

## MEDIA AND ITS COMPOSITION

### The composition of required medium for Quality Control Parameters

#### Soyabean Casein Digest Medium (Himedia-M011)

<b>Composition</b> (Ingredients g/litre)	- Pancreatic digest of casein 17.0 Peptic digest of soyabean meal 3.0 Sodium chloride 5.0 Dextrose (Glucose) 2.5 Dipotassium hydrogen phosphate 2.5 Final pH 7.3 +/- 0.2 at 25°C
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The medium (30g) was suspended in 1000ml distilled water containing 10ml glycerol in order get above composition. The mixture was boiled until a clear solution was obtained. The solution was sterilized by autoclaving at 15lbs pressure (121°C) for 15min.

#### Soyabean Casein Digest Agar (Himedia-M290)

<b>Composition</b> (Ingredients g/litre)	- Pancreatic digest of casein 15.0 Peptic digest of soyabean meal 5.0 Sodium chloride 5.0 Agar – 15.0 Final pH 7.3 +/- 0.2 at 37°C
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The medium (40g) was suspended in 1000ml distilled water containing 10ml glycerol in order get above composition. The mixture was boiled until a clear solution was obtained. The solution was sterilized by autoclaving at 15lbs pressure (121°C) for 15min.

#### Sabouraud Dextrose Agar (Himedia-M063)

<b>Composition</b> (Ingredients g / litre)	- Dextrose 40.0 Mycological peptone 10.0 Agar – 15.0 Final pH (at 25°C) 5.6±0.2
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The medium (65g) was suspended in 1000ml distilled water containing 10ml glycerol in order get above composition. The mixture was boiled until a clear solution was obtained. The solution was sterilized by autoclaving at 15lbs pressure (121°C) for 15min.

**MacConkey Agar w/o CV w/ 0.15% Bile Salts (HimediaM008)**

<b>Composition</b> (Ingredients g / litre)	- Peptic digest of animal tissue 17.000
	Proteose peptone 3.000
	Lactose 10.000
	Bile salts 1.500
	Sodium chloride 5.000
	Neutral red 0.030
	Agar 15.000
	Final pH (at 25°C) 7.1±0.2

The medium (51.53g) was suspended in 1000ml distilled water containing 10ml glycerol in order get above composition. The mixture was boiled until a clear solution was obtained. The solution was sterilized by autoclaving at 15lbs pressure (121°C) for 15min.

**Eosin Methylene Blue Agar, (Himedia M022)**

<b>Composition</b> (Ingredients g / litre)	- Peptic digest of animal tissue 10.000
	Dipotassium phosphate 2.000
	Lactose 10.000
	Eosin - Y 0.400
	Methylene blue 0.065
	Agar 15.000
	Final pH (at 25°C) 7.1±0.2

The medium (37.46g) was suspended in 1000ml distilled water containing 10ml glycerol in order get above composition. The mixture was boiled until a clear solution was obtained. The solution was sterilized by autoclaving at 15lbs pressure (121°C) for 15min.

**Brilliant Green Agar (Modified)****Composition** (Ingredients g / litre)

- Proteose peptone 10.0
- Yeast extract 3.0
- Lactose 10.0
- Sucrose 10.0
- Sodium chloride 5.0
- Phenol red 0.08
- Brilliant green 0.0125
- Agar 20.0
- Final pH (at 25°C) 6.9±0.2

The medium (29g) was suspended in 1000ml distilled water containing 10ml glycerol in order get above composition. The mixture was boiled until a clear solution was obtained. The solution was sterilized by autoclaving at 15lbs pressure (121°C) for 15min.

**Cetrimide Agar Base (Himedia M024)****Composition** (Ingredients g / litre)

- Pancreatic digest of gelatin 20.0
- Magnesium chloride 1.4
- Potassium sulphate 10.0
- Cetrimide 0.3
- Agar 15.0
- Final pH (at 25°C) 7.2±0.2

The medium (46.7g) was suspended in 1000ml distilled water containing 10ml glycerol in order get above composition. The mixture was boiled until a clear solution was obtained. The solution was sterilized by autoclaving at 15lbs pressure (121°C) for 15min.

**Baird-Parker Agar Base (Himedia CM0275)****Composition** (Ingredients g / litre)

- Tryptone 10.0
- Lab-Lemco powder 5.0
- Yeast extract 1.0
- Sodium pyruvate 10.0

Glycine 12.0  
Lithium chloride 5.0  
Agar 20.0  
Final pH (at 25°C) 6.8±0.2

The medium (63g) was suspended in one litre of distilled water in order to get above composition. The mixture was boiled until a clear solution was obtained. The solution was sterilised by autoclaving at 121°C for 15min. It was cooled to 50°C and 50ml of Egg Yolk Tellurite Emulsion (SR0054) and 3ml of potassium tellurite 3.5% (SR0030) were aseptically added.

### **The composition of required medium for Antimicrobial Studies**

#### **Nutrient Agar (Himedia - M001)**

**Composition** (Ingredients g/litre) - Peptic digest of animal tissue 5.0  
Sodium chloride 5.0  
Beef extract 1.5  
Yeast extract 1.5  
Agar 15.0  
Final pH (at 25°C) 7.4±0.2

The medium (28g) was suspended in 1000ml distilled water containing in order to get the above composition. The mixture was boiled until a clear solution was obtained. The solution was sterilized by autoclaving at 15lbs pressure (121°C) for 15min.

#### **Mueller Hinton Agar (M-H Agar) Sigma-70191**

**Composition** (Ingredients g/litre) - Beef infusion solids 4.0  
Starch 1.5  
Casein hydrolysate 17.5  
Agar 15.0  
Final pH 7.4 +/- 0.2 at 37°C

The required quantity (38g) was suspended in 1000ml of distilled water in order to get the above composition. The mixture was boiled until a clear solution was obtained. The solution was sterilized by autoclaving at 121°C for 15min.



## VITAMINS

Choline chloride 1.000  
D-Ca-Pantothenate 1.000  
Folic acid 1.000  
Nicotinamide 1.000  
Pyridoxal 1.000  
Riboflavin 0.100  
Thiamine hydrochloride 1.000  
i-Inositol 2.000

## OTHERS

D-Glucose 1000.000  
Phenol red sodium salt 11.000  
Sodium pyruvate 110.000

Suspend 9.8gms in 900ml tissue culture grade water with constant gentle stirring until the powder is completely dissolved. Add 2.2gms of sodium bicarbonate powder (TC230) or 29.3ml of 7.5% sodium bicarbonate solution (TCL013) for 1 litre of medium and stir until dissolved. Adjust the pH to 0.2 - 0.3 pH units below the desired pH using 1N HCl or 1N NaOH since the pH tends to rise during filtration. Make up the final volume to 1000ml with tissue culture grade water. Sterilize the medium immediately by filtering through a sterile membrane filter with a porosity of 0.22 micron or less, using positive pressure rather than vacuum to minimize the loss of carbon dioxide.

Aseptically add sterile supplements as required and dispense the desired amount of sterile medium into sterile containers. Store the liquid medium at 2-8°C and in dark till use.

### **Trypsin-EDTA Solution 1X (Himedia – TCL033)**

0.05% Trypsin,

0.02% EDTA in Hank's Balanced Salt Solution With Phenol red Product