

## TMV 359 – ANTIBIOTIC ASSAY MEDIUM NO.2 (BASE AGAR) (VEG.)

### INTENDED USE

For microbiological assay of antibiotics.

### PRODUCT SUMMARY AND EXPLANATION

Antibiotic Veg Assay Medium No. 2 is prepared by incorporating vegetable peptones in place of animal peptones, making the medium BSE, TSE risks free. It can be used for the same purpose of Antibiotic Assay Medium No. 2 and is recommended for use as base agar for microbiological agar diffusion assays for wide variety of antibiotics. Agar diffusion assays can be performed by cylinders, punched-hole or paper disc tests. The equivalent animal based medium is identical numerically with the name assigned by Grove and Randall that is equivalent to the Antibiotic Assay Medium No. B of Indian Pharmacopoeia.

This medium is widely used to prepare the base layer in the microbiological assay of antibiotics such as Bacitracin, Cephalexin, Cephalothin, Cephapirin, Cloxacillin, Dicloxacillin, Methicillin, Nafcillin, Oxacillin, Chloramphenicol, Novobiocin and Penicillin. To perform the antibiotic assay the Antibiotic Veg Assay medium No.2 is used as base agar. This medium should be prepared on the same day as the test. For the cylinder method, a base layer of 21 ml is required. Once the base medium has solidified, Antibiotic assay medium No.1 as seed agar, inoculated with the standardized culture can be overlaid. Even distribution of the layer is important.

### COMPOSITION

Ingredients	Gms / Ltr
Veg. Peptone	6.000
Veg. extract	1.500
Yeast extract	3.000
Agar	15.000

### PRINCIPLE

Veg Peptone, yeast extract and Veg extract provide the nitrogenous, vitamins and mineral requirement for the growth of test organisms. This medium provides solidified substratum for growth of organism and supports the over layering of soft agar.

### INSTRUCTION FOR USE

- Dissolve 25.5 grams in 1000 ml purified/ distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes. Cool to 45-50°C.
- Mix well and pour into sterile Petri plates.

Advice: Recommended for the microbiological assay of Bacitracin, Cephalexin, Cephaloglycin, Cephaloridine, Cephalothin, Cloxacillin, Cycloserine, Dicloxacillin, Methicillin, Nafcillin, Novobiocin, Oxacillin, Penicillin-G, Penicillin, Rifampicin, Spiramycin .

### QUALITY CONTROL SPECIFICATIONS

Appearance of Powder	: Cream to yellow homogeneous free flowing powder.
Appearance of prepared medium	: Amber coloured, clear to slightly opalescent gel forms in Petri plates.
pH (at 25°C)	: 6.6±0.2



## INTERPRETATION

Cultural characteristics observed after incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Basal layer	Incubation Temperature	Incubation Period
<i>Bacillus subtilis</i> subsp. <i>spizizenii</i>	6633	50-100	Luxuriant	>=70%	Spiramycin	35-37°C	18-48 Hours
<i>Micrococcus luteus</i>	10240	50-100	Luxuriant	>=70%	Bacitracin	35-37°C	18-48 Hours
<i>Staphylococcus aureus</i>	9144	50-100	Luxuriant	>=70%	Tylosin	35-37°C	18-48 Hours
<i>Staphylococcus aureus</i>	29737	50-100	Luxuriant	>=70%	Amikacin, Cephalothin, Cephapirin, Chlortetracycline, Nafcillin, Oxytetracycline, Rofloxacillin	35-37°C	18-48 Hours
<i>Staphylococcus epidermidis</i>	12228	50-100	Good-luxuriant	>=50%	Tetracycline	35-37°C	18-48 Hours
<i>Klebsiella pneumoniae</i>	10031	50-100	Luxuriant	>=70%	Capreomycin, Streptomycin, Troleandomycin, Gramicidin, Thiostrepton, Tobramycin	35-37°C	18-48 Hours
<i>Enterococcus hirae</i>	10541	50-100	Luxuriant	>=70%	-	35-37°C	18-48 Hours
<i>Escherichia coli</i>	10536	50-100	Luxuriant	>=70%	Chloramphenicol, Spectinomycin	35-37°C	18-48 Hours

## PACKAGING:

In pack size of 500 gm bottles.

## STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

**Product Deterioration:** Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

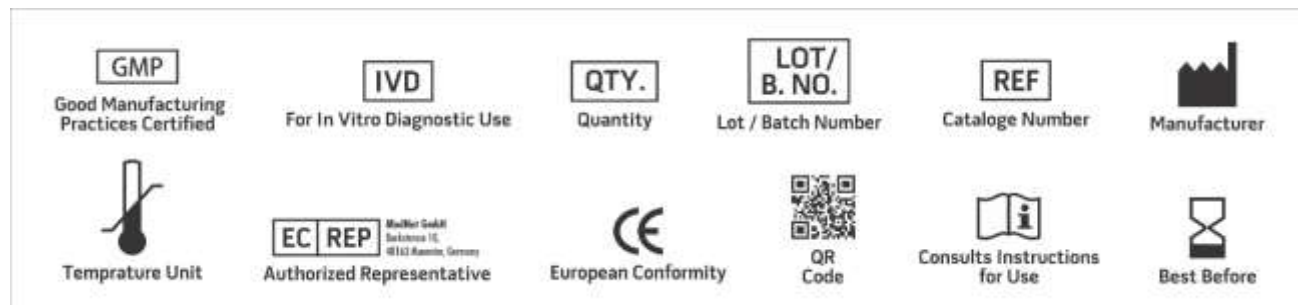
## DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.



## REFERENCES

1. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopedia, Inc. New York.
2. Indian Pharmacopoeia 2010, Ministry of Health and Family Welfare, Govt. of India, Delhi.



**NOTE:** Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

**\*For Lab Use Only**  
Revision: 08 Nov., 2019