foodproof®

Listeria Genus Detection LyoKit
Ready Reference Guide

Revision A, November 2023

Product No. KIT230088 (LP), KIT230089 (RP), KIT230330 (DP)

PCR Kit for the qualitative detection of the *Listeria* sensu stricto species (*L. monocytogenes*, *L. seeligeri, L. ivanovii, L. welshimeri, L. innocua* and *L. marthii*).

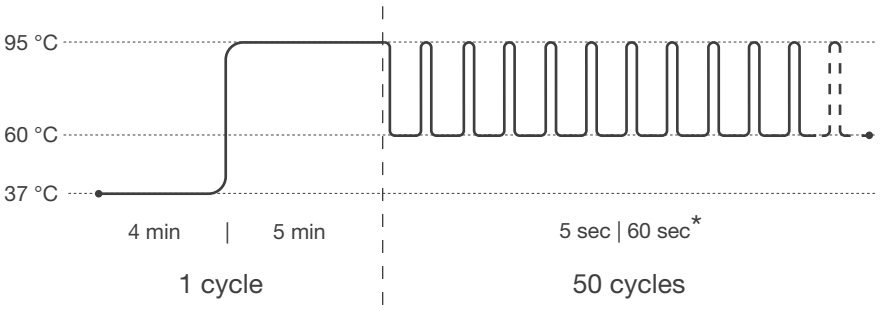
Before starting, it is strongly recommended to read the entire product manual available on our website.

PROGRAM SETUP

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

- FAM (*Listeria* sensu stricto) and VIC (Internal Control).

As an alternative to VIC, HEX can be used. For the PikoReal® 24, Yakima Yellow has to be selected.



Pre-incubation: 1 cycle
Step 1: 37 °C for 4 min
Step 2: 95 °C for 5 min
Amplification: 50 cycles
Step 1 : 95 °C for 5 sec
Step 2*: 60 °C for 60 sec

* 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.

For the Dualo 32 R2 real-time PCR instrument, please open the software, click on 'New', and select the respective template file. Template files can be added by clicking on 'Add' in the 'Select template file' window.

DATA INTERPRETATION

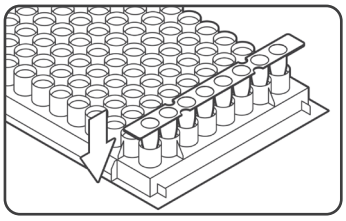
Verify results of positive (Control Template) and negative controls (H_2O), before interpreting sample results. Always compare samples to positive and negative control. Review data from each channel and interpret results as described in the table.

FAM	VIC	Result Interpretation
+	+ or -	Positive for *Listeria* sensu stricto
-	+	Negative for *Listeria* sensu stricto
-	-	Invalid

RR-KIT230088-089-330-RevA

PREPARATION OF THE PCR MIX

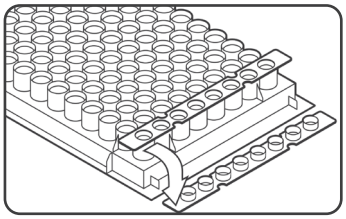
Take appropriate precautions to prevent contamination, e.g., by using filter tips and wearing gloves.



1. PLACE STRIPS IN RACK

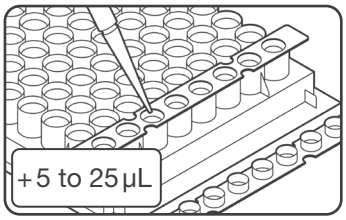
Take needed number of PCR tube strips out of aluminum bag.
Important: close bag tightly afterwards. Place strips in a suitable PCR tube rack.

If needed, gently tap the tubes to move the lyophilized pellets to the bottom of all tubes.



2. DECAP

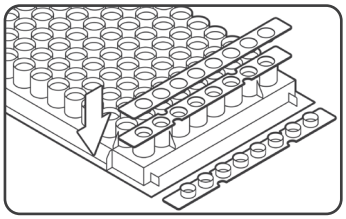
Carefully open strips immediately before filling and discard caps.
Do not leave open longer than necessary.



3. ADD SAMPLES AND CONTROLS

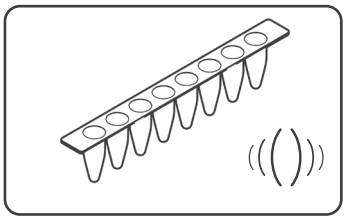
Pipette 5 to 25 µL of samples, 25 µL negative control (colorless cap) or Control Template (purple cap) into respective wells.

Varying sample volumes are due to different enrichment broths and DNA extraction procedures. If using less volume, add PCR-grade H_2O to reach a total volume of 25 µL.



4. SEAL

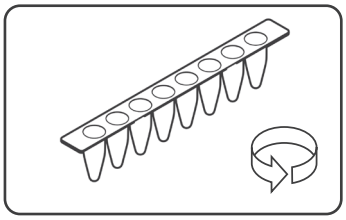
Carefully seal the tubes with the provided 8-cap strips.



5. MIX

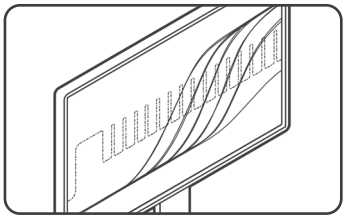
Resuspend pellet after sealing by mixing thoroughly.

Alternatively, resuspend pellet by pipetting up and down multiple times in Step 3.



6. CENTRIFUGE

Briefly spin strips, e.g., 5 seconds at 500 - 1,000 x g, in a suitable centrifuge.



7. START REAL-TIME PCR RUN

Cycle samples as described above.

Place tubes in a vertical, balanced order into the cycler, e.g., two strips can be placed in the first and last column.

foodproof®
Listeria Genus
Detection LyoKit

KIT230088 /089 /330
Kit for 96 reactions
Store kit at 2 to 8 °C

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

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