# Gold-standard TaqMan Assay technology enables highly accurate quantitation for copy number variation

Applied Biosystems<sup>™</sup> TaqMan<sup>™</sup> Copy Number Assays are the gold standard for accurate target quantitation, making them ideal for validating and screening specific copy number variations. These assays combine Applied Biosystems<sup>™</sup> TaqMan<sup>™</sup> Assay probe 5′ nuclease chemistry with Applied Biosystems<sup>™</sup> real-time PCR instruments to form a method for obtaining specific, reproducible, and easy-to-interpret results. TaqMan Copy Number Assays are run together with an Applied Biosystems<sup>™</sup> TaqMan<sup>™</sup> Copy Number Reference Assay in a duplex real-time polymerase chain reaction (PCR). Results are analyzed by the relative quantitation method using Applied Biosystems<sup>™</sup> CopyCaller<sup>™</sup> Software. On average, setup to primary analysis takes 3 to 4 hours, enabling users to obtain results with speed, confidence, and reproducibility.

## Features and benefits

#### 1. Performance

#### **Accuracy**

Assays are developed with a high degree of bioinformatics to overcome homology issues and help ensure accurate detection of a wide range of specific variations. Gain confidence in studying the right copy number variation to generate correct results. CopyCaller Software provides the calculated copy number and predicted copy number, along with confidence value and z-score quality metrics.



Applied Biosystems™ TaqMan™ Copy Number Assays, Custom Plus TaqMan™ Copy Number Assays, and Custom TaqMan™ Copy Number Assays.

#### **Specificity**

TaqMan Copy Number Assays use Applied Biosystems
TaqMan Assay chemistry and Applied Biosystems real-time
PCR instruments to obtain specific and reproducible copy
number results.

The TaqMan probe contains a FAM™ reporter dye on the 5′ end and a minor groove binder (MGB) and nonfluorescent quencher (NFQ) on the 3′ end. The MGB:

- Stabilizes the hybridized probe, resulting in stronger probe binding
- Allows the probe to be shorter than traditional duallabeled probes, enabling you to effectively target difficult sequences and increase specificity

## 2. Design and coverage

TaqMan Copy Number Assays are designed using Applied Biosystems<sup>™</sup> assay design algorithms, which are optimized to produce high-performing copy number assays.



TaqMan Copy Number Assays include predesigned collections for both human and mouse genomes. The human collection includes more than 1.6 million predesigned assays with genome-wide coverage targeting: known genes (exons, introns, and junctions), copy number variant (CNV) sequences within the Database of Genomic Variants (DGV), and extragenic/nongene regions. The mouse collection includes more than 180,000 assays targeting gene exons. Predesigned assays to common vector marker and reporter genes are also available for transgenic studies. Predesigned assays have been checked for genome quality as well as reference assay compatibility.

Custom Plus TaqMan Copy Number Assays are the optimal solution for studying variation in human and mouse genomic regions of interest for which a predesigned assay is not available. A target range is defined on the genome map, then premasked targets are created and submitted to the assay design pipeline. Benefits include genome quality checks and human/mouse reference assay compatibility checks.

Custom TaqMan Copy Number Assays are an option for additional targets of interest. The Applied Biosystems™ GeneAssist™ Copy Number Assay Tool enables you to submit your own premasked custom target sequences for assay design or primer/probe pair sequences for assay formulation. Custom assay designs do not go through genome quality checks, but can be compared with the human/mouse reference assays for compatibility in duplex reactions.

#### 3. Quality

Whether testing a large or small number of targets against a few or hundreds of samples, having confidence in the data you produce is paramount. The Applied Biosystems™ Copy Number Assay Design Pipeline has been optimized to create high-performing copy number assays based on R&D wet lab validation of assays. Additionally, we have developed the TaqMan™ Assays qPCR Guarantee to provide you with confidence in the data you generate with a predesigned TaqMan Assay. We guarantee that TaqMan Copy Number Assays will perform to your satisfaction. If you are not satisfied with the performance of a TaqMan Assay, we'll replace it at no cost, or credit your account for the purchase price of the assay.\* Learn more at

# thermofisher.com/taqmanguarantee

#### 4. Convenience

Choose only the specific assays you need for your project from the large variety of predesigned assays, select a genomic target region of interest for Custom Plus Assay design, or submit your target sequence for Custom Assay design. The custom workflow also allows for the submission of primer/probe sequences for assay formulation.

TaqMan Copy Number Assays are available in made-to-order small, medium, and large sizes.

TaqMan Copy Number Assays (excluding Reference Assays) are shipped in a preformulated liquid format at ambient temperature and arrive ready to use (no need to resuspend different components). There is no optimization required; just use universal cycling conditions for every assay.

#### **Ordering method**

TaqMan Copy Number Assays and other related products can be ordered at **thermofisher.com/ordertaqman** 

For help with online ordering, please refer to the GeneAssist Copy Number Assay Workflow Builder for predesigned, Custom Plus, and Custom Assays.

