Fast SYBR[™] Green Master Mix

Catalog Numbers 4385610, 4385612, 4385614, 4385616, 4385617, and 4385618

Pub. No. 4385371 Rev. C

Note: For safety and biohazard guidelines, see the "Safety" appendix in the *Fast SYBR*^T *Green Master Mix User Guide* (Pub. No. 4385372). Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

This quick reference provides simplified procedures for using the Applied Biosystems^T Fast SYBR^T Green Master Mix. For details on performing real-time PCR, refer to the *Fast SYBR*^T Green Master Mix User Guide (Pub. No. 4385372).

Contents and storage

Cat. No.	Number of 20-µL reactions	Amount	Storage
4385610	100	1 × 1 mL	
4385612	500	1 × 5 mL	
4385616	1,000	2 × 5 mL	2-8°C
4385617	2,500	5 × 5 mL	2-6 C
4385618	5,000	10 × 5 mL	
4385614	5,000	1 × 50 mL	

Methods

1	Prepare the PCR reaction mix	a. b.	 Prior to use, mix the Fast SYBR[™] Green Master Mix thoroughly by swirling the bottle. Calculate the volume of each component needed for all the wells in each assay, based on the number of reactions. Note: We recommend performing four replicates of each reaction. 			
			Component	Volume for one reaction		
			Fast SYBR™ Green Master Mix (2X)	10 µL		
			Forward and reverse primers ^[1]	Variable		
			cDNA template + RNase-free water ^[2]	Variable		
			Total volume	20 µL		
		c.	 [1] For optimal performance, use a minimum of 200 nM of each primer. [2] For optimal performance, use up to 20 ng of cDNA per 20-µL reaction. Cap the tube(s). 			
		d.	Vortex the tube(s) briefly to mix the solutions.			
			 Centrifuge the tube(s) briefly to spin down the contents and eliminate any air bubbles from t solutions. 			
2	Prepare the PCR reaction plate	a.	Transfer the appropriate volume of reaction mixture to each	n well of a plate:		
			Plate type	Volume per well		
			MicroAmp [™] Fast Optical 48-Well Reaction Plate	20 µL		

b. Seal the plate with an optical adhesive cover, then centrifuge the plate briefly.

MicroAmp[™] Fast Optical 96-Well Reaction Plate

MicroAmp[™] Optical 384-Well Reaction Plate



20 µL

20 µL

3 Run the PCR reaction plate **a**. Set the thermal cycling conditions as specified in the following table:

Select Fast Mode			
Step	Temperature	Duration	Cycles
AmpliTaq [™] Fast DNA Polymerase, UP activation	95°C	20 seconds	HOLD
Denature	95°C	3 seconds	
Anneal/extend	60°C	30 seconds	40
AmpliTaq [™] Fast DNA Polymerase, UP activation	95°C	20 seconds	HOLD
Denature	95°C	1 seconds	40
Anneal/extend	60°C	20 seconds	40
	Step AmpliTaq [™] Fast DNA Polymerase, UP activation Denature Anneal/extend AmpliTaq [™] Fast DNA Polymerase, UP activation Denature	StepTemperatureAmpliTaq™ Fast DNA Polymerase, UP activation95°CDenature95°CAnneal/extend60°CAmpliTaq™ Fast DNA Polymerase, UP activation95°CDenature95°C	StepTemperatureDurationAmpliTaq [™] Fast DNA Polymerase, UP activation95°C20 secondsDenature95°C3 secondsAnneal/extend60°C30 secondsAmpliTaq [™] Fast DNA Polymerase, UP activation95°C20 secondsDenature95°C1 seconds

/.	Analyze the results	Data analysis varies depending on the instrument. Refer to the <i>Fast SYBR™ Green Master Mix User Guide</i>
4	-	(Pub. No. 4385372) and your instrument user guide for information.

Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale found on Life Technologies' website at www.thermofisher.com/us/en/home/global/terms-and-conditions.html. If you have any questions, please contact Life Technologies at www.thermofisher.com/support.

The information in this guide is subject to change without notice.

DISCLAIMER

TO THE EXTENT ALLOWED BY LAW, LIFE TECHNOLOGIES AND/OR ITS AFFILIATE(S) WILL NOT BE LIABLE FOR SPECIAL, INCIDENTAL, INDIRECT, PUNITIVE, MULTIPLE, OR CONSEQUENTIAL DAMAGES IN CONNECTION WITH OR ARISING FROM THIS DOCUMENT, INCLUDING YOUR USE OF IT.

Revision history: Pub. No. 4385371

Revision	Date	Description
С	4 May 2016	Added instruments to Step 3
		Format, style, and legal updates
В	September 2007	Baseline for this revision history

Corporate entity: Life Technologies Corporation | Carlsbad, CA 92008 USA | Toll Free in USA 1 800 955 6288

Important Licensing Information: These products may be covered by one or more Limited Use Label Licenses. By use of these products, you accept the terms and conditions of all applicable Limited Use Label Licenses.

©2016 Thermo Fisher Scientific Inc. All rights reserved. All trademarks are the property of Thermo Fisher Scientific and its subsidiaries unless otherwise specified. AmpliTaq is a trademark of Roche Molecular Systems, Inc.

