foodproof®

GMO A2704-12 Soya Quantification Kit
Ready Reference Guide

Revision A, December 2023

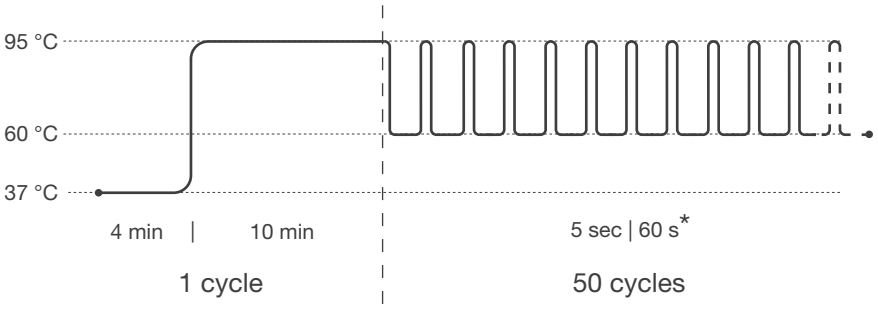
Product No. KIT230019

Before starting, it is strongly recommended to read the entire product instructions available on our website.

PROGRAM SETUP

Program your real-time PCR instrument before setting up the PCR reactions. Select the following channels:

- FAM: A2704-12 and Soya (*Le1)*



Pre-incubation: 1 cycle
Step 1: 37 °C for 4 min
Step 2: 95 °C for 10 min

Amplification: 50 cycles
Step 1 : 95 °C for 5 s
Step 2*: 60 °C for 60 s

* Fluorescence detection

For some real-time PCR instruments the probe quencher as well as the usage of a passive reference dye has to be specified. This kit contains probes with a non-fluorescent "dark" quencher and no passive reference dye.

PREPARATION OF STANDARD CURVE

Use Calibrator DNA (purple cap) and Dilution Buffer (blue cap) to prepare dilutions according to table below. For each dilution step, pipet 30 µL (60 µL for duplicates) Dilution Buffer into a new reaction tube. Transfer 10 µL (20 µL for duplicates) from preceding step to new dilution step. Mix well between pipetting steps.

The prepared dilutions can be used for both standard curves, GMO gene and reference gene.

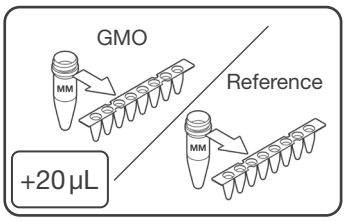
A typical experiment consists of 16 wells needed for standards and controls, plus 2 × n wells (n = number of food samples). Since a multiwell plate has 96 wells, 40 food samples can be analyzed during one PCR run if the GMO gene and the reference gene are analyzed in the same run. Some real-time PCR instruments provide the opportunity to import external standard curves generated in a previous run, then 46 food samples can be analyzed during one PCR run.

Dilution Step	Dilution Factor	Final Concentration
1	Undiluted	100
2	1:4	25
3	1:16	6.25
4	1:64	1.56
5	1:256	0.39
6	1:1024	0.098

RR-KIT230019-RevA

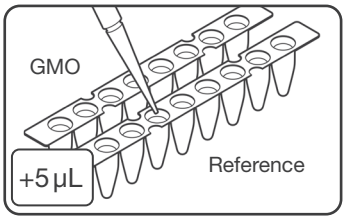
PREPARATION OF THE PCR MIX

Take appropriate precautions to prevent contamination, e.g., by using filter tips and wearing gloves.
Thaw reagents, mix (do not vortex!), and briefly spin vials before opening.
For data interpretation and calculation please refer to the entire product manual.



1. ADD PCR MIX

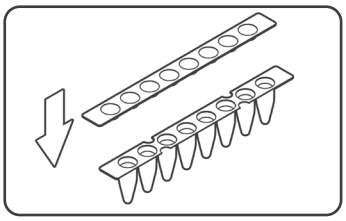
For each master mix - the GMO Gene (yellow cap) and the Reference Gene (green cap) - pipet 20 µL into different strip or plate wells for the number of samples, standards and negative control.



2. ADD SAMPLES AND CONTROLS

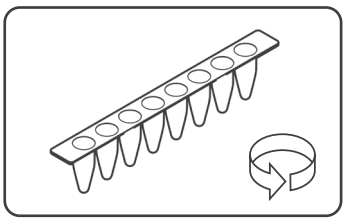
Sample DNA must be diluted at least 1:4 in the Dilution Buffer (blue cap).

To each PCR mix (GMO and Reference), pipet 5 µL of samples, standards, negative control (colorless cap) or Control Template (purple cap) into respective wells.



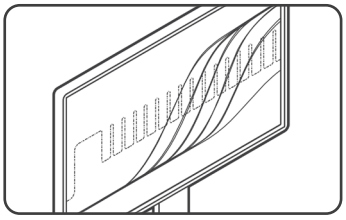
3. SEAL

Seal strips/plate accurately.



4. CENTRIFUGE

Briefly spin strips/plate in a suitable centrifuge.



5. START REAL-TIME PCR RUN

Cycle samples as described above.

foodproof®
GMO A2704-12 Soya
Quantification Kit

KIT230019
Kit for 2 x 64 reactions
Store kit at -15 to -25 °C

For food testing purposes
FOR IN VITRO USE ONLY
Made in Germany

hygiena®

Hygiena® | Camarillo, CA 93012 USA | diagnostics.support@hygiena.com | www.hygiena.com