TaqMan Copy Number Assays are provided with support documents that can be accessed at

## thermofisher.com/taqmanfiles

- Protocol for the TaqMan Copy Number Assays (PDF)
- Quick reference card for the TaqMan Copy Number Assays (PDF)
- Protocol for CopyCaller Software (PDF)
- Quick reference card for CopyCaller Software (PDF)
- Product insert for the TaqMan Copy Number Assays (PDF)
- Assay information file (AIF)
- Data sheet (PDF)
- Safety data sheet(s) (PDF)
- "Understanding your shipment" document (PDF)

# **Applied Biosystems instrument compatibility**

ViiA<sup>™</sup> 7 Real-Time PCR System
7900HT Fast Real-Time PCR System
7500 Fast Real-Time PCR System
StepOnePlus<sup>™</sup> Real-Time PCR System
QuantStudio<sup>™</sup> 3, 5, 7 & 12K Flex Real-Time PCR Systems

References and additional reading

**Application note:** Design pipeline and validation of TaqMan Copy Number Assays (135AP03-01)

**Protocol:** TaqMan Copy Number Assays (4397425)

Quick reference card: TaqMan Copy Number Assays

(4397424)

Quick reference card: CopyCaller Software (4400043)

User guide: CopyCaller Software (4400042)

**Additional product information** 

Online resources page: thermofisher.com/cnv

Web resources

Database of Genomic Variants: dgv.tcag.ca

Sanger Institute Copy Number Variation Project: sanger.

ac.uk

UCSC Genome Bioinformatics site: genome.ucsc.edu

Ensembl: ensembl.org dbVar: ncbi.nlm.nih.gov

**GeneAssist Copy Number Assay Workflow Builder** 

The GeneAssist Copy Number Assay Workflow Builder

enables users to search for predesigned human, mouse, and marker/reporters assays, to select a genomic human or mouse target region for Custom Plus Assay design, or to submit additional targets of interest for standard custom design/synthesis.

# CopyCaller Software

CopyCaller Software was developed specifically for TaqMan Copy Number Assay data analysis to enable users to obtain results quickly, robustly, and definitively (Figure 1). This free, easy-to-use software utilizes a graphical interface and quickly calculates the raw and possible copy numbers for a set of samples. It also gives a confidence value for each copy number call and has outlier removal functionality. CopyCaller Software can perform relative quantitation analysis using a known calibrator sample or without a calibrator sample.

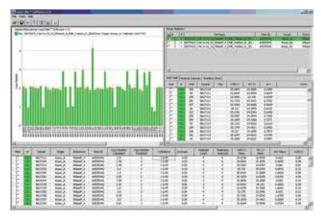


Figure 1. Determining copy number using CopyCaller Software.

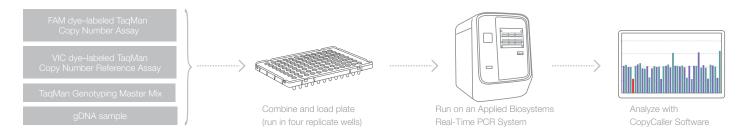


Figure 2. The complete TaqMan Copy Number Assay workflow. TaqMan Copy Number Assays have a simple workflow, and average setup to primary analysis takes only 3 to 4 hours (including a ~2 hour PCR run). Each copy number quantitation reaction contains four components: a TaqMan Copy Number Assay, a TaqMan Copy Number Reference Assay, TaqMan™ Master Mix, and purified genomic DNA sample, run in four replicate wells.



# **Ordering information**

TaqMan Copy Number Assays		Number of reactions		Cat. No.		
Assay scale	Concentration	384-well, 10 μL	96-well, 20 μL	Predesigned Assays	Custom Plus Assays	Custom Assays
Small	20X	720	360	4400291	4442487	4400294
Medium	20X	1,500	750	4400292	4442520	4400295
Large	60X	5,800	2,900	4400293	4442488	4400296

TaqMan Copy Numb	er Reference Assays	Number of react	Cat. No.	
Description	Concentration	384-well, 10 μL	96-well, 20 μL	
Human assays				
RNase P, 750 rxns	20X (1 tube)	1,500	750	4403326
RNase P, 3,000 rxns	20X (4 tubes)	6,000	3,000	4403328
TERT, 750 rxns	20X (1 tube)	1,500	750	4403316
TERT, 3,000 rxns	20X (4 tubes)	6,000	3,000	4403315
Mouse assays				
Tfrc, 750 rxns	20X (1 tube)	1,500	750	4458366
Tfrc, 3,000 rxns	20X (4 tubes)	6,000	3,000	4458367
Tert, 750 rxns	20X (1 tube)	1,500	750	4458368
Tert, 3,000 rxns	20X (4 tubes)	6,000	3,000	4458369

# Find out more at thermofisher.com/cnv



<sup>\*</sup>Subject to certain restrictions and terms and conditions. Go to thermofisher.com/taqmanguarantee for details.