



## QIAGEN Supplementary Protocol:

### LightCycler<sup>®</sup> 1.x Software Setup for the QuantiFast<sup>™</sup> SYBR<sup>®</sup> Green PCR Kit

This protocol shows the necessary parameters that need to be entered into the LightCycler 1.x software (version 3.5) when using the QuantiFast SYBR Green PCR Kit.

**IMPORTANT:** Please read the *QuantiFast SYBR Green PCR Handbook*, paying careful attention to the safety information, before beginning this procedure. The QuantiFast SYBR Green PCR Kit is intended for research use. No claim or representation is intended to provide information for the diagnosis, prevention, or treatment of a disease.

#### Procedure

1. Launch the software and set up the cycle program data as described in the next few steps.
2. Select "None" and set up the parameters for the initial activation step as shown below.

The screenshot shows the 'Cycle Program Data' window in the LightCycler 1.x software. The 'Analysis Mode' is set to 'None'. The 'Cycles' field is set to 1. The 'Temperature Targets' section is visible, with the following parameters set: Target Temperature (°C) at 95, Incubation Time (hrs:min:sec) at 5:00, Temperature Transition Rate (°C/s) at 20.00, Secondary Target Temperature (°C) at 0, Step Size (°C) at 0.0, Step Delay (cycles) at 0, and Acquisition Mode set to NONE. There are 'Ins' and 'Del' buttons for each parameter field.

3. Select "Quantification" and set up the parameters for PCR cycling as shown below. Be sure to select "SINGLE" for "Acquisition Mode" at the 60°C step.

Cycle Program Data

Analysis Mode: None  
Quantification  
Melting Curves

Cycles: 40

**Temperature Targets**

Target Temperature (°C)  
Incubation Time (hrs:min:sec)  
Temperature Transition Rate (°C/s)  
Secondary Target Temperature (°C)  
Step Size (°C)  
Step Delay (cycles)  
Acquisition Mode

Ins	95	10	20.00	0	0.0	0	NONE	Del
Ins	60	30	20.00	0	0.0	0	SINGLE	Del
Ins								

4. Select "Melting Curves" and set up the parameters for melting curve analysis as shown below. Be sure to select "CONT" for "Acquisition Mode" at the 95°C step.

Cycle Program Data

Analysis Mode: None  
Quantification  
Melting Curves

Cycles: 1

**Temperature Targets**

Target Temperature (°C)  
Incubation Time (hrs:min:sec)  
Temperature Transition Rate (°C/s)  
Secondary Target Temperature (°C)  
Step Size (°C)  
Step Delay (cycles)  
Acquisition Mode

Ins	95	15	20.00	0	0.0	0	NONE	Del
Ins	60	15	20.00	0	0.0	0	NONE	Del
Ins	95	0	0.10	0	0.0	0	CONT	Del
Ins								

5. Select "None" and set up the parameters for cooling as shown below.

Cycle Program Data

Analysis Mode: None

Cycles: 1

Quantification  
Melting Curves

**Temperature Targets**

Target Temperature (°C): 40

Incubation Time (hrs:min:sec): 30

Temperature Transition Rate (°C/s): 20.00

Secondary Target Temperature (°C): 0

Step Size (°C): 0.0

Step Delay (cycles): 0

Acquisition Mode: NONE

Ins Del

Ins

6. Load your PCR capillaries and start the program.

QIAGEN handbooks can be requested from QIAGEN Technical Service or your local QIAGEN distributor. Selected handbooks can be downloaded from [www.qiagen.com/literature/handbooks/default.aspx](http://www.qiagen.com/literature/handbooks/default.aspx).

Material safety data sheets (MSDS) for any QIAGEN product can be downloaded from [www.qiagen.com/ts/msds.asp](http://www.qiagen.com/ts/msds.asp).

Trademarks: QIAGEN®, QuantiFast™ (QIAGEN Group); LightCycler® (Roche Group); SYBR® (Molecular Probes, Inc.).

Purchase of the QuantiFast SYBR Green PCR Kit is accompanied by a limited, non-transferable immunity from suit to use it with detection by a dsDNA-binding dye as described in U.S. Patents Nos. 5,994,056 and 6,171,785 and corresponding patent claims outside the United States for the purchaser's own internal research. No real-time apparatus or system patent rights or any other patent rights, and no right to use this product for any other purpose are conveyed expressly, by implication or by estoppel.

The melting curve technology is covered by United States Patent No. 5,871,908, and corresponding foreign patents, owned by Evotec Biosystems GmbH and licensed to Roche Diagnostics GmbH. The purchase of this product does not convey to the buyer any right under these patent rights to perform the melting curve technology claimed in those patents. In particular, the purchase of this product does not include nor carry any right or license to use, develop, or otherwise exploit the melting curve technology commercially, and no rights are conveyed to the buyer to use the product or components of the product for any other purposes, including without limitation, provision of services to a third party, generation of commercial databases, research and development, human diagnostics or veterinary diagnostics. Roche Diagnostics GmbH reserves all rights under these patent rights. For information on purchasing a license to the patent rights for uses in conjunction with this product or to use the melting curve technology for other purposes, please contact Roche Diagnostics GmbH, Patent Department, Werk Penzberg, Nonnenwald 2, 82372 Penzberg, Germany.

PCR51 Apr-07 © 2007 QIAGEN, all rights reserved.



---

Sample & Assay Technologies