
BAIRD-PARKER AGAR WITH EGG YOLK TELLURITE

ENUMERATION OF COAGULASE POSITIVE STAPHYLOCOCCI

1 INTENDED USE

Baird-Parker Agar with egg yolk and potassium tellurite is a selective medium for the detection and enumeration of coagulase-positive staphylococci (*Staphylococcus aureus* and other species) in animal origin biological samples, pharmaceutical products, cosmetics, foods and water.

The typical composition is that defined in the food microbiology directives NF EN ISO 6888-1, NF EN ISO 6888-3 and NF V08-057-1.

2 HISTORY

The formula with egg yolk, developed by Baird-Parker in 1962, was found to be particularly appropriate for the enumeration of coagulase positive staphylococci. In 1964, Smith and Baird-Parker showed that adding sulfamethazine to the medium inhibited the growth of *Proteus* and in 1971, Tardio and Baer observed, that among 18 selective isolation media tested, that the Baird-Parker formulation was less inhibitory than Vogel-Johnson medium, used previously with some frequency.

3 PRINCIPLES

The growth of staphylococci is favored by sodium pyruvate and glycine.

Accompanying microflora is inhibited by lithium chloride and potassium tellurite (added extemporaneously), as well as a high concentration of glycine.

The addition of sulfamethazine (optional) after autoclaving inhibits most *Proteus* and thus limits the invasion of the medium by this species.

Enrichment with egg yolk aids in identification by showing the action of lecithinase.

Staphylococcus aureus presents black colonies (due to the reduction of tellurite to telluride), surrounded by clearing zones in the cloudy egg yolk medium.

In principle, other microorganisms are inhibited. However, it is possible to observe brown or greenish micrococci, white yeast colonies or brown *Bacillus* or *Proteus*.

Coagulase negative staphylococci are nearly totally inhibited and if a culture does develop, it will not be surrounded by clearing zones.

4 TYPICAL COMPOSITION

The composition can be adjusted to obtain optimal performance.

For 1 liter of complete medium:

- Tryptone	10,0 g
- Meat extract	5,0 g
- Yeast extract	1,0 g
- Sodium pyruvate	10,0 g
- Glycine	12,0 g
- Lithium chloride	5,0 g
- Bacteriological agar.....	15,0 g
- Egg yolk emulsion	47,0 mL
- Potassium tellurite at 3,5%.....	3,0 mL
- Sulfamethazine (if necessary).....	50,0 mg

pH of the ready-to-use medium at 25°C: 7.2 ± 0.2.

For 58 g dehydrated base BK055

- Tryptone	10,0 g
- Meat extract	5,0 g
- Yeast extract	1,0 g
- Sodium pyruvate	10,0 g
- Glycine	12,0 g
- Lithium chloride	5,0 g
- Bacteriological agar	15,0 g

For one vial of supplement BS060

(50 mL)	
- Egg yolk emulsion	47,0 mL
- Potassium tellurite at 3,5%	3,0 mL

For one vial of supplement BS036

(900 mL)	
- Egg yolk emulsion	846,0 mL
- Potassium tellurite at 3,5%	54,0 mL

For one vial of supplement BS028

- Sulfamethazine	25,0 mg
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5 PREPARATION

- Suspend 58.0 g of dehydrated base medium (BK055) in 950 mL of distilled or deionized water.
- Slowly bring to boiling, stirring with constant agitation until complete dissolution.
- Dispense in flasks by adding 95 mL per flask.
- Sterilize in an autoclave at 121°C for 15 minutes.
- Cool and maintain at 44-47°C.
- Aseptically add 5 mL of Egg Yolk Tellurite Enrichment (BS060) per flask.
- Mix rapidly and thoroughly.
- Pour into sterile Petri dishes and let solidify on a cool surface.
- Dry the plates in an incubator with the covers partially removed.

✓ **Reconstitution:**
58,0 g for 950 mL

✓ **Sterilization:**
15 min at 121 °C

✓ **Additives:**
50 mL BS060

NOTE:

After cooling to 44-47 °C, it is possible to add sulfamethazine to limit invasion by *Proteus*. In this case, rehydrate the freeze-dried supplement (BS028) with 5 mL of sterile distilled water and add 1 mL per vial of base media.

