

# Data Sheet

## HOT FIREPol® Probe qPCR Mix Plus (ROX), 5x

Cat. No.	Pack Size	20 μl rxn
08-14-0000S	0.2 ml	50
08-14-00001	1 ml	250
08-14-00001-5	5 x 1 ml	1250
08-14-00001-10	10 x 1 ml	2500
08-14-00020	20 ml	5000

For in vitro use only

#### **Description:**

HOT FIREPol® Probe qPCR Mix Plus (ROX) is optimized for amplifying up to 2 targets in a single reaction in real-time quantitative PCR assays and contains all the components necessary to perform qPCR, with the exception of template, primers, and probe. The qPCR Mix contains optimized components and HOT FIREPol® DNA Polymerase supplied in a proprietary reaction buffer that enables detection of low copy number targets.

HOT FIREPol® Probe qPCR Mix Plus (ROX) is optimized for DNA hydrolysis probes based on the 5' flap endonuclease activity.

HOT FIREPol® DNA Polymerase is activated by a 12 min incubation step at 95°C. This prevents extension of non-specifically annealed primers and primer-dimers formed at low temperatures during qPCR setup.

## **Applications:**

- Detection and quantification of DNA and cDNA targets
- Profiling gene expression
- Microbial detection
- Viral load determination

## **Mix Composition:**

- HOT FIREPol<sup>®</sup> DNA Polymerase
- 5x Probe qPCR buffer
- 15 mM MgCl<sub>2</sub> 1x PCR solution – 3 mM MgCl<sub>2</sub>
- dNTPs
- Internal reference based on ROX dye

The dye is used to normalize the fluorescent reporter signal generated in qPCR. The product is compatible with both low ROX and high ROX system requirements.

## **Shipping and Storage conditions:**

Routine storage: -18°C to -28°C

Shipping and temporary storage for up to 1 month at room temperature has no detrimental effects on the quality of the product.

#### Recommendations:

Reaction setup at room temperature.

In order to prevent contamination, we recommend you to setup the reaction under laminar or in PCR box.

## Recommended qPCR reaction mix:

Component	Volume	Final conc.	
HOT FIREPol® Probe qPCR Mix Plus (5x)	4 µl	1x	
Forward primer (10 µM)	0.4–0.8 µl	200–400 nM	
Reverse primer (10 µM)	0.4–0.8 µl	200–400 nM	
Probe	x µl	100–250 nM	
DNA template	Variable	Variable <sup>1</sup>	
H <sub>2</sub> O PCR grade	up to 20 µl		
Total	20 µl		

<sup>&</sup>lt;sup>1</sup> Conc. of cDNA 0.1 pg/µl-10 ng/µl; gDNA 10 pg/µl-4 ng/µl

## Recommended qPCR cycling protocol:

Cycle step	Temp.	Time	Cycles
Initial activation <sup>2</sup>	95°C	12 min	1
Denaturation	95°C	15–20 s	10
Annealing/Extension <sup>3</sup>	60°C	60 s	40

<sup>&</sup>lt;sup>2</sup> To activate the polymerase, include an incubation step **at 95°C for 12 minutes** at the beginning of the qPCR cycle.

Manufactured by Solis BioDyne in compliance with the ISO 9001 and ISO 13485 certified Quality Management System.

FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES.

<sup>&</sup>lt;sup>3</sup> The annealing/extension temperature (Ta) depends on the melting temperature (Tm) of the primers. A Ta that is about 2 to 5°C lower than the Tm of the primers is generally suitable. Performing temperature gradient is recommended.

## Safety warnings and precautions:

This product and its components should be handled only by persons trained in laboratory techniques. It is advisable to wear suitable protective clothing, such as laboratory overalls, gloves and safety glasses. Care should be taken to avoid contact with skin or eyes. In case of contact with skin or eyes, wash immediately with water. Refer to Safety Data Sheet for more information.

### **Technical support:**

Contact your sales representative for any questions or send an email to support@solisbiodyne.com

Online chat is available at www.solisbiodyne.com

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