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**Technical Data Sheet** 

### GranuCult<sup>®</sup> prime BPLS (Brilliant-green Phenol-red Lactose Sucrose) agar ref. to ISO 6579 Ordering number: 1.00207.0500

For the isolation and differentiation of *Salmonella* spp. from food and animal feed, water and other materials.

BPLS (Brilliant-green Phenol-red Lactose Sucrose) agar ref. to ISO 6579 is also known as Brilliant green agar (BGA) and Phenol red brilliant green agar.

This culture medium complies with the specifications given for Agar medium L (Brilliant green, phenol red, lactose monohydrate, sucrose agar) by 6<sup>th</sup> Edition of European Pharmacopoeia (2008), 2.6.13 A. Microbiological examination of non-sterile products: Test for specified Microorganisms - Method of the European Pharmacopeia (non-harmonised method), APHA and reference to EN ISO 6579-1.

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 lactose, whose degradation to acid is indicated by the pH indicator phenol red, which changes its color to yellow. The indicator exhibits a pink-red color in the alkaline range.

In this highly selective culture medium, the increased growth of accompanying microorganisms is considerably prevented by raising the concentration of brilliant green.

The growth of *Salmonella* is improved by the richer nutrient base, but *Salmonella* Typhi and *Shigella* spp. are largely inhibited by brilliant green.

Most *Salmonellae* are not able to ferment either lactose or sucrose. The sucrose contained in this medium allows identification of accompanying, weakly lactose-positive or lactose-negative, but sucrose-positive microorganisms. Phenol red serves as a pH indicator and yields a yellow color as a result of in the fermentation of lactose and/or sucrose in the medium. Agar is the solidifying agent.

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#### **Typical Composition**

There is no composition specified by EN ISO 6579-1 or APHA for Brilliant green agar.

Specified by EP 6 <sup>th</sup> Edition Chapter 2.6.13 A. (non-harmonised method)		GranuCult® prime BPLS (Brilliant-green Phenol-red Lactose Sucrose) agar ref. to ISO 6579		
Peptones	10.0 c/l	Enzymatic digest of casein	5.0 g/l	
(meat and casein)	10.0 g/i	Enzymatic digest of meat	5.0 g/l	
Yeast extract	3.0 g/l	Yeast extract	3.0 g/l	
Sodium chloride	5.0 g/l	Sodium chloride	5.0 g/l	
Lactose	10.0 g/l	Lactose	10.0 g/l	
Sucrose	10.0 g/l	Sucrose	10.0 g/l	
Phenol red	80 mg/l	Phenol red	0.08 g/l	
Brillant green	12.5 mg/l	Brillant green	0.0125 g/l	
Agar	20.0 g/l*	Agar-agar**	13.0 g/l	
Water	1000ml	Water n/		
pH at 25 °C	$6.9 \pm 0.2$	pH at 25 °C 6.9 ± 0.2		

\* Depending on the gel strength of the agar.

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

#### Preparation

Dissolve 51.0 g in 1 liter of purified water. Heat in boiling water and agitate frequently until completely dissolved. Autoclave (15 minutes at 121°C) and pour plates.

The dehydrated medium is a granulate with red-brown color.

The prepared medium is clear to slightly opalescent and red. The pH value at 25  $^{\circ}$ C is in the range of 6.7 - 7.1.

Before inoculation, allow the prepared medium to equilibrate at room temperature if stored before 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 6579-1, inoculate the surface of the medium from the selective enriched cultures so that well-isolated colonies will be obtained.

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Incubate the inoculated plates inverted under aerobic conditions, e.g. acc. to EN ISO 6579-1 at (37  $\pm$  1 °C) for (24  $\pm$  2 h).

BPLS (Brilliant-green Phenol-red Lactose Sucrose) agar ref. to ISO 6579, *Salmonella* spp. and other lactose- and sucrose-negative microorganisms produce pink-red colored colonies, with or without a pink-red colored zone in the medium.

Lactose- and/or sucrose-positive microorganisms produce yellow-green colonies mostly surrounded by a yellow-green colored zone in the medium.

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

Appearance of colonies	Microorganisms		
Pink-red mostly surrounded by a pink-red zone	Lactose- and sucrose-negative: Salmonella spp. and others.		
Yellow-green mostly surrounded by a yellow-green zone	Lactose- and/or sucrose-positive: Escherichia coli, Citrobacter spp., Proteus vulgaris, Klebsiella spp. and others. Occasionally complete inhibition of growth.		

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.

Self-prepared plates can be stored acc to Corry et al. (2012) at  $(5 \pm 3 \text{ °C})$  for up to 4 days in the dark and protected against evaporation.

