

# RNaseP ENDOGENOUS CONTROL qPCR KIT USER GUIDE

Catalogue No's: YSL-qP-ENDO-CY5 / YSL-qP-ENDO-HEX 100 reactions

**VERSION 1.0** 

For Research Use Only



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### INTENDED USE

This kit is designed to be used as a Human endogenous control for a qPCR reaction. The endogenous control is detected in the Yellow (VIC/HEX) or Blue (Cy5) channel of your qPCR instrument in multiplex with your target of interest. The kit is deliberately primer limited to avoid any risk on inhibiting detection of your target of interest. It is designed to be used by trained users in a suitable molecular biology laboratory environment.

### KIT CONTENTS

|  | Cap Colour | Volume |  |
|--|------------|--------|--|
| Endogenous control primer/probe (Cy5 or VIC/HEX probe) |            | 11Ο μΙ |  |

### MATERIALS REQUIRED BUT NOT PROVIDED

Extraction Kit - This qPCR test kit will work well with high quality DNA/RNA derived from any extraction kit with minimal PCR inhibitors present.

Pipettes, micro-centrifuge tubes and general laboratory equipment.

# qPCR BENCH SIDE PROTOCOL

Clean and decontaminate all work surfaces, pipettes, and other equipment prior to use to remove potentially contaminating nucleic acids.

### REACTION SET UP

Combine the following reagents to create a final test reaction:

| Component                       | Volume |
|---------------------------------|--------|
| 2X qPCR MasterMix               | 10 µl  |
| Target primer/probe             | × μΙ   |
| Endogenous Control primer/probe | 1 μΙ   |
| Extracted Sample DNA/RNA        | х µl   |
| Final Volume                    | 20 μΙ  |



## qPCR AMPLIFICATION PROTOCOL

### RNA SAMPLES:

Run the following PCR protocol:

Please note: This YouSeg kit will work with any gPCR instrument capable fluorescence of either Cy5 or HEX (VIC).

|           | Temperature | Time       |
|-----------|-------------|------------|
| RT Step   | 55°C        | 10 minutes |
| Hot Start | 95°C        | 3 minutes  |
| 45 cycles | 95°C        | 15 seconds |
|           | 60°C*       | 60 seconds |

<sup>\*</sup>Make sure to collect fluorogenic data through all the required target channels.

### **DNA SAMPLES:**

Run the following PCR protocol:

Please note: This YouSeq kit will work with any qPCR instrument capable fluorescence of either Cy5 or HEX (VIC).

|           | Temperature | Time       |
|-----------|-------------|------------|
| Hot Start | 95°C        | 3 minutes  |
| 45 cycles | 95°C        | 15 seconds |
|           | 60°C*       | 60 seconds |

<sup>\*</sup>Make sure to collect fluorogenic data through all the required target channels.

### INTERPRETATION OF RESULTS

Detection of the endogenous control is through the Cy5 or HEX channel depending on the kit version you have chosen. Cq values in the range 28±3 indicate a good quality extraction has taken place. If your Cq value is later than 28, this indicates that only a small amount of DNA/RNA was present in your sample. You may wish to repeat sample collection and then repeat the test in order to confirm the result.



### PRODUCT SPECIFICATIONS

### Storing your kit

Store at -20C from arrival. The kit's shelf life is outlined as an expiry date on the pouch label.

#### Use good quality DNA/RNA

Poor quality input nucleic acid is the biggest cause of test failure. The kit will work well with any source of good quality DNA/RNA. Good quality is defined as DNA/RNA with high integrity (not degraded) and with low levels of inhibitors present.

#### Regulatory status

This product has been developed for Research Use Only and is not intended for diagnostic use. It should not be used for diagnosis of disease unless specifically approved by the regulatory authorities in the country of use.

#### **Quality Control**

In accordance with the YouSeq Ltd ISO EN 13485-certified Quality Management System, each lot of this kit is tested against predetermined specifications to ensure consistent product quality.

#### **Technical Assistance**

For customer support, please contact:

e-mail: support@youseq.com phone: +44 (0)333 577 6697

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