



ViPrimePLUS Lyophilized qPCR Master Mix

Product code: QLMM05
Packsize: 150 reactions

DESCRIPTION

ViPrimePLUS Lyophilized qPCR Master Mix is next generation first choice mix for a fast and easy real-time PCR reaction set up. The master mix has been freeze-dried to produce a room temperature stable preparation. The improved formulation of master mix contains pure Taq DNA Polymerases, highest quality MgCl₂ and buffer components at optimal concentrations. Taq DNA Polymerases in the master mix provide mechanism which releases more active enzyme and requires much shorter activation time to achieve excellent results in reaction efficiency, correlation coefficient and slope.

ViPrimePLUS Lyophilized qPCR Master Mix can be used to amplify any DNA template including genomic, cDNA and viral sequences. The improved formulation of qPCR master mix can detect the extremely low copy number targets very specifically with high efficiency. The qPCR master mix has been designed to prevent and reduce the formation of primer dimers and non-specific products leading to optimum sensitivity and specificity.

ViPrimePLUS Lyophilized qPCR Master Mix supplied with re-suspension buffer and ROX dye to be used with most of real-time PCR instruments which include hardware platforms that need ROX as a passive reference dye. The improved sensitivity and consistency of ViPrimePlus Lyophilized qPCR Master Mix in standard cycling conditions allow for industry leading performance in fast cycling conditions.

APPLICATIONS

All kinds of DNA sample material suited for qPCR amplification can be used.

FEATURES

- Lyophilized form – room temperature stable preparation
- Fast and easy real-time PCR reaction set up
- Rapid extension rate for early Ct values
- Highest sensitivity and specificity
- Increased limit of detection
- Compatible on most of the real-time PCR platforms

COMPONENTS

- 3 x Lyophilized qPCR Master Mix
- 1 x Lyophilized ROX
- 4 x Re-suspension buffer

STORAGE

Store at ambient temperature on arrival and stable at ambient temperature up to the expiry date.

Once re-suspended with the provided buffer, the master mix should be stored at -20°C up to the expiry date stated. Keep in aliquot to reduce freeze-thaw cycles.

QUALITY CONTROL

As part of the ISO9001:2008 quality assurance systems, each lot of ViPrimePLUS Lyophilized qPCR Master Mix has been tested against predetermined specifications to ensure consistent product quality and highest levels of performance and reliability.

LIMITATION OF USE

For research use only. Not recommended or intended for diagnosis of disease in humans or animals. Do not use internally or externally in humans or animals.

PROTOCOL

Master Mix Preparation

1. Re-suspend each tube of Lyophilized qPCR Master Mix with 525µl of re-suspension buffers.
 - a. Do not replace the re-suspension buffer with water or any other buffer.
 - b. The master mix is ready to be used as a 2X qPCR Master Mix.
2. ROX is required for platforms that use ROX as a passive reference guide. If ROX is required then follow the steps below.
 - a. Re-suspend the tube of lyophilized ROX in the correct volume of re-suspension buffer according to the table below.
 - b. Add the re-suspended ROX to each master mix at the correct level according to the table below.

Real-time PCR platforms	ROX re-suspension volume	ROX addition per tube of master mix
Applied Biosystems 7000, 7700, 7900, 7300 StepOne, StepOnePLUS and ViA7 platforms, Roche capillary Lightcyclers.	100µl	20µl
All Stratagene platforms	200µl	15µl
Applied Biosystems 7500 platform	700µl	10µl
All other machines	Not Required	Not Required

Reaction Preparation

1. Keep the qPCR master mix protected from light until use it.
2. Aliquot the qPCR master mix to minimize freeze-thaw cycles and light exposure.
3. Reserve plate positions for positive (control DNA) and negative (water or buffer) controls.
4. When preparing mixes, always calculate the volume according to the number of reactions that needed plus one extra.
5. After the mixture is done and aliquot into tubes, place into qPCR platform.

SUGGESTED MIXTURE

- a. When using ViPrimePLUS gene detection kits:

Components	Reaction (1X)
ViPrimePLUS Lyophilized qPCR Master Mix	10µl
Primer/Probe Mix	1µl
Template (25ng)	5µl
Nuclease free water	4µl
Final Volume	20µl

- b. When using user supplied primers and probe:

Components	Reaction (1X)
ViPrimePLUS Lyophilized qPCR Master Mix	10µl
Primers (6pmols Forward & Reverse)	X µl
Probe (3pmols)	X µl
Template (25ng)	X µl
Nuclease free water	X µl
Final Volume	20µl

CYCLING PROGRAM

- a. For Taqman® gene detection kits

Step	Cycles	Temp	Time
Enzyme activation	1	95°C	2mins
Denaturation	50	95°C	10secs
Data Collection*		60°C	60secs

*Fluorogenic data should be collected during this step through the FAM channel.

- b. For SYBR® green detection kits

Step	Cycles	Temp	Time
Enzyme activation	1	95°C	2mins
Denaturation	50	95°C	10secs
Data Collection*		60°C	60secs
Melt Curve**			

*Fluorogenic data should be collected during this step through the SYBR® green channel.

**A post PCR run melt curve can be used to prove the specificity of primers. See the manufactures instructions for your hardware platform.

PREVENTION OF CONTAMINATION

qPCR amplification is a very sensitive DNA amplification reaction; therefore extra care should be taken to eliminate the possibility of contamination with any foreign DNA templates.

- Use separate clean areas for preparation of samples, reaction mixture and for cycling.
- Clean lab bench and equipments periodically with 3% hydrogen peroxide or 70% ethanol.
- Wear fresh gloves. Change gloves whenever suspect that they are contaminated.
- Use sterile tubes and pipette tips with aerosol filters for PCR reaction set up.
- With every PCR reaction set up, perform a contamination control reaction without template RNA.

LEGAL DISCLAIMER

Purchase of product does not include a license to perform any patented applications; therefore it is the sole responsibility of users to determine whether they may be required to engage a license agreement depending upon the particular application in which the product is used.

WARRANTY AND LIMITED LIABILITY

The performance characteristics stated were obtained using the assay procedures in the insert. Failure to comply with the instructions may derive inaccurate results. In such event, manufacturer disclaims all warranty expressed, implied or statutory including the implied warranty of merchantability and the fitness of use.

The manufacturer will not be liable for any damage caused by misuse, improper handling and storage; non-compliance with precautions and procedures, and damages caused by events occurring after the product is released.

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