

18795 Brucella Agar

For the isolation and cultivation of *Brucella* species, in particular the pathogens *Bruc. melitensis*, *Bruc. abortus* and *Bruc. suis*, from clinical material and comestibles of animal origin, modified according to Wundt. This medium is also formulated to support luxuriant growth of Streptococci, Pneumococci, *Listeria*, *Neisseria meningitidis* and *Haemophilus influenzae*.

Composition:

Ingredients	Grams/Litre
Meat peptone	10.0
Casein peptone	10.0
Yeast extract	2.0
D(+)-Glucose	1.0
Sodium chloride	5.0
Agar	13.0

Final pH 7.0 +/- 0.2 at 25°C

Store prepared media below 8°C, protected from direct light. Store dehydrated powder, in a dry place, in tightly-sealed containers at 2-25°C.

Directions:

Dissolve 41 g in 1 litre distilled water, autoclave at 121°C for 15 minutes and pour plates.

The plates are clear and yellowish-brown.

For the Brucella differential agar sterilize Brucella agar, cool 50°C and adjust pH to 6.7 ± 0.1 . Add an aqueous 1 ml 2% basic fuchsin solution (Cat. No. 03968). Another possibility is to produce a 2% solution from thionine acetate (Cat. No. 88930). Boil the solution first for 20 minutes.

For the Brucella selective agar sterilize the Brucella agar, cool to 50°C and add the following compounds to 1 litre medium: Bacitracin (Cat. No. 11702) 500mg, Polymyxin B sulfate (Cat. No. P4932) 1mg, Cycloheximide (Cat. No. 01810) 100 mg and if required ethyl violet (Cat. No. 228842) 1.25 mg.

For isolation of Campylobacter species cool media to 50°C and add sterile filtered solution with 125 mg sodium pyruvate (Cat. No. P2256), 125 mg sodium metabisulfite (Cat. No. 31448) and 125 mg ferrous sulfate (Cat. No. 31236) in 2 ml of distilled water and 5-7% sterile defibrinated sheep blood and may as well 25 ml heat inactivated horse serum to 500 ml of medium base.

Principle and Interpretation:

Meat peptone, Casein peptone and Yeast extract provide nitrogenous nutrients and Vitamin B complexes. D(+)-Glucose serves as an energy source. It can be enriched with 5% defibrinated horse blood. The growth of accompanying microbial flora can be suppressed by addition of bacitracin, polymyxin, cycloheximide and possibly ethyl violet. Circulin, which also has been recommended originally, is no longer used. The various *Brucella* species can be differentiated by their different sensitivity to the dyes thionine and fuchsin. Differential culture media can be prepared by adding these two compounds to Brucella agar.



Cultural characteristics after 24-72 hours at 35°C.

Organisms (ATCC)	Growth	on thionine	on basic fuchsin
<i>Brucella abortus</i> (4315)	+++	growth	no
<i>Brucella melitensis</i> (4309)	+++	growth	growth
<i>Brucella suis</i> (4314)	+++	no	growth
<i>Escherichia coli</i> (25922)	+++	-	-

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Precautions and Disclaimer

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