# **Product Use Limitation & Warranty**

This product is intended to be used for life science research only. It has not been approved for drug or diagnostic purpose. YEASTERN's products should not be resold, modified for resole, or used to manufacture commercial products without written approval by YEASTERN. YEASTERN guarantees the performance of all products in the manner described in our protocol. The purchaser must determine the suitability of the product for its particular use. Should any product fail to perform satisfactorily due to any reason other than misuse, YEASTERN will replace it free of charge.

Copyright® 2011 All rights reserved. Yeastern Biotech Co., Ltd.

Copyright® 2011 All rights reser

ved. Yeastern Biotech Co., Ltd

YB



**Cat. No.** FYT105-100P FYT105-400P FYT106-100P FYT106-400P



No part of these protocols may be reproduced in any form or by any mean, transmitted, or translated into a machine language without the permission of YEASTERN BIOTECH CO., LTD.

> Address: 61-3, 23 Lane 169, Kang Ning St., Shijr, Taipei, 22180 Taiwan. Tel: +886-2-2695-3922 Fax: +886-2-2695-3979 Email: yeastern@yeastern.com.tw

# EZtime Real-Time PCR Premix (2X, For TaqMan® Probe) (2X, For TaqMan® Probe, ROX)

# Description

EZtime™ Real-Time PCR Premix with TaqMan<sup>®</sup> Probe is a ready-to-use, 2X concentrated premix reagent that includes Holstart Taq, TaqMan<sup>®</sup> Probe, ROX, optimized reation buffer and AIPFs for running real-time quantitive PCR(qPCR) and 2-step aRT-PCR. This premix can be used for detection of gene and quantification of gene expression with high sensitivity, good specificity, wide dynamic range and reproducibility.

Cat. No.	Product	Volume	Package
FYT105-100P	Eztime™ Real-Time PCR Premix	1.25 ml	100 rxns
FYT105-400P	(2X, For TaqMan® Probe)	5 ml	400 rxns
FYT106-100P	Eztime™ Real-Time PCR Premix	1.25 ml	100 rxns
FYT106-400P (2X, For TaqMan® Probe, ROX)	5 ml	400 rxns	

### Contents

- EZtime<sup>™</sup> Real-Time PCR Premix (2X, TaqMan<sup>®</sup> Probe, FYT106 with ROX)
- Protocol
- Hotstart Tag DNA Polymerase
- TaqMan<sup>®</sup> Probe Real-Time PCR Buffer
- dNTP mix including dATP, dCTP, dGTP, dTTP, 5 mM MgCl<sub>2</sub>

#### Storage

-20°C, avoid repeated freezing and thawing, protected from light.

### Procedure

## A. Preparation of PCR Master Mix

- 1. Thawing all reagents completely and vortex well.
- 2. Prepare a master mix according to Table 1

## Table 1. Reaction Components for real-time PCR master mixture

Component	Volume/ reaction	Final conc.
Template DNA	2 µl	n/a
Eztime™ Real-Time PCR Premix	12.5 µl	1X
Forward Primer (10µM)	0.75 µl	0.3~0.6 µM
Reversed Primer (10µM)	0.75 µl	0.3~0.6 µM
ddH <sub>2</sub> O	9 µl	
Total	25 µl	

3. Mix the master mix thoroughly by pipetting up and down. 4. Dispense 23 ul of master mix into PCR tubes or plates.

5. Add 2 µl of the DNA or cDNA; mix carefully by pipetting up and down.

# B. Performing Real-time PCR

1. Program your instrument according to **Table 2**. Users can choose either running **(A)** 2-step real-time PCR or a traditional **(B)** 3-step real-time PCR.

# Table 2. Thermal cycling conditions.







\* X: optimal annealing temperature is depending on user's primer sequences.

2. Place the PCR tubes or PCR plates in the thermal cycler and start the cycling program.

## Applications

- Quantitative real-time PCR
- Quantitative 2-step RT-PCR
- Quick and accurate detection and quantification of target gene through real-time PCR

# Note

For research use only. Not for use in diagnostic or the rapeutic procedures.