6 INSTRUCTIONS FOR USE

Enumeration of coagulase positive staphylococci (Food microbiology, NF EN ISO 6888-1)

- To the surface of prepared media or pre-poured plates (BM018, BM091) brought to room temperature, transfer 0.1 mL of the sample to analyze and its serial tenfold dilutions.
- Spread the inoculum on the surface of the agar with a sterile triangle.
- Incubate at 35 °C or at 37 °C for 24 to 48 ± 4 hours.

✓ **Inoculation:**
0,1 mL surface plating

✓ **Incubation:**
24 & 48 h at 35-37 °C

Detection of *Staphylococcus aureus* (Cosmetics, NF EN ISO 22718)

- To the surface of prepared media or pre-poured plates (BM018, BM091) brought to room temperature, inoculate by streaking a loop of enrichment broth (Eugon LT 100).
- Incubate at 30-35 °C for 24 to 48 hours.

✓ **Inoculation:**
Streaking on surface

✓ **Incubation:**
24 to 48 h at 30-35 °C

7 RESULTS

Staphylococcus aureus is characterized by the formation of black or grey colonies (reduction of tellurite to telluride), which are surrounded by clear zones (egg yolk reaction) resulting from proteolysis. An opaque zone due to the action of lecithinase may appear inside the halo.

Non-characteristic colonies are black or grey with or without a clear zone.

Presumptive *Staphylococcus aureus* must be confirmed by the coagulase tube test (Lyophilized Rabbit Plasma, BR002) or by re-streaking onto Baird Parker with Rabbit Plasma Fibrinogen (BM067).

See ANNEX 1: PHOTO SUPPORT

NOTE: For more details on non-characteristic colonies, please refer to NF EN ISO 6888-1.

8 QUALITY CONTROL

Dehydrated base medium: cream-white powder, free-flowing and homogeneous.

Aspect of liquid supplement: yellowish emulsion, opaque, having a precipitate that can be resuspended.

Aspect of freeze-dried supplement: white pellet, after reconstitution giving a clear, limpid solution.

Prepared media (complete): yellow agar, cloudy.

Typical culture response after 48 hours of incubation at 37°C (NF EN ISO 11133):

Microorganisms	Growth (Productivity Ratio : P_R)	Characteristics / halo of clearing
<i>Staphylococcus aureus</i> WDCM 00034	$P_R \geq 50\%$	Black colonies, with halo
<i>Staphylococcus epidermidis</i> WDCM 00036	Slowed, score 0-1	Black colonies, without halo
<i>Staphylococcus saprophyticus</i> WDCM 00159	Slowed, score 0-1	Black colonies, without halo
<i>Escherichia coli</i> WDCM 00013	Inhibited, score 0	-

9 STORAGE / SHELF LIFE

Dehydrated base medium: 2-30 °C.

Egg yolk tellurite emulsion: 2-8 °C, shielded from light.

Pre-poured media in Petri dishes: 2-8 °C.

Sulfamethazine 25 mg selective supplement: 2-8 °C.

The expiration dates are indicated on the labels.

Prepared base media in vials (*): 180 days at 2-8 °C.

Prepared complete media in plates (*): 30 days at 2-8 °C.

Prepared complete media in vials (*): Not recommended; the reliquification will denature the media.

Reconstituted sulfamethazine selective supplement (*): 30 days at 2-8 °C.

(*): Benchmark value, determined in standard conditions of preparation, following manufacturer's instructions.

10 PACKAGING

Dehydrated base media (without egg yolk or tellurite)

500 g bottle BK055HA

5 kg drum BK055GC

Sterile Egg yolk emulsion with Potassium tellurite

Kit of 10 vials x 50 mL BS06008

Sterile Egg yolk emulsion with Potassium tellurite

1 vial of 900 mL BS03608

Sulfamethazine 25 mg selective supplement

Kit of 10 vials qsp 500 mL..... BS02808

Pre-poured media in Petri dishes (Ø 90 mm)

20 plates BM01808
120 plates BM09108

11 BIBLIOGRAPHY

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12 ADDITIONAL INFORMATION

The information provided on the labels take precedence over the formulations or instructions described in this document and are susceptible to modification at any time, without warning.

Document code : BAIRD PARKER EYT_EN_v13
Creation date : 02-2003.
Updated : 06-2020.
Origin of revision : Modification of the results part.

Baird-Parker agar with Egg yolk Tellurite

Detection and enumeration of *Staphylococcus aureus*

Results:

Incubation 48 hours at 37°C (surface)

Staphylococcus aureus

Characteristic colony:
Grey to black color, shiny, surrounded
by an opaque ring and a halo of
clearing.

