



# TETRA™ 2X qPCR MASTERMIX

CAT NO. YS-qP-TMM-5/10/20

5ml / 10ml / 20ml

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VERSION 3.0

For Research Use Only



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## INTENDED USE

YouSeq Tetra™ MasterMix is a convenient 2X mix designed for real-time PCR applications. The MasterMix formulation has been specifically engineered for highly processivity, powerful multiplexing capability and to be highly resistant to inhibitors.




The high accuracy and enhanced 5'-3' exonuclease activity of our Polymerase result in very efficient probe cleave activity for superior probe-based assay performance. The enzyme is ideally suited to applications where sensitivity and speed to result is key. YouSeq Tetra™ MasterMix uses an advanced buffer system including dNTPs, Mg and enhancers, enabling efficient PCR of a wide range of target types from all sort of sequence types including small GC-rich genomes with minimal or no optimisation required.

## KIT CONTENTS




### Standard Packs:

|                          | Cap Colour  | 5 ml Pack Size | 10 ml Pack Size | 20 ml Pack Size |
|--------------------------|---|----------------|-----------------|-----------------|
| Tetra™ 2X qPCR MasterMix |  | 5 x 1 ml       | 10 x 1 ml       | 20 x 1 ml       |




### Low ROX:

|                          | Cap Colour  | 5 ml Pack Size | 10 ml Pack Size | 20 ml Pack Size |
|--------------------------|---|----------------|-----------------|-----------------|
| Tetra™ 2X qPCR MasterMix |    | 5 x 1 ml       | 10 x 1 ml       | 20 x 1 ml       |
| ROX Passive Reference    |   | 10 µl          | 10 µl           | 10 µl           |
| DNase/RNase Free Water   |  | 1.5 ml         | 1.5 ml          | 1.5 ml          |

### Mid ROX:

|                          | Cap Colour  | 5 ml Pack Size | 10 ml Pack Size | 20 ml Pack Size |
|--------------------------|---|----------------|-----------------|-----------------|
| Tetra™ 2X qPCR MasterMix |  | 5 x 1 ml       | 10 x 1 ml       | 20 x 1 ml       |
| ROX Passive Reference    |  | 10 µl          | 10 µl           | 2 x 10 µl       |
| DNase/RNase Free Water   |  | 1.5 ml         | 1.5 ml          | 1.5 ml          |

### High ROX:

|                          | Cap Colour  | 5 ml Pack Size | 10 ml Pack Size | 20 ml Pack Size |
|--------------------------|---|----------------|-----------------|-----------------|
| Tetra™ 2X qPCR MasterMix |  | 5 x 1 ml       | 10 x 1 ml       | 20 x 1 ml       |
| ROX Passive Reference    |  | 3 x 10 µl      | 5 x 10 µl       | 10 x 10 µl      |
| DNase/RNase Free Water   |  | 1.5 ml         | 1.5 ml          | 1.5 ml          |

## ROX (PLATFORM DEPENDENT)

ROX is required for platforms that use ROX as a passive reference. The table below outlines the hardware platforms that require the addition of ROX.

If ROX is required, dilute the ROX supplied according to the table below, then add 5µl to each of the Tetra™ MasterMix tubes.

|                      | Instruments  | Step 1: Volume of water to add to each ROX tube | Step 2: Add to MasterMix tube |
|----------------------|--|---|-------------------------------|
| High ROX Instruments | Applied Biosystems 7700, 7000, 7900, 7300, StepOne, StepOne Plus, and Roche capillary Lightcyclers 2.0 | No Dilution Required                            | 5 µl                          |
| Mid ROX Instruments  | Stratagene MX  | 75 µl   | 5 µl                          |
| Low ROX Instruments  | Applied Biosystems 7500 Platform, ViiA7 platforms, Quantstudio   | 130 µl  | 5 µl                          |
| All Other Machines   |  | Not Required                                    | Not Required                  |

## SUGGESTED USE CONDITIONS AND OPTIMISATIONS

### Denaturation

A denaturation step at 95°C is recommended. However, when using a template with high GC content, this may be increased to 98-100°C to improve results.

### Annealing

The recommended annealing temperature is between 60°C and 70°C. The annealing temperature may be optimised by performing a temperature gradient starting at 60°C then increasing in 2°C increment to remove non-specific product.

### Template Concentration

The recommended amount of template is 1-50 ng DNA per reaction, although the mix is extremely sensitive and capable of quantitation at pico-gram levels.

## qPCR BENCH SIDE PROTOCOL

Clean and decontaminate all work surfaces, pipettes, and other equipment prior to use to remove potentially contaminating nucleic acids.

### REACTION SET UP

Combine the following reagents to create a final test reaction:

| Component                | Volume |
|--------------------------|--------|
| Tetra™ 2X qPCR MasterMix | 10 µl  |
| Primer/probe mix         | 1 µl   |
| Extracted Sample DNA     | 2-9 µl |
| DNase/RNase Free Water   | x µl   |
| Final Volume             | 20 µl  |

## qPCR AMPLIFICATION PROTOCOL

Run the following PCR protocol:

|           | Temperature | Time       |
|-----------|-------------|------------|
| Hot Start | 95°C        | 3 minutes  |
| 45 cycles | 95°C        | 15 seconds |
|           | 60°C*       | 60 seconds |

## PRODUCT SPECIFICATIONS

### Storing your kit

On arrival the kit should be stored between -30°C and -15°C. Avoid prolonged exposure to light. If stored correctly the kit will retain full activity for 12 months. The kit can go through 10 freeze/thaw cycles with no loss of activity. We recommend storing the MasterMix in small aliquots if you plan to utilise it over more than 10 occasions.

### Regulatory status

This product has been developed for Research Use Only and is not intended for diagnostic use. It should not be used for diagnosis of disease unless specifically approved by the regulatory authorities in the country of use.

### Quality Control

In accordance with the YouSeq Ltd ISO EN 13485-certified Quality Management System, each lot of YouSeq Lyophilised Tetra™ 2X qPCR MasterMix is tested against predetermined specifications to ensure consistent product quality.

### Technical Assistance

For customer support, please contact:

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