Technical Data Sheet

ReadyPlate™ PEMBA (Polymyxin pyruvate egg yolk mannitol bromothymol blue agar) acc. ISO 21871

Ordering number: 1.46711.0100

General

The composition of medium complies with ISO 21871

Mode of Action

This selective culture medium utilizes two characteristic properties of B. cereus. B. cereus does not form acids from mannitol. Mannitol-positive bacteria grow yellow colonies with a yellow border. B. cereus, however, grows with turquoise to blue colonies, since the medium is slightly alkalized by its metabolites (bromothymol blue reaction). The yolk reaction (by lecithinase) also leads to a precipitate surrounding the colony, which appears in the color of the agar or similar to the colony.
Growth and lecithinase activity are often so strong that the precipitation is already evident before a colony is visible. Polymyxin B inhibits the growth of gram-negative bacteria. Some Proteus spp. and gram-positive cocci can grow.

Typical Composition (g/liter):

Specified by ISO 21871		ReadyPlate™ PEMBA ISO 21871	
Enzymatic digest of casein	1	Enzymatic digest of casein	1
D-Mannitol	10	D-Mannitol	10
Sodium pyruvat	10	Sodium pyruvat	10
Magnesium sulfate heptahydrate	0,1	Magnesium Sulfate · 7 H2O	0.1
Sodium chloride	2	Sodium chloride	2
Disodium hydrogen phosphate	2,5	Disodium hydrogen phosphate	2,5
Potassium dihydrogen phosphate	0,25	Potassium dihydrogen phosphate	0,25
Bromthymol blue	0,12	Bromthymol blue	0,12
Egg yolk emulsion 20%	50 ml	Egg yolk emulsion 20%*	50 ml
Polymyxin B sulfate	100,000 IU	Polymyxin B sulfate	100,000 IU
Agar	9-18	Agar	12,5
Water	940	Water	n/a
pH at 25°C	7.2±0.2	pH at 25°C	7.2±0.2

** SO 21871 states that for usage of a commercial egg yolk emulsion, the concentration should be used according to the manufacturer's instruction.*

Application and Interpretation

Following the procedure for detection method given by ISO 21871. Add 1ml of the initial suspension to a 9ml single strength Tryptone Soya Polymixin Broth (TSPB) or 10ml of the initial suspension to a 10ml double strength TSPB and incubate at 30°C for 48h ± 4

Mix the incubated broth thoroughly and using an inoculation loop streak the culture onto the surface of the PEMBA or MYP agar and incubate downwards at 37°C for PEMBA or 30°C for MYP for 18 to 24 hours.

Result: Typical colonies of preseumptive Bacillus *cereus* are about 2 to 5 mm in size. Turquoise to peacock blue with possible greyish white colony center against a blue background and has precipitation halo up to 5mm wide.

Following the procedure for enumeration method given by ISO 21871. Inoculate 10ml of the initial suspension into 3 tubes of double strength TSPB. In parallel inoculate 1ml of the initial suspension into 3 tubes of single strength of single strength TSPB. Incubate at 30°C for 48h ± 4

Mix the incubated broth thoroughly and using an inoculation loop streak the culture onto the surface of the PEMBA or MYP agar and incubate downwards at 37°C for PEMBA or 30°C for MYP for 18 to 24 hours. Additional 24 hours incubation, if necessary.

Select 3 colonies and confirm using Haemolysis test or microscopic examination from the PEMBA plates. Calculate the MPN.

Storage and Shelf life

The product can be used for tests until the expiry date if stored upright, protected from light and properly sealed at +2 °C to +8 °C.

The testing procedures as described on the CoA can be started up to the expiry date printed on the label.

Condensation can be prevented by avoiding quick temperature shifts and mechanical stress.

Quality Control

Function	Incubation	Control strains	Reference medium	Method of control	Criteria (% Recovery)	Characteristic reactions
Productivity	(21±3) h to (44±4) h / (37±1)°C	*Bacillus cereus* ATCC® 11778 WDCM 00001	-	Qualitative	Good growth	Turquoise-blue colonies with precipitation halo
Selectivity	(44±4) h / (37±1)°C	*Escherichia coli* ATCC® 8739 WDCM 00012	-	Qualitative	Total inhibition	-
		Escherichia coli ATCC® 25922 WDCM 00013				
Specificity		Bacillus *subtilis* ATCC® 6633 WDCM 00003	-	Qualitative	-	White colonies without precipitation halo

The performance test is in accordance with the current version of EN ISO 11133

Literature

Mossel, D. A. A., Koopman, M. J., Jongerius, E. (1967): Enumeration of *Bacillus cereus* in foods. Appl. Microbiol. **15**, 650-653.

Holbrook, R., Anderson, J. M. (1980): An im-proved selective and diagnostic medium for the isolation of *Bacillus cereus* in foods. Can. J. Microbiol. **26**, 753-759.

Farrar, W. E., Reboli, A. C. (1991): The Genus Bacillus. In: Balows, A. et al. eds. The Prokaryotes. Second Edit., Vol. II: 1746-1768.

DIN EN ISO 21871:2006-04: Microbiology of food and animal feeding stuffs - Horizontal method for the determination of low numbers of presumptive Bacillus cereus - Most probable number technique and detection method.

Ordering Information

Product	Cat. No.	Pack size
ReadyPlate™ PEMBA ISO 21871	1.46711.0100	100 x 90mm
ReadyPlate™ MYP ISO 7932, 21871	1.46160.0020	20 x 90mm
Sheep Blood Agar	1.46740.0020	20 x 90mm
Certistain® Malachite green oxalate (C.I. 42000)	1.15942.0025	25g
Safranine solution	1.09217.0500	500ml

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