RAPID’S Staph Agar

356-3960
356-4704

AREA OF APPLICATION
Medium used for the enumeration of coagulase-positive Staphylococcus aureus at 37°C in 24h from food.

PRINCIPLE
RAPID’S Staph medium is based on a Baird Parker formula optimized for the detection and enumeration of Staphylococcus aureus in 24h (± 2h). The principle of the medium relies on the capacity of Staphylococcus aureus to reduce tellurite (black colonies) and to provoke proteolysis of egg yolk (clear halo around colonies).

AFNOR VALIDATION
RAPID’S Staph has AFNOR approval as a valid alternative method to the NF ISO 6888-1 standard for the enumeration of coagulase-positive staphylococci (Staphylococcus aureus and other species) for all food products destined for human consumption, according to ISO 16140 protocol, under Attestation No: BRD - 07/09-02/05.

AOAC VALIDATION
RAPID’S Staph has been validated by the AOAC Research Institute under the Performance Tested Method Program for enumeration of coagulase-positive Staphylococcus aureus in pasteurized whole milk, custard pie, processed ham and smoked salmon. Typical colonies on RAPID’S Staph are presumptive and should be confirmed by standard reference methods appropriate for the food type being tested.

STANDARD REFERENCES
FDA Bacteriological Analytical Manual, Chapter 12 Staphylococcus aureus.

PRESENTATION
Pre-poured plates
- 90mm x 20 356-3960
Dehydrated
- 500g 356-4704

STORAGE / VALIDITY / BATCH
- Pre-poured and supplements: + 2 - 8 °C in a dark place.
- Dehydrated: 15-25°C, in tightly closed bottle in a cool dry place.
- Petri dishes prepared by user: 5 days, + 2 - 8°C in a dark place.
- Expiration date and batch number are indicated on the pack.

COMPLETE FORMULA
Proprietary peptone mixture 10 g
Yeast extract 1 g
Meat extract 5 g
Lithium chloride 5 g
Agar 16 g
L-Glycine 12 g
Sodium pyruvate 10 g
Potassium tellurite 0.1 g
Egg yolk emulsion 10 ml
Sulfamethazine 0.05 g
Distilled water 1000 ml

Final pH = 7.2 ± 0.2

OTHER PRODUCT(S) REQUIRED (NOT SUPPLIED)
Butterfield’s Phosphate Buffer
Egg yolk with potassium tellurite
- 5ml x 1 vial 355-4201
- 25ml x 1 flask 355-4205
Sulfamethazine 0.2 %
- 2.5ml x 1 vial 356-2682

OTHER EQUIPMENT REQUIRED (NOT SUPPLIED)
- Autoclave
- Balance
- Blender and weigh bags
- Bottles, 125ml, autoclavable
- Hot plate with stirring
- Sterile Petri dishes (Ø = 90 or 140 mm)
- Sterile pipettes (0.1 ml, 1 ml, etc.)
- Sterile spreader
- Stomacher bags
- Thermostatically controlled incubator capable of maintaining 37 ± 1 °C
- Water bath accurate to ± 1 °C
PREPARATION OF THE DEHYDRATED MEDIUM

Always shake before use
Dissolve 57g of powder in 1 liter of distilled water. Wait 5 minutes, then mix until a homogenous suspension is obtained. Heat gently, agitating frequently, then bring to a boil until completely dissolved. Dispense 90ml of medium per bottle. Sterilize in autoclave at 121°C (± 1°C) for 15 minutes.

At moment of use, add the following solutions to 90ml of base, previously melted and cooled to between 44°C and 47°C:
- 5 ml egg yolk with potassium tellurite
- 2.5 ml of 0.2% sulfamethazine
Mix thoroughly. Pour into Petri dishes and leave to solidify on a level surface.

500g of powder makes 8.7 liters of medium

PROTOCOL

Preparation of samples
Weigh 50g of food (or measure 50ml of liquid) and add 450ml Butterfield’s phosphate buffer. Homogenize in a blender for 2 minutes on high speed (16,000 – 18,000 rpm).

Inoculation and incubation
For samples with >150 S. aureus/g, spread 0.1ml of the sample to be analyzed and/or 0.1ml of its dilutions over the surface of a dried plates.

For samples with <150 S. aureus/g, spread 1ml of the sample to be analyzed and/or 1ml of its dilutions over the surface of 3 dried plates (0.4ml, 0.3ml, 0.3ml).

Turn the dishes over and incubate at 37°C (± 1°C) for 24h (± 2h)

READING AND INTERPRETATION

After each period of incubation, proceed with counting of typical colonies. Coagulase-positive Staphylococcus aureus form black colonies on this opaque medium with a clear halo around the colony, corresponding to a zone of proteolysis (lightening of egg yolk). Presumptive colonies must be confirmed by coagulase test according to standard reference methods.

Count only those plates with 15-150 colonies.

To obtain total S. aureus/g count, multiply the number of typical colonies by the percentage of colonies that confirmed (coagulase test). Multiply this by the appropriate dilution factor.

PRECAUTIONS

- Respect Good Laboratory Practice. Appropriate protection, such as gloves and lab coat, should be worn when working with live organisms.

- Media that have come in contact with food samples should be considered contaminated and should be autoclaved prior to disposal.

- Before using RAPID Staph, allow plates to dry, according to standard ISO 7218 at 25°C-50°C, until all drops on the surface of the media have disappeared. Avoid prolonged drying, however, since this could affect the performance of the medium.

- Do not add supplements to base medium at temperatures exceeding 47°C.

TECHNICAL SUPPORT

In the United States, for technical assistance please call (800) 4BIORAD. Select option 2 for technical support and option 2 again for the food science division. To place an order, please call (800) 4BIORAD and press option 1 for customer service.

QUALITY CONTROL

All products manufactured and marketed by Bio-Rad are incorporated into a quality assurance procedure from reception of raw materials through commercialization of the end product. Each batch of end product undergoes quality control, and is only marketed if it complies with acceptance criteria. Documentation concerning production and verification of each batch is archived. Material Safety Data Sheets are available on the web at www.foodscience.bio-rad.com
### TEST QUALITY AND PERFORMANCE

<table>
<thead>
<tr>
<th>Strains</th>
<th>Results after 24H at 37°C</th>
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</thead>
<tbody>
<tr>
<td><strong>Tellurite reduction</strong></td>
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</tr>
<tr>
<td><strong>Halo</strong></td>
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<tr>
<td><strong>Growth</strong></td>
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<tr>
<td><strong>Staphylococcus aureus</strong></td>
<td></td>
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<tr>
<td>ATCC 6538</td>
<td>Tellurite reduction: Positive, Black colonies</td>
</tr>
<tr>
<td><strong>Staphylococcus aureus</strong></td>
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</tr>
<tr>
<td>ATCC 25923</td>
<td>Tellurite reduction: Positive, Black colonies</td>
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<tr>
<td><strong>Staphylococcus epidermidis</strong></td>
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<tr>
<td>ATCC 12228</td>
<td>Tellurite reduction: Gray/black colonies</td>
</tr>
<tr>
<td><strong>Escherichia coli</strong></td>
<td></td>
</tr>
<tr>
<td>ATCC 25922</td>
<td>No growth</td>
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</tbody>
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