Technical Data Sheet

GranuCult® prime

MacCONKEY (MAC) agar
acc. ISO 21150, ISO 21567, FDA-BAM and EP, USP, JP

Ordering number: 1.00205.0500 / 1.00205.5000

For the isolation and confirmation of *Escherichia coli* and other *Enterobacteriaceae*, e.g. *Salmonella* and *Shigella*, from the food chain, cosmetic, pharmaceutical and other materials.

This culture medium complies with the specifications given by EN ISO 21150, EN ISO 21567, FDA-BAM Medium M91, APHA Part 9225 and Part 9260, AOAC Official Method 967.25, GB 4789.5, GB 4789.6 and with the specifications given by the harmonized methods of EP, USP, JP for Microbial Examination of Non-sterile Products: Tests for Specified Microorganisms.

This culture medium is released by the quality control laboratory of Merck KGaA, Darmstadt, Germany. The laboratory is accredited by the German accreditation authority DAkkS as registered test laboratory D-PL-15185-01-00 according to DIN EN ISO/IEC 17025 for the performance testing of media for microbiology according to DIN EN ISO 11133.

Mode of Action

This culture medium contains crystal violet and bile salts that inhibit the growth of the Gram-positive microbial flora whilst it allows the Gram-negative bacteria to grow. Peptones provide nitrogen, vitamins and amino acids. Lactose and the pH indicator neutral red are used to detect lactose degradation. The fermentation of lactose is causing a local drop down of the pH, resulting in a colour change of the pH indicator neutral red. By this, reddish-pink colonies are formed and the medium around the colonies changes its colour to reddish. Often a bile precipitation zone around the colony is formed. Bile salts and crystal violet inhibits the growth of Gram-positive bacteria. Agar is the solidifying agent.

Typical Composition

APHA specifies no composition for MacConkey (MAC) agar.

Specified by EN ISO 21150		Specified by EN ISO 21567, GB 4789.5, GB 4789.6		Specified by EP/JP/USP, FDA-BAM Medium M91, AOAC 967.25		GranuCult® prime MacConkey (MAC) agar acc. ISO 21150, ISO 21567, FDA-BAM and EP/USP/JP	
Pancreatic digest of gelatin	17.0 g/l	Enzymatic digest of casein and animal tissues*	20.0 g/l	Pancreatic digest of gelatin **	17.0 g/l	Pancreatic digest of gelatin	17.0 g/l
Peptic digest of casein	1.5 g/l			Peptones (meat and casein)**	3.0 g/l	Peptic digest of casein	1.5 g/l
Peptic digest of animal tissue	1.5 g/l					Peptic digest of animal tissue	1.5 g/l
NaCl	5.0 g/l	NaCl	5.0 g/l	NaCl	5.0 g/l	NaCl	5.0 g/l
Lactose	10.0 g/l	Lactose	10.0 g/l	Lactose	10.0 g/l	Lactose	10.0 g/l
Bile salt mixture	1.5 g/l	Bile salt mixture	1.5 g/l	Bile salts	1.5 g/l	Bile salt mixture	1.5 g/l
Neutral red	0.04	Neutral red	0.04	Neutral red	0.03	Neutral red	0.03
Crystal violet	0.001	Crystal violet	0.001	Crystal violet	0.001	Crystal violet	0.001
Agar	13.5 g/l	Agar	9.0 g to 18.0 g/l ***	Agar	13.5 g/l	Agar-agar ****	13.5 g/l
Water	1000 ml	Water	1000 ml	Water	1000 ml	Water	n/a
pH at 25 °C	7.1 ± 0.2	pH at 25 °C	7.1 ± 0.2	pH at 25 °C	7.1 ± 0.2	pH at 25 °C	7.1 ± 0.2

* GB 4789.5 and GB 4789.6 specify "Peptone 20 g/l".

** FDA-BAM Medium M91 specifies "Peptone or gelysate 17.0 g/l" and "Proteose peptone or polypeptone 3 g/l". Proteose peptone is a term for a mixed peptone from animal tissues and casein.

*** Depending on the gel strength of the agar.

**** Agar-Agar is equivalent to other different terms of agar.

Preparation

Dissolve 50.0 g in 1 liter of purified water. Heat in boiling water and agitate frequently until completely dissolved. Autoclave (15 minutes at 121 °C). Pour to plates.

The dehydrated medium is a granulate with red brown color.

The prepared medium is clear and dark-red. The pH value at 25 °C is in the range of 6.9 - 7.3.

Before inoculation, allow the prepared medium to equilibrate at room temperature if it was stored at a lower temperature.

There should be no visible moisture on the plates before use. When moisture is present, the plates should be dried for the minimum time required to remove visible moisture, following the procedure as described by EN ISO 11133.

Experimental Procedure and Evaluation

Depend on the purpose for which the medium is used.

Following the procedure given by EN ISO 21150, ISO 21567, GB 4789.5, GB 4789.6
inoculate the surface of the medium from
the selective enriched cultures so that well-isolated colonies will be obtained.

Incubate the inoculated plates inverted under aerobic conditions, e.g.

