

qPCR Instrument Calibration Kit

Cat. No. 11742-100 Store at -20°C in the dark

Components: 8 tubes

Description

The qPCR Instrument Calibration Kit provides qualified reagents for calibrating your real-time qPCR system. The kit includes common dyes used in real-time qPCR (SYBR® Green I, ROX, TAMRA) and dyes for use with LUX™ Fluorogenic Primers (FAM, JOE, HEX, TET, and AlexaFluor® 546). Real-time qPCR instruments should be calibrated as part of regular instrument maintenance and prior to using new dyes for the first time when performing qPCR. Refer to your real-time qPCR instrument's user manual for specific instructions on calibration.

Components

The qPCR Instrument Calibration Kit contains one tube of each of 8 calibration dyes in amber tubes:

Dye	Volume	Concentration
FAM Calibration Dye	0.6 ml	10X
SYBR [®] Green Calibration Dye	0.6 ml	10X
JOE Calibration Dye	0.6 ml	10X
HEX Calibration Dye	0.6 ml	10X
TET Calibration Dye	0.6 ml	10X
AlexaFluor® 546 Calibration Dye	0.6 ml	10X
TAMRA Calibration Dye	0.6 ml	10X
ROX Calibration Dye	0.6 ml	10X

Instrument Compatibility

This calibration kit is for use with real-time qPCR instruments that have the option of being calibrated manually. The calibration kit is compatible with, but not limited to the following real-time qPCR instruments:

ABI PRISM® 7000

ABI PRISM® 7500

ABI PRISM® 7700

ABI PRISM® 7900 HT

Storage

Store 10X or diluted calibration dyes at -20° C in the dark. When thawing and using calibration dyes, protect from direct light. For most instruments, calibration is recommended every 3-6 months. Save the diluted dyes in the plates/tubes used to do the calibration at -20° C in the dark until next calibration. Calibration dyes may be reused for at least one year when stored correctly.

Caution

Wear powder-free gloves when handling the calibration dyes. Contact Technical Service to obtain the MSDS for further precautions on kit components.

Quality Control

Calibration dyes are tested first by measuring the emission peaks, and second by analyzing the raw spectra data on an ABI PRISM® 7700.

Part no. 11742.pps Rev. Date: 23 Feb 2005

This product is distributed for laboratory research use only. CAUTION: Not for diagnostic use. The safety and efficacy of this product in diagnostic or other clinical uses has not been established.

For technical questions about this product, call the Invitrogen Tech-LineSM 800 955 6288

Printed on Recycled Paper

General Protocol

The following protocol describes the steps involved in calibrating real-time qPCR instruments. For most systems, calibration should occur at initial setup, before the use of new dyes, and after maintenance service from the manufacturer. For more detailed instructions, refer to the manual for your particular instrument. If you are using an ABI PRISM® real-time qPCR system, refer to the next page for more details.

Important

If you have previously calibrated your real-time instrument for a dye included with this kit and wish to avoid overwriting the calibration setting, you should give the new calibration setting a different name (for example, "Invitrogen ROX").

Protocol

- 1. Thaw the 10X calibration dyes at room temperature away from direct light.
- 2. Dilute each 10X calibration dye 1:10 in 1X TE for a total volume of 5 ml in a new tube.
- 3. Aliquot the diluted calibration dyes into tubes or plates according to the suggested configuration and format for your real-time qPCR system. If you are using an ABI PRISM® instrument, see the guidelines on the next page. Return any unused 1X calibration dyes to -20°C in light-safe tubes.
- 4. Centrifuge the diluted calibration dyes in tubes or plates for 2 minutes at low speed to ensure that bubbles are removed.
- 5. Place diluted calibration tubes or plates into the sample tray of your real-time qPCR machine.
- 6. Run a pure dye spectral calibration on your real-time qPCR system using the protocol described in the instrument's user manual.
- Analyze the data after completing the calibration run using the protocol described in the instrument's user manual.
- 8. After processing, wrap the diluted calibration dyes in aluminum foil to protect from light and return to -20°C. If you are calibrating each dye with an individual plate, return to step 5 and repeat until all plates have been processed.

Guidelines for Calibrating ABI PRISM® Machines

If you are using a different real-time qPCR system than the models described below, refer to the system's instrument manual for calibration guidelines.

