3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone 800-325-5832 • (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

**Control Primer Set for Arabidopsis Actin-2 Gene**

**ProductInformation**

Product Number **C 3615**
Storage Temperature -20 °C

# TECHNICAL BULLETIN

## Product Description

This product consists of a primer set for the Arabidopsis housekeeping gene Actin-2 (At3g18780). It is intended for use as an internal control for Q-RT-PCR for expression analysis of any Arabidopsis gene.

Of our three control primer series for Arabidopsis gene expression, Actin-2 control primer is designed as a control for high-abundant gene expression. The other two control primers in the series are nuclear cap-binding protein (CBP20, Product Code C 3115) and ubiquitin (UBC, Product Code C 3240) designed as controls for medium- to low-abundant gene expression.

## Components

Each tube contains enough primer (forward or reverse) for 100 RT-PCR reactions (50 µl reaction volume)

- Forward Primer ($T_m$ = 68 °C based on % GC method in 1 M salt concentration), 20 µM, in TE buffer, Product Code A 7728.
- Reverse Primer ($T_m$ = 67 °C based on % GC method in 1 M salt concentration), 20 µM, in TE buffer, Product Code A 6978.

## Precautions and Disclaimer

This product is for R&D use only. Not for drug, household or other uses. Consult the MSDS for information regarding hazards and safe handling techniques.

## Storage/Stability

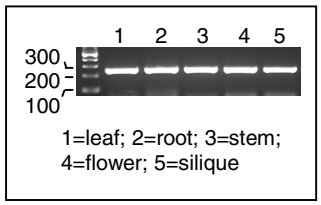
Primer should be stored at –20 °C. Primers are stable at –20 °C for at least 1 year and can be frozen and thawed at least 10 times.

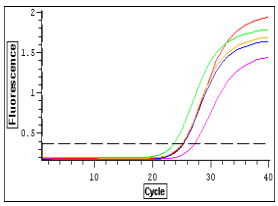
## Recommended PCR Procedure

| Step | Temperature | Time |
|---|---|---|
| For Cycles 1-30: | | |
| Denaturation | 94 °C | 15 sec. |
| Annealing | 65 °C | 45 sec. |
| Extension | 72 °C | 1 min. |
| Plate Read (real-time) | 80 °C | 1 sec. |
| Incubate | 72 °C | 10 min. |
| | 4 °C | Indefinite |

## Product Profile

The primers have been tested by quantitative RT-PCR with total RNA isolated from five different Arabidopsis tissues, leaf, root, stem, flower, and silique. The size of the amplicon produced by this set of primers is 257 bp. The following figure shows performance of the primer set on different Arabidopsis tissues. Total RNA was isolated from each tissue using GenElute Total RNA Kit (RTN10). Samples were treated with amplification grade DNAse I, Product Code AMP-D1, to remove genomic DNA. Equal amounts of total RNA, based on OD measurements at 260 nm, were added in each RT-PCR reaction. The gel-based RT-PCR was conducted using the JumpStart RED HT RT-PCR kit (Product Code J 3520), whereas the real time RT-PCR data was generated using the SYBR Green Quantitative RT-PCR kit, Product Code QR0100.



**References**

1. Lovatt, A., et al., Validation of quantitative PCR assays. BioPham, March 2002, p. 22-32.
2. Bustin, S. A., Quantification of mRNA using real-time reverse transcription PCR (RT-PCR): trends and problems. J. Mol. Endocrinol., **29,** 23-29 (2002).
3. Ginzinger, D. G., Gene quantification using real-time quantitative PCR: an emerging technology hits the mainstream. Exp. Hematol., **30,** 503-512 (2002).

**Related Products**

GenElute Total RNA Kit, Product Codes RTN10, RTN70, and RTN350
GenElute Total RNA Purification Kit, Product Codes NA6000 & NA0610
JumpStart RED HT RT-PCR kit, Product Code J 3520
SYBR Green Quantitative RT-PCR kit, Product Code QR0100

JWM 4/04