

Data Sheet

HOT FIREPol® Multiplex qPCR Mix (Purple), 5x

| Cat. No. | Pack Size | 20 µl rxn |
|----------------|-----------|-----------|
| 08-03-0000S | 0.2 ml | 50 |
| 08-03-00001 | 1 ml | 250 |
| 08-03-00001-5 | 5 x 1 ml | 1250 |
| 08-03-00001-10 | 10 x 1 ml | 2500 |
| 08-03-00020 | 20 ml | 5000 |

For *in vitro* use only

Description:

HOT FIREPol® Multiplex qPCR Mix (Purple) is optimized for amplifying up to 4 targets in a single reaction in real-time quantitative PCR assays. The qPCR Mix comprises all the components necessary (except primers, probes and template) to perform qPCR: HOT FIREPol® DNA Polymerase, optimized buffer components, ultrapure dNTPs and MgCl₂, Purple passive reference dye according to system requirements

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

HOT FIREPol® DNA Polymerase is activated by a 10 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® DNA Polymerase**
- **5x Multiplex qPCR buffer**
- **15 mM MgCl₂**
1x PCR solution – 3 mM MgCl₂
- **dNTPs**, including dUTP
The mix allows UNG treatment to prevent carryover contamination from previous runs. IMPORTANT: UNG is not included in the HOT FIREPol® Multiplex qPCR Mix and should be purchased separately.
- **Purple dye**
Purple dye is an internal passive reference dye used to normalize the fluorescent reporter signal generated in qPCR. If Cy5 or similar dye is used as one of the fluorophores, Purple passive reference dye might interfere with the signal. A version without passive reference or with ROX reference dye is available.

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.

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.

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® Multiplex qPCR Mix (Purple) (5x) | 4 µl | 1x |
| Forward primer (10 µM) | 0.4–0.8 µl | 200–400 nM (each) |
| Reverse primer (10 µM) | 0.4–0.8 µl | 200–400 nM (each) |
| Probe | x µl | 100–250 nM (each) |
| OPTIONAL: UNG (Uracil-N-glycosylase) | Variable | Variable ¹ |
| DNA template | Variable | Variable ² |
| H ₂ O PCR grade | up to 20 µl | |
| Total | 20 µl | |

¹ Please add UNG according to manufacturer's specification.

² 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 | Cycle s |
|---------------------------------------|-----------------------|-----------------------|---------|
| OPTIONAL: UNG treatment ³ | Variable ³ | Variable ³ | 1 |
| Initial activation³ | 95°C | 10 min | 1 |
| Denaturation | 95°C | 15–20 s | 40 |
| Annealing/Extension ⁴ | 60°C | 60 s | |

³ **OPTIONAL!** Add UNG treatment step **ONLY** if UNG enzyme is added in the reaction mix for carryover contamination removal. Use UNG according to manufacturer's specification.

³ To activate the polymerase, include an incubation step **at 95°C for 10 minutes** at the beginning of the qPCR cycle.

⁴ The annealing temperature (T_a) depends on the melting temperature (T_m) of the primers. A T_a that is about 2 to 5°C lower than the T_m of the primers is generally suitable. Performing temperature gradient is recommended.

Cycler compatibility:

HOT FIREPoI® Multiplex qPCR Mix (Purple) is compatible with qPCR cyclers, such as Applied Biosystems™ 7500 Fast Real-Time PCR System, 7500 Real-Time PCR System, ViiA™ 7 PCR System, and QuantStudio™ 6 Flex, 7 Flex, and 12K Flex Real-Time PCR systems, which enable using Mustang Purple™ channel for reference signal normalization.

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

DS-08-03 v3
Revised 12.04.2022

Permitted Use: This product is supplied for research use only (the **Permitted Use**). If the customer wishes to use the product for any purpose other than the Permitted Use, including (without limitation) resale or alteration, the customer should obtain the appropriate licence from Solis BioDyne. Some applications of this product may require a license/licenses from one or more third parties which are not provided by the purchase of this product. Users should obtain the licence if required. Covered by the patent EP2501716, made by the methods of US Patent No 9,321,999.

Trademark information: FIREPoI® is an EU registered trademark of Solis BioDyne OÜ. Applied Biosystems is a trademark of Applied Biosystems LLC, QuantStudio, ViiA and Mustang Purple are trademarks of Life Technologies Corporation.

Warranty and Disclaimer: This product shall comply with its relevant specification and be fit for its stated purpose, but Solis BioDyne gives no other warranty and makes no representation as to description or quality. Any such warranty or representation is excluded, to the fullest extent permitted by law. In particular, but without limiting the foregoing, Solis BioDyne shall not be liable for the failure of the product to comply with its relevant specification where such failure arises as a result of: (i) customer negligence or because the customer failed to follow any of the applicable technical data or safety sheets, standard user materials, use guidelines or any other information provided by Solis BioDyne as to the storage, transportation, handling, use or maintenance of the products or other good practice regarding the same, or (ii) the customer altering the products in any way without the prior written consent from Solis BioDyne, or (iii) the products differing from the relevant specification as a result of changes made to ensure their compliance with applicable statutory or regulatory requirements.

Nothing shall limit or exclude Solis BioDyne's liability for death or personal injury caused by its negligence, fraud or fraudulent misrepresentation or any matter in respect of which it would be unlawful for Solis BioDyne to exclude or restrict liability. Without limiting the foregoing, Solis BioDyne shall under no circumstances whatever be liable to the customer, whether in contract, tort (including negligence), breach of statutory duty, or otherwise, for any loss of profit, or any indirect or consequential loss arising under or in connection with the products and Solis BioDyne's total liability to the customer in respect of all other losses arising under or in connection with the product, whether in contract, tort (including negligence), breach of statutory duty, or otherwise, shall in no circumstances exceed the price of the products supplied in respect of which the liability has arisen.

Solis BioDyne

Teaduspargi 9, 50411 Tartu, Estonia, tel: +372 740 9960, fax: +372 740 2079, e-mail: info@solisbiodyne.com, www.solisbiodyne.com