

Manual

2X HotStart qPCR MasterMix

Research use only.

This product is intended for Research Use Only. This product is manufactured under ISO 13485:2016 Quality System Requirements and is available for use as a Raw Material for use in IVD applications. Please contact LGC for further details. For SDS and Certificate of Analysis please contact techsupport@lgcgroup.com.

BIOSEARCH[™]
TECHNOLOGIES

Manual

2X HotStart qPCR MasterMix

1. Product description

HotStart qPCR MasterMix is a 2X ready-to-use master mix for quantitative, real-time amplification of dsDNA using intercalating dyes or fluorescent probe detection. HotStart qPCR MasterMix contains a chemically modified version of Taq DNA polymerase that is inactive at ambient temperatures but is rapidly activated upon heating to 95 °C during the initial 2-minute melting step of PCR. HotStart qPCR MasterMix includes dNTPs, MgCl₂, and an optimised buffer with additives that improve resistance to common environmental and biological inhibitors of PCR, making it amendable to direct sample testing applications. Although this product has been optimised for use with hydrolysis probe technology, it is also suitable for use with other probe-based detection systems.

2. Concentration

HotStart qPCR MasterMix: 2X

3. Storage and handling

Store at -20 °C upon arrival until provided expiration date. See individual component labels for additional storage recommendations. Minimise freeze/thaw cycles to avoid loss of performance.

4. Quick protocol

The following reaction set up and general cycling conditions are recommended but can vary depending on the template and primers being used. The following set up is for a 20 µl reaction size.

Component	Volume	Final
2X HotStart qPCR MasterMix	10 µl	1X
Target specific primers and probe	1 µl	1X
Sample	X µl	100 ng to 1 pg
EvaGreen® Dye, 20X in water (if required, purchase separately)	1 µl	1X
ROX Reference Dye, 25 µM (see table 2, if required, purchase separately)	See table 2	See table 2
Nuclease free water to volume	X µl	Not applicable
Total	20 µl	

Table 1: Reaction set-up for a 20 µL reaction.

Manual

2X HotStart qPCR MasterMix

Type	Company	Instrument	Final concentration
No ROX	Roche	LightCycler 480, LightCycler 2.0	None
	BioRad	iCycler, MyiQ, MiQ 2, iQ 5, CFX-96, CFX-384, Chromo4, MJ Opticon, Option2, MiniOpticon	
	Qiagen	Roto-Gene Q, Roto-Gene3000, Roto-Gene 6000	
	Illumina	Eco RealTime PCR System	
	Eppendorf	Mastercycler realplex	
	Cepheid	SmartCyler	
Low ROX	ABI	7500, 7500 Fast	30 nM
	Stratagene	MX4000P, MX3000P, MX3005P	
Hi ROX	ABI	5700, 7000, 7300, 7700, 7900, 7900HT, 7900HT Fast, StepOne, StepOne plus, Vii7	300 nM

Table 2: ROX concentration recommendation for different instruments.

5. Recommended use conditions

The following general cycling conditions are recommended but can vary depending on the template and primers being used.

Cycling Step	Temperature	Holding time	Cycles
Enzyme Activation	95 °C	2 minutes	1
Melting	95 °C	10 seconds	40
Amplification**	60 °C	60 seconds	

Table 3: Recommended cycling conditions.

** Temperature and holding time will be based off primer set used.

6. Ordering information

Item number	Size	Volume
300ITMM-1	200 reactions	1 mL × 2 tubes
300ITMM-2	600 reactions	1 mL × 6 tubes

7. Further support

If you require any further support, please do not hesitate to contact our Technical Support Team: techsupport@lgcgroup.com

Notice to purchaser:

EvaGreen® is a registered trademark of Biotium, Inc. Use of this product is covered by the following US patents and foreign equivalents: US7,803,943 B2, US7,776, 567, B2, including any patent application from which any of the listed patents directly or indirectly matured, any divisional, continuation and continuation-in-part application of any of the foregoing patent applications.

Research use only.

✕ **f in** @LGCBiosearch | biosearchtech.com

All trademarks and registered trademarks mentioned herein are the property of their respective owners. All other trademarks and registered trademarks are the property of LGC and its subsidiaries. Specifications, terms and pricing are subject to change. Not all products are available in all countries. Please consult your local sales representative for details. No part of this publication may be reproduced or transmitted in any form or by any means, electronic or mechanical, including photocopying, recording or any retrieval system, without the written permission of the copyright holder. © LGC Limited, 2025. All rights reserved. GEN/1209/SW/0225

BIOSEARCH™
TECHNOLOGIES