

Taqman Master Mix-Multiplex

(Catalog# M1125-500; 2X QPCR Master Mix for 500 reactions (20 µl); 4 X 1.25 ml; Store at -20°C)

I. Introduction:

Taqman Master Mix-Multiplex is designed specifically for TaqMan probe-based real-time PCR analysis of DNA samples. Taqman 2X QPCR Master Mix is designed for high throughput quantitative PCR using TaqMan® probe-based chemistry. Available with the option of ROX or fluorescein as the internal passive reference dye, BioVision's TaqProbe 2X QPCR Master Mix offers superb performance in sensitivity and signal-to-noise ratio. The multiplex formulation supports quantitative amplification and detection of up to four targets simultaneously with consistent and reliable results. Due to variations in QPCR instrument performance, we offer different Taqman QPCR Master Mix formulations optimized for different machines. Please use BioVision's QPCR Mix that will be most compatible with your choice of a particular instrument/model. Taqman 2X Master Mix-Multiplex (Cat# M1125-500) is suited for Multiplex TaqMan assay in the following instruments including ABI® 7000,7300,7500,7700,7900; StepOne™, StepOnePlus™; BioRad® iCycler®, iQ™5, CFX96, Opticon™2; Chromo® 4; Corbett Rotor-gene® 6000,3000; Stratagene® Mx3000, Mx4000; Roche LightCycler® 2.0, 480.



II. Key Features:

- Enable streamlined protocol in a simple reaction set-up
- Allow accurate quantification of a variety of gene targets
- Reduce pipetting steps to minimize the risk of contamination
- Compatible with most real-time PCR instruments

III. Applications:

- Gene-expression analysis
- Gene knockdown validation
- SNP genotyping assays
- CHiP
- Copy number variation
- Microarray validation
- High throughput applications
- Virus detection and quantification

IV. User Supplied Reagents and Equipment:

- PCR Tubes
- QPCR Instrument
- Pipettes
- Water, Nuclease-free
- Primers (forward and reverse)
- Template DNA

V. Shipment and Storage:

Upon arrival, the Taqman Master Mix should be stored at -20°C and protected from light. After each experiment, the leftover mix (completely thawed and thoroughly homogenized) can be stored at 4°C if it is to be used within the next 3 months. Taqman Master Mix is stable for 1 year from the date of shipping when stored and handled properly

VI. Reagent Preparation and Storage Conditions:

- Aliquot reagents to avoid contamination and to avoid repeated freeze-thaw cycles.
- The Taqman Master Mix components are light sensitive and therefore, avoid prolonged exposure to direct light.
- Ideally, start the PCR as soon as the reaction mixture is prepared. If not, then make sure that the reaction mixture is kept chilled till starting up the PCR.
- Primer concentration should not be high, a concentration of 100 nM to 300 nM of each primer usually gives the best results.
- A very effective way to get tight Ct among replicates is to reduce pipetting error, this can be achieved by: preparing amplicon specific pre-mix, using repeating pipet, and keeping pipetting volume in manufacture suggested range.
- For optimal results, it is recommended that the primers are 18-22 nucleotides in length with a T_m of 58°C-60°C and the size of target is about 100-250 bp.

VII. QPCR Protocol:

1. Thaw the Taqman 2X Master Mix-Multiplex, template DNA, primers and nuclease-free water on ice. Mix each solution thoroughly. Prepare a reaction Master Mix as follows:

Components	Volume (20 µl)	Volume (25 µl)	Volume (50 µl)	Final concentration
Taqman Master Mix-Multiplex	10 µl	12.5 µl	25 µl	1X

Forward Primer	Varies	Varies	Varies	100-500 nM
Reverse Primer	Varies	Varies	Varies	100-500 nM
Template DNA	Varies	Varies	Varies	≤10 ng/reaction
TaqMan Probe	Varies	Varies	Varies	100 - 300 nM
Water, Nuclease-free	to 20 µl	to 25 µl	to 50 µl	-
Total Volume	20 µl	25 µl	50 µl	-

2. Perform QPCR reactions using the following 3-step cycling program:

Step	Temperature	Duration (Standard)	Duration (Fast)	Cycle(s)
Enzyme Activation	95°C	10 min	10 min	1
Denaturation	95°C	15 secs	3 secs	40
Annealing/Extension	60°C	60 secs	30 secs	

VIII. Related Products:

BV Product Name	BV Cat. No.
Jade™ Master Mix	M1105-500
Jade™ Mix-iCycler	M1106-500
Jade™ Mix-Low ROX	M1107-500
Jade™ Mix-ROX	M1108-500
Jade Express™ Mix	M1109-500
Jade Express™ Mix-iCycler	M1110-500
Jade Express™ Mix-Low ROX	M1111-500
Jade Express™ Mix-ROX	M1112-500
Jade Smart™ Mix	M1113-500
Jade Smart™ Mix-iCycler	M1114-500
Jade Smart™ Mix-Low ROX	M1115-500
Jade Smart™ Mix-ROX	M1116-500
Jade miRNA™ Mix	M1117-500
Jade miRNA™ Mix-iCycler	M1118-500
Jade miRNA™ Mix-Low ROX	M1119-500
Jade miRNA™ Mix-ROX	M1120-500
Taqman Master Mix	M1121-500
Taqman Master Mix-iCycler	M1122-500
Taqman Master Mix-Low ROX	M1123-500
Taqman Master Mix-ROX	M1124-500
Taqman Master Mix-Multiplex	M1125-500
QPCR Positive Control Kit	M1126-100

FOR RESEARCH USE ONLY! Not to be used on humans.