foodproof®

Staphylococcus Detection LyoKit
Ready Reference Guide

Revision B, June 2025

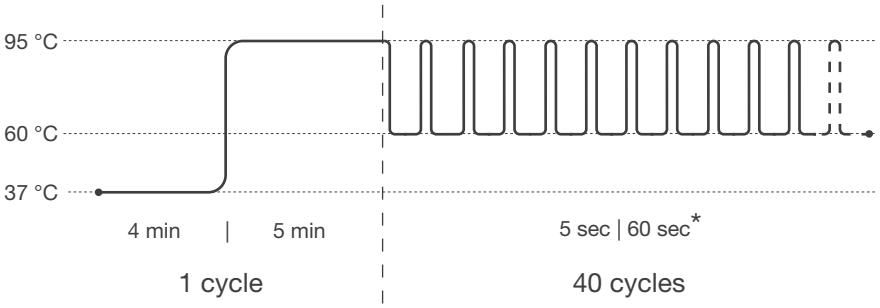
Product No.: KIT230103 (LP), KIT230104 (RP)

PCR kit for the qualitative detection of the most important coagulase-positive *Staphylococcus* species including the simultaneous identification of *Staphylococcus aureus*.

PROGRAM SETUP

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

- FAM (coagulase-positive *Staphylococcus* spp.), HEX (*Staphylococcus aureus*) and ROX (Internal Control).



Pre-incubation: 1 cycle
Step 1: 37 °C for 4 min
Step 2: 95 °C for 5 min
Amplification: 40 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. A Color Compensation is necessary for users of the LightCycler® 480 System: Color Compensation Set 5 (Product No.: KIT230011).

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.

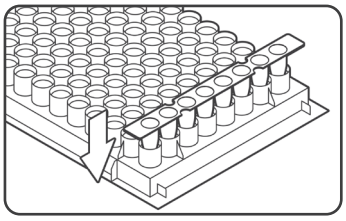
FAM	HEX	ROX	Result Interpretation
+	+	+ or -	Positive for *S. aureus*
+	-	+ or -	Positive for coagulase-positive *Staphylococcus* spp. other than *S. aureus*
-	-	+	Negative for coagulase-positive *Staphylococcus* spp.
-	-	-	Invalid

If the Cq-value for FAM or HEX is > 35, the signal may have been caused by DNA from dead bacteria.
In this case, a prolongation of enrichment and a new analysis is recommended.

RR-KIT230103-04-REVB

PREPARATION OF THE PCR MIX

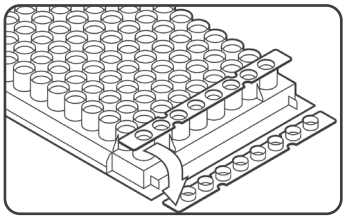
Take appropriate precautions to prevent contamination, e.g., by using filter tips and wearing gloves.



1. PLACE STRIPS IN RACK

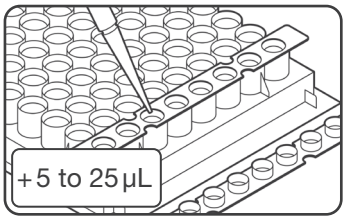
Take needed number of PCR tube strips out of aluminum bag.
Important: close bag tightly afterwards. Place strips in a suitable PCR tube rack.

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



2. DECAP

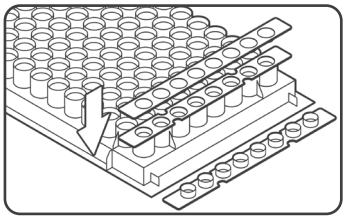
Carefully open strips immediately before filling and discard caps.
Do not leave open longer than necessary.



3. ADD SAMPLES AND CONTROLS

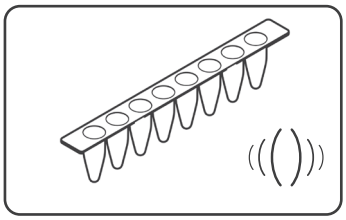
Pipette 5 to 25 µL of samples, 25 µL negative control (colorless cap) or Control Template (purple cap) into respective wells.

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

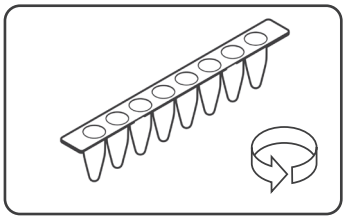
Carefully seal the tubes with the provided 8-cap strips.



5. MIX

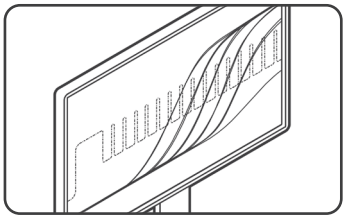
Resuspend pellet after sealing by mixing thoroughly.

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®
Staphylococcus
Detection LyoKit

KIT230103 /104
Kit for 96 reactions
Store kit at 2 to 8 °C

For food testing purposes
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Made in Germany

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