



# Technical Data

## Dichloran Rose Bengal Chloramphenicol (DRBC) Agar

M1000F

### Intended Use:

Recommended for selective isolation and enumeration of spreader moulds associated with food spoilage in accordance with FDA BAM.

### Composition\*\*

Ingredients	g / L
Peptone	5.000
Dextrose (Glucose)	10.000
Potassium dihydrogen phosphate	1.000
Magnesium sulphate, heptahydrate	0.500
Rose bengal	0.025
Dichloran	0.002
Chloramphenicol	0.100
Agar	15.000
Final pH ( at 25°C)	5.6±0.2

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 31.37 grams (the equivalent weight of dehydrated medium per litre) in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

### Principle And Interpretation

Dichloran Rose Bengal Chloramphenicol Agar is formulated as described by FDA BAM (1) for isolation and enumeration of yeasts in the presence of moulds.

Peptone provides nitrogen, vitamins and minerals. Dextrose is a carbohydrate source. Phosphate buffers the medium. Magnesium sulfate provides divalent cations and sulfate. Dichloran is an antifungal agent, added to the medium to reduce colony diameters of spreading fungi. Rose bengal exhibits an improved inhibitory activity at pH 5.6 and therefore the final pH of the medium is reduced to 5.6 for the inhibition of spreading fungi. The presence of rose bengal in the medium suppresses the growth of bacteria and restricts the size and height of colonies of the more rapidly growing moulds. The concentration of rose bengal is reduced for optimal performance with dichloran. Additionally, rose bengal is taken up by yeast and mould colonies, which allows these colonies to be easily recognized and enumerated. Addition of Chloramphenicol inhibits a wide range of Gram-positive and Gram-negative bacteria which makes the medium selective for fungi

Note: Incubate plates in the dark at 25°C. Do not stack plates higher than 3 and do not invert. Let plates remain undisturbed until counting.

### Type of specimen

Food sample

### Specimen Collection and Handling:

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (1).

### Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

### Limitations

1. The medium should not be exposed to direct light as rose bengal undergoes photo-degradation leading to formation of toxic chemicals for fungi
2. Certain pathogenic fungi may show poor growth on this medium.

Please refer disclaimer Overleaf.

3. Presence of chloramphenicol may inhibit certain pathogenic fungi.
4. Overheating of the medium may result in low productivity and softening of gel.
5. It is recommended to use fresh plates
6. Further biochemical and serological tests must be carried out for complete identification.

## Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

## Quality Control

### Appearance

Light yellow to pink homogeneous free flowing powder

### Gelling

Firm, comparable with 1.5% Agar gel

### Colour and Clarity of prepared medium

Pink coloured, clear to slightly opalescent gel forms in Petri plates

### Reaction

Reaction of 3.14% w/v aqueous solution at 25°C. pH : 5.6±0.2

### pH

5.40-5.80

### Cultural Response

Cultural characteristics observed after an incubation at 25-30°C for upto 6 days.

Organism	Inoculum (CFU)	Growth	Recovery
<i>Candida albicans</i> ATCC 10231 (00054*)	50-100	good-luxuriant	≥50%
<i>Saccharomyces cerevisiae</i> ATCC 9763 (00058*)	50-100	good-luxuriant	≥50%
# <i>Aspergillus brasiliensis</i> ATCC 16404 (00053*)	50-100	good-luxuriant	
<i>Mucor racemosus</i> ATCC 42647 (00181*)	50-100	good-luxuriant	
** <i>Bacillus spizizenii</i> ATCC 6633 (00003*)	≥10 <sup>4</sup>	inhibited	0%
<i>Escherichia coli</i> ATCC 25922 (00013*)	≥10 <sup>4</sup>	inhibited	0%

Key : (\*) - Corresponding WDCM numbers

\*\*Formerly known as *Bacillus subtilis* subsp. *spizizenii* #Formerly known as *Aspergillus niger*

## Storage and Shelf Life

Store the dehydrated powder between 15-25°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Product performance is best if used within stated expiry period.

## Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (2,3).

## Reference

1. Valerie Tournas, Michael E. Stack, Philip B. Mislivec, Herbert A. , Koch and Ruth Bandler, April 2001 (Content current as of 11/07/2022); BAM Chapter 18: Yeasts, Molds and Mycotoxins, Media M183
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2<sup>nd</sup> Edition.
3. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

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**Disclaimer :**

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