating the need to optimize, simplifying duplexing, and providing higher specificity and sensitivity, NuPCR assays save you time, resources, and money. In addition, the budget-friendly price is a fraction of the cost of expensive competitor probes.



#### Summary

NuPCR assays enable more accurate gene expression analysis with higher specificity and sensitivity, an easier probe design, and a lower cost than other methods. Amplify your story. Get the full details at www.illumina.com/nupcr.

Product	No. Reactions	Catalog No.
NuPCR Assays		
NuPCR Gene Expression Assays - FAM	250	EC-309-1001
	500	EC-309-1002
	2000	EC-309-1003
NuPCR Gene Expression Assays - HEX	250	EC-309-1011
	500	EC-309-1012
	2000	EC-309-1013
NuPCR Gene Expression Assays - ROX	250	EC-309-1021
	500	EC-309-1022
	2000	EC-309-1023
NuPCR Gene Expression Assays - Q670	250	EC-309-1031
	500	EC-309-1032
	2000	EC-309-1033
NuPCR Master Mixes		
NuPCR Gene Expression Master Mix	250	EC-305-1001
	500	EC-305-1002
	2000	EC-305-1003

Illumina • 1.800.809.4566 toll-free (U.S.) • +1.858.202.4566 tel • techsupport@illumina.com • www.illumina.com

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