mCCD Agar ACCORDING TO ISO 10272:2017

INSTRUCTION FOR USE READY-TO-USE PLATED MEDIA

For professional use

Intended use: mCCD Agar is used for the isolation and cultivation of Campylobacter spp.

Ref.:	Type of medium:	Packaging:
201008	ready-to-use medium-plate	1x10 pcs (90 mm)

1. Principle: meat extract, enzymatic digest animal tissue and enzymatic digest of casein serve as sources of essential nutrients and amino acids. The combination of activated charcoal, iron (II) sulfate and sodium pyruvate are used to absorb radicals and peroxides that might be inhibitory to the microaerophilic *Campylobacter*. Sodium chloride maintains the osmotic balance of the medium. The selective agents desoxycholate, cefoperazone and amphotericin B inhibit the accompanying bacterial flora as well as yeasts and moulds Agar is the solidifying agent.

2. Formula/Liter:	Supplements/Liter:		
Meat extract	10.0 g	Amphotericin B	0.01 g
Enzymatic digest of animal tissue	10.0 g	Cefoperazone	0.032 g
Sodium chloride	5.0 g		
Activated charcol	4.0 g		
Enzymatic digest of casein	3.0 g		
Sodium deoxycholate	1.0 g		
Iron (II) sulfate hydrate	0.25 g		
Sodium puryvate	0.25 g		
Agar	14.0 g		

3. pH: 7.4 ± 0.2 at 25°C.

4. Appearance:

Prepared Appearance: prepared medium is black and homogenous.

5. Sample: samples taken from the liquid medium Bolton Broth (Ref.: 3412), Preston Broth (Ref.: 3413), food samples or samples from the primary production stage.

6. Test procedure: <u>detection procedure A</u>: if the agar plate has been refrigerated, allow to warm to room temperature before inoculation. Using the culture obtained in the enrichment medium Bolton Broth (Ref.: 3412), inoculate with a sterile 10 μ l loop the surface of the medium. Incubate plates at 41.5 °C in an microaerobic atmosphere for 44±4°C. <u>Detection procedure B</u>: if the agar plate has been refrigerated, allow to warm to room temperature before inoculation. Using the culture obtained in the enrichment medium Preston Broth (Ref.: 3413), inoculate with a sterile 10 μ l loop the surface of the medium. Incubate plates at 41.5 °C in an microaerobic atmosphere for 44±4°C. <u>Detection procedure B</u>: allow to warm to room temperature before inoculation. Using the culture obtained in the enrichment medium Preston Broth (Ref.: 3413), inoculate with a sterile 10 μ l loop the surface of the medium. Incubate plates at 41.5 °C in an microaerobic atmosphere for 44±4°C. <u>Detection procedure C</u>: if the agar plate has been refrigerated, allow to warm to room temperature before inoculation. For caecal or faecal samples, use loop or a sterile swab to bring some of the well-mixed sample material onto the first half plate. Use another loop to streak out on the second half of the plate. For all other samples, add an appropriate amount of liquid (e.g peptone salt solution or Preston Broth), for example , 1 in 2 (volume fraction), mix well and either streak the plate using a loop, or dispense a suitable volume an spread it over the plate. Incubate plates at 41.5 °C in an microaerobic atmosphere for 44±4°C.

7. Results: after incubation examine the plates for typical and / or suspect colonies of *Campylobacter*. Make a confirmation tests.

8. Quality control: perform quality control testing for both negative and positive reaction by inoculating a representative sample of plates with pure cultures of stable control organisms that produce known, desired reactions according to ISO 11133:2014.

Microorganism:	Method of control:	Criteria:	Appearance of colony:	
Campylobacter jejuni WDCM 00156	productivity: quantitative	PR ≥0,5	greyish, flat and moist colonies, sometimes with metallic sheen	
Campylobacte coli WDCM 00004	productivity: quantitative	PR ≥0,5	greyish, flat and moist colonies, sometimes with metallic sheen	
Escherichia coli WDCM 00013	selectivity: qualitative	Total or partial inhibition (0-1)		
Staphylococcus aureus WDCM 00034	selectivity: qualitative	Total inhibition (0)	—	

9. Precautions: due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

10. Disposal of waste: after use, all plates and any other contaminated materials must be sterilized or disposed of in line with appropriate internal procedures and in accordance with local legislations. Plates can be destroyed by autoclaving at 121°C for at least 20 minutes.

11. Storage: on receipt, store plates at 2-12°C away from direct sun light in an inverted position. Do not overload a refrigerator with excessive amounts of plates to avoid water condensation on the lids during storage. Plates must not come into direct contact with the inner walls of refrigerator, as the media may freeze, invalidating the tests. Prepared plates, stored in their original sleeve wrapping at 2-12°C until just prior to use, may be inoculated up to the expiration date and incubated for recommended incubation times. Plates from opened stacks of 10 plates should be used for two weeks when stored in a clean area at 2 to 12° C. Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking or others signs of deterioration. Allow the medium to warm to the room temperature before inoculation.

All microbiological media containing dyes or light-sensitive components should be protected from light and stored in the dark.

Note that shelf life of the growth media changes after the addition of supplements. Complete media containing protein supplement tend to degrade faster than basal media alone.

12. Shelf life: 3 months.

13. Required supplements not supplied together with medium base: not applicable

14. References: available on request.



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