Technical Data Sheet

GranuCult™
DG 18 (Dichloran Glycerol Chloramphenicol) Agar (Base)

acc. ISO 21527 and FDA-BAM

Ordering number: 1.00465.0500

Recommended for the enumeration of xerophilic moulds in dried and semi-dried foods, such as dried fruits, meat and fish products, spices, confectionery, cereals, nuts.

This culture medium complies with the specifications given by ISO 21527-2 and FDA-BAM.

Mode of Action

By reducing the water activity from approx. 0.99 to 0.95 with 18 % (w/w) glycerol and addition of chloramphenicol growth of bacteria is prevented. The inclusion of dichloran serves to inhibit the rapid spreading of mucoraceous fungi and restricts colony sizes of other genera, easing the colony count. The enzymatic digest of casein is a nitrogen source containing a high level of free amino acids and glucose provides an energy source for the growth whilst agar is the solidifying agent.

Typical Composition

Specified by ISO 21527-2		FDA-BAM M184		GranuCult™ DG18	
Casein Enzymatic Digest	5 g/l	Peptone	5 g/l	Enzymatic Digest of Casein	5 g/l
D-Glucose	10 g/l	Glucose	10 g/l	D(+)-Glucose	10 g/l
KH_2PO_4	1 g/l	KH_2PO_4	1 g/l	KH_2PO_4	1 g/l
$MgSO_4$	0.5 g/l	$MgSO_4$	0.5 g/l	$MgSO_4$	0.5 g/l
Dichloran	0.002 g/l	Dichloran	0.002 g/l	Dichloran	0.002 g/l
Chloramphenicol	0.1 g/l	Chloramphenicol	0.1 g/l	Chloramphenicol	0.1 g/l
Agar	12-15 g/l	Agar	15 g/l	Agar-Agar*	15 g/l
Glycerol, anhydrous	220 g/l	Glycerol	220 g/l	Glycerol, anhydrous	220 g/l
Water	1000 ml/l	Water	800 ml/l	Water	n/a
pH at 25 °C	5.6 ± 0.2	pH at 25 °C	5.6 ± 0.2	pH at 25 °C	5.6 ± 0.2

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

Preparation

Dissolve 31.6 g in 1 l (acc. ISO 21527-2) of purified water. Heat in boiling water and agitate frequently until completely dissolved. Add 175 ml / 220 g of glycerol (article number 104092) to the medium and autoclave 15 min at 121 °C. Cool to 50 °C. Mix well and pour into plates.

The prepared medium is clear to slightly opalescent and yellowish-brown.

According to ISO 21527-2, chlortetracycline (50 mg/l) may be added as a sterile solution to the medium after autoclaving when bacterial overgrowth may be a problem. In order for moulds to exhibit their full morphology trace elements may be added to the medium prior to autoclaving, see ISO 21527-2 for details.

Experimental Procedure and Evaluation

Depend on the purpose for which the medium is used.

Incubate the inoculated plates under aerobic conditions, lids uppermost, e.g. acc. to ISO 21527-2 at 24-26 °C for 5-7 days. If necessary, leave the plates to stand in diffuse daylight for 1-2 days.

If the presence of *Xeromyces bisporus* is suspected, incubate the plates for 10 days.

It is recommended that the plates be incubated in an open plastic bag in order not to contaminate the incubator in the event of dissemination of the moulds out of the dishes.

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.

According to Corry et al. (2012), self-prepared plates can be stored at +2 °C to +8 °C in the dark and protected against evaporation for up to 7 days.

Quality Control

Function	Control strains	Incubation	Reference medium	Method of control	Expected results
Productivity	*Wallemia sebi* ATCC® 42694	5 days at 24-26 °C	Sabouraud Dextrose Agar (SDA)	Quantitative	Recovery ≥ 50 %
	Saccharomyces cerevisiae ATCC® 9763				
	Aspergillus caesiellus (formerly *A. restrictus*) ATCC® 42693				
	Eurotium rubrum ATCC® 42690				
	Mucor racemosus ATCC® 42647		-	Qualitative	Growth good to very good
	Penicillium cyclopium CMI 19759				
	Rhizopus stolonifer ATCC® 15441				

Function	Control strains	Incubation	Reference medium	Method of control	Expected results
Selectivity	*Escherichia coli* ATCC® 8739	5 days at 24-26 °C	Sabouraud Dextrose Agar (SDA)	Qualitative	No Growth
	Escherichia coli ATCC® 25922				
	Bacillus subtilis subsp. *Spizizenii* ATCC® 6633				

Please refer to the actual batch related Certificate of Analysis.

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

A recovery rate of 50 % is equivalent to a productivity value of 0.5.

Literature

Beuchat, L.R. and Hwang, C.-A. (1995): Evaluation of modified dichloran 18% glycerol (DG18) agar for enumerating fungi in wheat flour. Int. J. Food Microbiol. **29**: 161-166.

Corry, J.E.L., Curtis, G.D.W. and Baird, R.M. (2012): Handbook of Culture Media for Food and Water Microbiology, pp. 735-737. Royal Society of Chemistry, Cambridge, UK.

FDA-BAM (2001) Chapter No. 18: Yeasts, Molds and Mycotoxins. U.S. Food and Drug Administration - Bacteriological Analytical Manual

Hocking, A.D. and Pitt, J. I. (1980): Dichloran-glycerol medium for enumeration of Xerophilus fungi from low-moisture foods. Appl Environ. Microbiol. **39**: 488-492.

ISO International Standardisation Organisation. Microbiology of food and animal feeding stuffs - Horizontal method for the enumeration of yeasts and moulds - Part 2: Colony count technique in products with water activity less than or equal to 0,95. ISO 21527-2:2008.

ISO International Standardisation Organisation. Microbiology of food, animal feed and water - Preparation, production, storage and performance testing of culture media. EN ISO 11133:2014.

Ordering Information

Product	Cat. No.	Pack size	Other pack sizes available
GranuCult™ DG 18 Agar ISO 21257	1.00465.0500	500 g	
Glycerol for analysis EMSURE®	1.04092.1000	1000 ml	
ReadyPlate™ DG 18 Agar ISO 21257	1.46161.0020	20 plates	100 plates

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Find contact information for your country at:
www.EMDmillipore.com/offices

For Technical Service, please visit:
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