Technical Data Sheet

Differential Reinforced Clostridial Broth (DRCM)

Ordering number: 1.11699 .0500

Medium proposed by GIBBS and FREAME (1965) for the enumeration of all clostridia by the MPN method in foodstuffs and other materials.

This culture medium was successfully utilized by FREAME and FITZPATRICK (1971) and GIBBS (1973) to isolate and count clostridia. The Institute for Food Technology and Packing of the Technical University of Munich (Institut für Lebensmitteltechnologie und Verpackung der TU München) (1976) recommends this medium for the examination of packing materials. It complies with the requirements of the DIN Norm 38411 for the examination of water.

Mode of Action

Differential Reinforced Clostridial Broth represents a development of the Reinforced Clostridial Media proposed by HIRSCH and GRINDSTED (1954) and GIBBS and HIRSCH (1956). The redox indicator resazurin is used to monitor anaerobiosis. Clostridia reduce sulfite to sulfide, the formed iron sulfide causes the culture medium to turn black. As other bacteria can also produce sulfide, vegetative forms must first be removed from the culture by a relevant treatment (e.g. pasteurization), and the anaerobic spore-forming microorganisms must then be identified. GIBBS and FREAME (1956) inhibited the growth of most non-spore-forming microorganisms by adding polymyxin (70 IU/ml) to the broth.

Typical Composition (g/L)

Differential Reinforced Clostridial Broth (DRCM)	
Peptone from casein	5.0
Peptone from meat	5.0
Meat extract*	8.0
Yeast extract	1.0
Starch	1.0
D(+)glucose	1.0
L-cysteinium chloride	0.5
Sodium acetate	5.0
Sodium di-sulfite	0.5
ammonium iron(III) citrate	0.5
Sodium resazurin	0.002

* Meat extract is equivalent to the term beef extract.

Preparation

Suspend 27.5 g/litre, dispense into test tubes, autoclave (15 min at 121°C).

pH: 7.1 ± 0.2 at 25°C.

The ready-to-use broth in the tube is clear and reddish-brown.

Experimental Procedure and Evaluation

Inoculate the culture medium, cover with a 3 to 5 mm layer of sterilized paraffin viscous and pasteurize (30 min at 75 °C in a water bath!).

Incubation: at least 7 days at 30°C.

Microbial growth can usually be seen after 3-4 days. The cultures should be observed for up to 4 weeks as occasionally some time is required for spore germination to start. The cultures should be checked for a black colouration. Further tests should be performed to identify the clostridia.

Note for testing milk samples:

Milk can curdle or coagulate in the presence of strongly acid- and gas-forming microorganisms. This curding prevents blackening of the broth. If milk coagulation and strong gas formation occur in testing a milk sample, this may be suspected to contain Clostridium perfringens. Further tests should follow for a more precise identification.

Storage

The prepared culture medium can be stored for up to 2 weeks.

Quality Control

Control strains	Growth	Blackening
Escherichia coli ATCC 25922 (WDCM 00013)	Good to very good	-
Bacillus cereus ATCC 11778 (WDCM 00001)	Fair to good	-
Pseudomonas aeruginosa ATCC 27853 (WDCM 00025)	Poor to fair	-
Clostridium bifermentans ATCC 19299	Good to very good	+
Clostridium perfringens ATCC 10543	Good to very good	+
Clostridium perfringens ATCC 13124 (WDCM 00007)	Good to very good	+
Clostridium sporogenes ATCC 11437	Good to very good	+
Clostridium sporogenes ATCC 19404 (WDCM 00008)	Good to very good	+

Please refer to the actual batch related Certificate of Analysis.

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Literature

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Ordering Information

Product	Cat. No.	Pack size
Differential Reinforced Clostridial Broth (DRCM)	1.11699.0500	500 g
Paraffin viscous	1.07160.1000	1 l
Polymyxin	5291	

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