- acc. to EN ISO 21150 at (32,5 ± 2,5 °C) for at least 24 h;
- acc. to EN ISO 21567 at (37 ± 1 °C) for between 20 h and 24 h;
- acc. to FDA-BAM Chapter No. 6 at (35 ± 2 °C) for 20 h;
- acc. to EN ISO GB 4789.5 at (36 ± 1 °C) for between 20 h and 24 h;
- acc. APHA Part 9225 at (35 ± 0,5 °C) for at least 24 h;
- acc. APHA Part 9260 at 35 °C overnight;
- acc. to APHA Chapter No. 37 at (35 ± 2 °C) for (16 to 24 h);
- acc. to EP/USP/JP at (30 - 35 °C) for (18 to 24 h);

On MacCONKEY (MAC) agar acc. ISO 21150, ISO 21567, FDA-BAM and EP, USP, JP :
Shigella spp. produce colourless (sometimes to pale pink) translucent colonies without precipitation zone.
Salmonella spp. produce colourless translucent colonies and medium yellowished coloured around the colonies.
Escherichia coli produce reddish-pink colonies mostly with precipitation zone and medium reddish coloured around the colonies.

Colonies of the most important bacteria usually have the appearance described below:

Appearance of colonies	**Microorganisms**
Colourless, translucent	*Salmonella, Shigella* and others
Large, red, surrounded by a turbid zone of precipitation	*Escherichia coli*
Large, pink, mucoid	*Enterobacter, Klebsiella*
Very small, opaque, isolated colonies	Enterococci, staphylococci others

This presumptive evidence must be confirmed by carrying out the usual tests.

Storage

Store at +15 °C to +25 °C, dry and tightly closed. Do not use clumped or discolored medium. Protect from UV light (including sun light). For *in vitro* use only.

Acc. to EN ISO 21567, self-prepared plates can be stored in the dark and protected against evaporation at (5 ± 3 °C) for up to two weeks.

Microbiological Performance

The performance test is in accordance with the current version of EN ISO 11133 and EP/JP/USP.
Test method: Performance testing of solid culture media - Quantitative and qualitative methods

Qualitative method for solid media		
Test strain	**Specification**	
	Growth	**Typical reaction**
Escherichia coli ATCC® 25922 [WDCM 00013]	good	reddish-pink colonies with precipitation zone, medium reddish coloured
Salmonella Typhimurium ATCC® 14028 [WDCM 00031]	good	colourless translucent colonies without precipitation zone, medium yellowish coloured
Salmonella Enteritidis ATCC® 13076 [WDCM 00030]	good	colourless translucent colonies without precipitation zone, medium yellowish coloured
Shigella flexneri ATCC® 12022 [WDCM 00126]	good	colourless translucent colonies without precipitation zone, medium yellowish coloured
Shigella flexneri ATCC® 29903 [WDCM 00125]	good	colourless translucent colonies without precipitation zone, medium yellowish coloured
Shigella sonnei ATCC® 29930 [WDCM 00127]	good	colourless translucent colonies without precipitation zone, medium yellowish coloured
Proteus mirabilis ATCC® 29906 [WDCM 00023]	good	colourless translucent colonies without precipitation zone, medium yellowish coloured
Enterococcus faecalis ATCC® 29212 [WDCM 00087]	no growth or partial inhibition	if growth: red small colonies without precipitation zone
Staphylococcus aureus ATCC® 6538 [WDCM 00034]	no growth	not applicable

Incubation: 20 ± 2 h at 35 ± 1 °C, aerobic.

Quantitative method for solid media			
Test strain	**Specification**		
	Inoculum	**Recovery rate**	**Typical reaction**
Escherichia coli ATCC® 8739 [WDCM 00012]	10 – 100 cfu	50 – 150 %	reddish-pink colonies, with or without precipitation zone, medium reddish coloured

Incubation: ≤ 18 hours at 30 – 35 °C, aerobic.
Reference medium: Tryptic Soy agar.

Please refer to the actual batch related Certificate of Analysis.

Literature

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EN ISO International Standardisation Organisation. Microbiology of food, animal feed and water - Preparation, production, storage and performance testing of culture media + Amendment 1 + Amendment 2. EN ISO 11133:2014/Amd1:2018/Amd2:2020.

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United States Pharmacopeial Convention. (2020): The United States Pharmacopeia 43/National Formulation 38. Chapter <62> Microbiological examination of non-sterile products: Test for specified products. Rockville, Md., USA.

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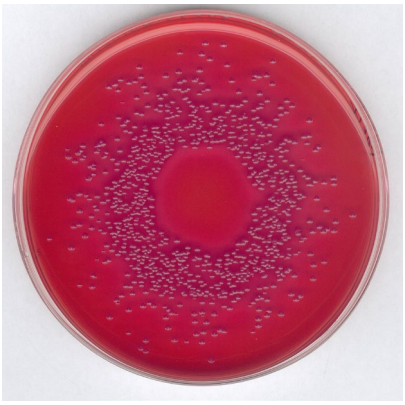

Escherichia coli ATCC 11775
on MacConkey agar

Salmonella Typhimurium ATCC 14028
on MacConkey agar

Ordering Information

Product	Cat. No.	Pack size
GranuCult® prime MacConkey (MAC) agar acc. ISO 21150, ISO 21567, FDA-BAM and EP/USP/JP	1.00205.0500	500 g
GranuCult® prime MacConkey (MAC) agar acc. ISO 21150, ISO 21567, FDA-BAM and EP/USP/JP	1.00205.5000	5 KG