# **Product Use Limitation & Warranty**

This product is intended to be used for life science research only. It has not been approved for drug or diagnostic purpose. YEASTERN's products should not be resold, modified for resole, or used to manufacture commercial products without written approval by YEASTERN. YEASTERN guarantees the performance of all products in the manner described in our protocol. The purchaser must determine the suitability of the product for its particular use. Should any product fail to perform satisfactorily due to any reason other than misuse, YEASTERN will replace it free of charae.

No part of these protocols may be reproduced in any form

language without the permission of YEASTERN BIOTECH CO., LTD.

Email: yeastern@yeastern.com.tw

Copyright® 2011 All rights reserved. Yeastern Biotech Co., Ltd

Copyright® 2011 All rights reser ved. Yeastern Biotech Co., Ltd





**Cat. No.** FYT103-100P FYT103-400P FYT104-100P FYT104-400P

# EZtime Real-Time PCR Premix (2X, For SYBR® Green) (2X, For SYBR® Green, ROX)

### Description

EZTime<sup>™</sup> Real-Time PCR Premix with SYBR® Green is a ready-to-use, 2X concentrated premix reagent that includes Hotstart Taq, SYBR® Green I, ROX, optimized reaction buffer and AURPs for running real-time quantita -tive PCR (qPCR) and 2-step aRT-PCR. This premix can be used for detec -tion of gene and quantification of gene expression with high sensitivity, wide dynamic range and reproducibility.

Cat. No.	Product	Volume	Package
FYT103-100P	Eztime™ Rea⊦Time PCR Premix	1.25 ml	100 rxns
FYT103-400P	(2X, For SYBR <sup>®</sup> Green)	5 ml	400 rxns
FYT104-100P	Eztime™ Real-Time PCR Premix	1.25 ml	100 rxns
FYT104-400P	(2X, For SYBR® Green, ROX)	5 ml	400 rxns

#### Contents

- EZtime<sup>™</sup> Real-Time PCR Premix (2X, SYBR<sup>®</sup> Green, FYT104 with ROX)
- Protocol
- Hotstart Tag DNA Polymerase
- SYBR\* Green Real-Time PCR Buffer
- dNTP mix including dATP, dCTP, dGTP, dTTP, 5 mM MgCl<sub>2</sub>

#### Storage

-20°C, avoid repeated freezing and thawing, protected from light.

### Procedure

## A. Preparation of PCR Master Mix

- 1. Thawing all reagents completely and vortex well.
- 2. Prepare a master mix according to Table 1

## Table 1. Reaction Components for real-time PCR master mixture

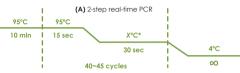
Component	Volume/ reaction	Final conc.
Template DNA	2 µl	n/a
Eztime™ Real-Time PCR Premix	12.5 µl	1X
Forward Primer (10 µM)	0.75 µl	0.3~0.6 µM
Reversed Primer (10 µM)	0.75 µl	0.3~0.6 µM
ddH <sub>2</sub> O	9 µl	
Total	25 µl	

Mix the master mix thoroughly by pipetting up and down.
Dispense 23 µl of master mix into PCR tubes or plates.
Add 2 µl of the DNA or cDNA. Mix carefully by pipetting up and down.

# B. Performing Real-time PCR

1. Program your instrument according to **Table 2**. Users can choose either running **(A)** 2-step real-time PCR or a traditional **(B)** 3-step real-time PCR.

# Table 2. Thermal cycling conditions.



\* 2-step program is used only for the length of target gene smaller than 300 bp.



\* X: optimal annealing temperature is depending on user's primer sequences.

- 2. Place the PCR tubes or PCR plates in the thermal cycle and start the cycling program.
- 3. Perform a melting curve analysis of the PCR product.

## Applications

- Quantitative real-time PCR
- Quantitative 2-step RT-PCR
- Quick and accurate detection and quantification of target gene through real-time PCR

## Note

For research use only. Not for use in diagnostic or the rapeutic procedures.