#### **Microbiological Performance**

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

Test method: Performance testing of solid culture media - Qualitative testing

	Specification			
Test strain	Growth Typical reaction			
Salmonella Typhimurium ATCC <sup>®</sup> 14028 [WDCM 00031]	good	pink-red colored colonies, with or without a pink-red zone in the medium		
Salmonella Enteritidis ATCC <sup>®</sup> 13076 [WDCM 00030]	good	pink-red colored colonies, with or without a pink-red zone in the medium		
<i>Escherichia coli</i> ATCC <sup>®</sup> 25922 [WDCM 00013]	weak to good	yellow colonies, with a yellow zone in the medium		
Enterococcus faecalis ATCC <sup>®</sup> 29212 [WDCM 00087]	none to weak	If growth: yellow colonies, with a yellow zone in the medium		

Incubation:  $22 \pm 2$  h at  $37 \pm 1$  °C, aerobic.

Please refer to the actual batch related Certificate of Analysis.

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#### Literature

APHA (2015) Chapter No. 67: Microbiological media, reagents and stains. Compendium of Methods for the Microbiological Examination of Foods. 5<sup>th</sup> ed. American Public Health Association, Washington, D.C.

EN ISO International Standardisation Organisation. Microbiology of the food chain - Horizontal method for the detection, enumeration and serotyping of *Salmonella* - Part 1: Horizontal method for the detection of *Salmonella* spp. + Amendment 1. EN ISO 6579-1:2017/Amd1:2020.

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.

European Directorate for the Quality of Medicines and Healthcare (2008): The European Pharmacopoeia. 6<sup>th</sup> Ed. Chapter 2.6.13 A. Microbiological examination of non-sterile products: Test for specified Microorganisms - Method of the European Pharmacopeia (non-harmonised method). Strasbourg, France.

Andrews, W.H., Wilson, C.R., Poelma, P.L. and Romero, A. (1979): Relative Productivity of Five Selective Plating Agars for the Recovery of *Salmonella* from Selected Food Types. J. Assoc. Off. Anal. Chem. 62: 320-326.

Andrews, W.H., Poelma, P.L. and Wilson, C.R. (1981): Comparative Efficiency of Brilliant Green, Bismuth Sulfite, Salmonella-Shigella, Hektoen Enteric, and Xylose Lysine Desoxycholate Agars for the Recovery of *Salmonella* from Foods: Collaborative Study. J. Assoc. Off. Anal. Chem. **64**: 899-928.

Corry, J.E.L., Curtis, G.D.W. and Baird, R.M. (2012): Phenol red brilliant green agar. In: Handbook of Culture Media for Food and Water Microbiology, pp. 866-869. Royal Society of Chemistry, Cambridge, UK.

Mooijman, K.A. (2012): Culture media for the isolation of *Salmonella*. In: Handbook of Culture Media for Food and Water Microbiology. (Corry, J.E.L., Curtis, G.D.W. and Baird, R.M. eds). pp. 261-286. Royal Society of Chemistry, Cambridge, UK.



Salmonella Enteritidis ATCC<sup>®</sup> 14028 [WDCM 00031]



Proteus vulgaris ATCC<sup>®</sup> 13315



Escherichia coli ATCC<sup>®</sup> 25922[WDCM 00013]

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#### **Ordering Information**

Product	Cat. No.	Pack size	Other pack sizes available
GranuCult® prime BPLS (Brilliant-green Phenol-red Lactose Sucrose) agar ref. to ISO 6579	1.00207.0500	500 g	
GranuCult <sup>®</sup> Buffered Peptone Water acc. ISO 6579, ISO 21528, ISO 22964, FDA-BAM and EP	1.07228.0500	500 g	5 kg, 25 kg
GranuCult <sup>®</sup> MKTTn (MULLER-KAUFFMANN Tetrathionate Novobiocin) Broth (Base) acc. ISO 6579	1.05878.0500	500 g	
Potassium Iodide	1.05043.0250	250 g	
Iodine resublimed	1.04761.0100	100 g	
GranuCult <sup>®</sup> RVS (RAPPAPORT-VASSILIADIS-Soya) Broth (Base) acc. ISO 6579	1.07700.0500	500 g	
Novobiocin Sodium Salt	N6160-1-G	1 g	
MSRV (Modified Semi-solid RAPPAPORT - VASSILIADIS) Medium (Base) acc. ISO 6579	1.09878.0500	500 g	
MSRV Selective Supplement	1.09874.0010	1 x 10 vials	
ChromoCult <sup>®</sup> RAMBACH <sup>®</sup> agar ref. to ISO 6579 (Kit)	1.00188.0002	4 x 250 ml	
ChromoCult <sup>®</sup> RAMBACH <sup>®</sup> agar ref. to ISO 6579 (Kit)	1.00188.0004	4 x 1000 ml	
GranuCult <sup>®</sup> prime Bismuth sulfite (BS) agar acc. WILSON and BLAIR acc. FDA-BAM	1.00191.0500	500 g	
Triple Sugar Iron Agar	1.03915.0500	500 g	
Urea Agar (Base) acc CHRISTIANSEN acc. ISO 6579, ISO 10273, ISO 19250, ISO 21567	1.08492.0500	500 g	
Urea GR for Analysis ACS, Reagent Ph Eur	1.08487.0500	500 g	
MR-VP (Methyl Red-VOGES-PROSKAUER) Broth	1.05712.0500	500 g	
KOVACS' Indole Reagent	1.09293.0100	100 ml	

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