

## COMPOSITION OF MEDIA AND REAGENTS

### SIMMONS CITRATE AGAR

### HI-MEDIA

<b>Ingredients</b>	<b>Quantity(gm/lit)</b>
Magnesium sulphate hepta hydrate	0.2g
Ammonim di hydrogen phosphate	1g
Di cregpotassium hydrogen phosphate	1g
Trisodium citrate dehydrate	2g
NaCl	5g
Bromothymol blue	0.08g
Agar	15g
Distill water	1000ml
pH 6.8±2	

### CLED AGAR

### HI-MEDIA

<b>Ingredients</b>	<b>Quantity(gm/lit)</b>
Peptic digest of animal tissue	4g
Casein enzymichydrosylate	4g
Beef extract	3g
Lactose	10g
L- cystine	0.128g
Bromothymol blue	0.02g
Agar	15g
pH(at 25°C) 7.3±0.2	

**NUTRIENT AGAR**

**HI-MEDIA**

**Ingredients**

**Quantity(gm/lit)**

Peptic digest of animal tissue

5g

Beef extract

1g

Yeast extract

2g

Sodium chloride

5g

Agar

15g

pH (at 25°C) 7.4±0.2

**PEPTONE**

**HI-MEDIA**

**Ingredients**

**Quantity(gm/lit)**

Peptic digest of animal tissue

1gm

NaCl

0.5gm

**TRIPLE SUGAR IRON AGAR**

**HI-MEDIA**

**Ingredients**

**Quantity(gm/lit)**

Casein enzymichydrolysate

15.0 g

Peptic digest of animal tissue

5.0 g

Meat extract

3g

Yeast extract

3g

NaCl

5g

Lactose monohydrate

10g

Sucrose

10g

Glucose anhydrous	1g
Ferric ammonium citrate anhydrous	0.5g
Sodium thiosulphate anhydrous	0.5g
Phenol red	0.024g
Agar agar	12g
Distill water	1000ml
Ph 7.2±0.2	

**NUTRIENT AGAR**

**HIMEDIA**

**Ingredients**

**Quantity(gm/lit)**

Peptic digest of animal tissue	5g
Beef extract	1.5g
Yeast extract	1.5g
NaCl	5g
Agar agar	15g
Distill water	1000ml
Ph 7.4±0.2	

**MUELLER HINTON AGAR**

**HI-MEDIA**

Meat infusion powder	2g
Casamino acids	17.5g
Starch soluble	5g
Agar agar	17g

Distill water 1000ml

pH 7.3±.2

**MAC CONKEY AGAR**

**HI-MEDIA**

Peptic digest of animal tissue

1.5g

Casein enzymichydrolysate

1.5g

Pancreatic digest of gelatin

17g

NaCl

5g

Lactose monohydrate

10g

Bile salts

1.5g

Neutral red

0.03g

Crystal violet

0.001g

Agar agar

13.5g

Distill water

1000ml

pH 7.1±.2

**NUTRIENT BROTH**

**HI-MEDIA**

Peptic digest of animal tissue

10g

NaCl

5g

Beef extract

10g

Distill water

1000ml

pH 7.3±1

**TRYPTONE**

**HI-MEDIA**

Pancreatic digest of casein

**Glycerol (C<sub>3</sub>H<sub>8</sub>O<sub>3</sub>)**

**HI-MEDIA**

**Acetic acid glacial extrapure (C<sub>2</sub>H<sub>4</sub>O<sub>2</sub>)**

**HI-MEDIA**

**Urea agar base(christensen)**

**HI-MEDIA**

**Gram stain**

**HI-MEDIA**

**Crystal violet**

**Solution A**

**Ingredients**

**Quantity(gm/lit)**

Crystal violet (90% dye content)

2g

Ethyl alcohol (95%)

20ml

**Solution B**

Ammonium oxalate

0.8g

Distilled water

80ml

**Gram's iodine**

**Ingredients**

**Quantity(gm/lit)**

Iodine

1g

Potassium iodide

2g

Distilled water

80ml

**Ethyl alcohol (95%)**

Ethyl alcohol (100%)	95ml
Distilled water	5ml

**Saffranine**

Saffranine (25% solution in 95% ethyl alcohol)	10ml
Distilled water	100ml

**ANTIBIOTICS USED**

**HI-MEDIA ( $\mu\text{g}/\text{disc}$ )**

• Nalidixic acid	30
• Norfloxacin	10
• Ciprofloxacin	5
• Ofloxacin	5
• Levofloxacin	5
• Sparfloxacin	5
• Lomefloxacin	5
• Gatifloxacin	5
• Gemifloxacin	5
• Ampicillin	10
• Co-trimoxazole	25
• Tigecycline	15
• Gentamicin	120
• Amikacin	30
• Imipenem	10
• Ceftazidime	30
• Cefotaxime	30
• Ceftriaxone	30
• Polymixin B	300U

**SOC medium preparation:**

<b>Ingredients</b>	<b>Quantity(gm/lit)</b>
2% w/v tryptone	20 g
0.5% w/v yeast extract	5g
10mM NaCl	0.584g
2.5mM KCl	0.186g
10mM MgCl <sub>2</sub>	0.952g
10mM MgSO <sub>4</sub>	1.204g
0.4% glucose	0.4g
Distilled water	1000ml
<b>Tris borate EDTA buffer</b>	
<b>Agarose</b>	<b>HI-MEDIA</b>
<b>Nuclease free water</b>	<b>HI-MEDIA</b>
<b>0.1M CaCl<sub>2</sub></b>	
<b>DNA ladder</b>	<b>MERCK</b>
<b>Plasmid mini prep kit</b>	<b>QIAGEN</b>