Loading Volume

Real-Time qPCR Instrument	Format	Volume/well		
ABI PRISM® 7000	96-well plate	50 μl/well		
ABI PRISM® 7500	96-well plate	50 μl/well		
ABI PRISM® 7700	96-well plate	50 μl/well		
ABI PRISM® 7900 HT	96-well plate	50 μl/well		
	384-well plate	20 μl/well		

Plate Configurations

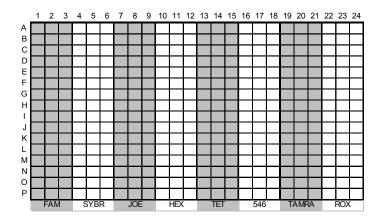
ABI PRISM® 7000 and 7500 Plate Configuration:

For the ABI PRISM $^{\circ}$ 7000 and 7500 models, each calibration dye needs to be aliquoted into every well of a 96-well plate, for a total of 8 separate calibration plates

ABI PRISM® 7700 and ABI PRISM® 7900 96-Well Plate Configuration:

_	1	2	3	4	5	6	7	8	9	10	11	12
Α					FAM	FAM	FAM	FAM				
В					SYBR	SYBR	SYBR	SYBR				
С					JOE	JOE	JOE	JOE				
D					HEX	HEX	HEX	HEX				
Ε					TET	TET	TET	TET				
F					546	546	546	546				
G					TAMRA	TAMRA	TAMRA	TAMRA				
Н					ROX	ROX	ROX	ROX				

ABI PRISM® 7900 HT 384-Well Plate Configuration:



Additional Products

Product	Amount	Catalog no.	
Custom Designed LUX [™] Fluorogenic Primers	www.invitrogen.com/lux		
Platinum [®] Quantitative PCR SuperMix UDG	100 rxns	11730-017	
SuperScript [™] III Platinum [®] Two-Step qRT-PCR Kit	100 rxns	11734-050	
SuperScript [™] III Platinum [®] One-Step qRT-PCR Kit	100 rxns	11732-020	

Purchaser Notification

Limited Use Label License No:154, SYBR Technology

The manufacture, use, sale or import of this product may be subject to one or more of US patent Nos. 5,436,134, 5,534,416, 5,545,535, 5,658,751, 5,863,753; 6,664,047; pending applications and corresponding foreign equivalents, owned by Invitrogen Corp. The purchase of this product conveys to the buyer the non-transferable right to use the purchased amount of the product and components of the product in research conducted by the buyer (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes. The buyer may transfer information or materials made through the use of this product to a scientific collaborator, provided that such transfer is not for any Commercial Purpose, and that such collaborator agrees in writing (a) to not transfer such materials to any third party, and (b) to use such transferred materials and/or information solely for research and not for Commercial Purposes. Commercial Purposes means any activity by a party for consideration and may include, but is not limited to: (1) use of the product or its components in manufacturing; (2) use of the product or its components to provide a service, information, or data; (3) use of the product or its components for therapeutic, diagnostic or prophylactic purposes; or (4) resale of the product or its components, whether or not such product or its components are resold for use in research. Invitrogen Corporation will not assert a claim against the buyer of infringement of the above patents based upon the manufacture, use or sale of a therapeutic, clinical diagnostic, vaccine or prophylactic product developed in research by the buyer in which this product or its components was employed, provided that neither this product nor any of its components was used in the manufacture of such product. If the purchaser is not willing to accept the limitations of this limited use statement, Invitrogen is willing to accept return of the product with a full refund. For information on purchasing a license to this product for purposes other than research, contact Molecular Probes, Inc., Business Development, 29851 Willow Creek Road, Eugene, OR 97402. Tel: (541) 465-8300. Fax: (541) 335-0504.

Limited Use Label License No:183, AlexaFluor® Dyes

The manufacture, use, sale or import of this product may be subject to one or more of U.S. Patent Nos. 5,696,157; 6,130,101; 6,716,979; pending applications and corresponding foreign equivalents, owned by Invitrogen Corp. The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and components of the product in research conducted by the buyer (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes. The buyer may transfer information or materials made through the use of this product to a scientific collaborator, provided that such transfer is not for any Commercial Purpose, and that such collaborator agrees in writing (a) to not transfer such materials to any third party, and (b) to use such transferred materials and/or information solely for research and not for Commercial Purposes. Commercial Purposes means any activity by a party for consideration and may include, but is not limited to: (1) use of the product or its components in manufacturing; (2) use of the product or its components to provide a service, information, or data; (3) use of the product or its components for therapeutic, diagnostic or prophylactic purposes; or (4) resale of the product or its components, whether or not such product or its components are resold for use in research. Invitrogen Corporation will not assert a claim against the buyer of infringement of the above patents based upon the manufacture, use or sale of a therapeutic, clinical diagnostic, vaccine or prophylactic product developed in research by the buyer in which this product or its components was employed, provided that neither this product nor any of its components was used in the manufacture of such product. If the purchaser is not willing to accept the limitations of this limited use statement, Invitrogen is willing to accept return of the product with a full refund. For information on purchasing a license to this product for purposes other than research, contact Molecular Probes, Inc., Business Development, 29851 Willow Creek Road, Eugene, OR 97402, Tel: 541)465-8300. Fax: (541)335-0354.

ABI PRISM® is a registered trademark of the Applera Corporation

©2005 Invitrogen Corporation. All rights reserved.

For research use only. Not intended for any animal or human therapeutic or diagnostic use.