

## MCDB 201 Medium

With Trace elements, L- Glutamine and 30mM HEPES buffer  
Without Sodium bicarbonate

Product Code: AT136

### Product Description :

MCDB media were developed for the culture of specific cell types without a serum supplement. The media were supplemented with growth factors, hormones, trace elements, or low levels of dialyzed fetal bovine serum protein (FBSP). Each MCDB medium was formulated for a specific cell type. MCDB 105 and 110 were formulated for rapid clonal growth of normal human diploid cells. MCDB 131 medium was originally developed for the clonal growth of human micro-vascular endothelial cells (HMVEC). MCDB 151, 201 and 302 were originally developed for human keratinocytes, clonal growth of chick embryo fibroblasts and CHO cells.

AT136 is MCDB 201 with trace elements, L-glutamine and 30mM HEPES buffer. HEPES, a zwitterionic buffer having a pKa of 7.3 at 37°C prevents the initial rise in pH that tends to occur at the initiation of a culture and increases the buffering capacity of the medium. Users are advised to review the literature for recommendations regarding medium supplementation and physiological growth requirements specific for different cell lines.

### Composition :

Ingredients	mg/L
<b>INORGANIC SALTS</b>	
Ammonium metavanadate	0.000006
Calcium chloride dihydrate	294.000
Cupric sulphate pentahydrate	0.00025
Disodium hydrogen phosphate anhydrous	70.990
Ferrous sulphate heptahydrate	1.668
Magnesium sulphate anhydrous	180.570
Manganese sulphate	0.000075
Molybdic acid ammonium tetrahydrate	0.000618
Nickel chloride hexahydrate	0.000012
Potassium chloride	223.650
Sodium chloride	7597.000
Sodium metasilicate nonahydrate	0.142
Sodium selenite	0.000865
Zinc sulphate heptahydrate	0.028744
<b>AMINO ACIDS</b>	

Glycine	7.510
L-Alanine	8.910
L-Arginine hydrochloride	63.200
L-Asparagine monohydrate	150.000
L-Aspartic acid	13.310
L-Cysteine hydrochloride monohydrate	35.130
L-Glutamic acid	14.710
L-Glutamine	146.150
L-Histidine hydrochloride monohydrate	20.970
L-Isoleucine	13.120
L-Leucine	39.350
L-Lysine hydrochloride	36.540
L-Methionine	4.480
L-Phenylalanine	4.960
L-Proline	5.760
L-Serine	31.530
L-Threonine	35.750
L-Tryptophan	6.130
L-Tyrosine sodium salt	11.350
L-Valine	35.130
<b>VITAMINS</b>	
Choline chloride	13.960
D-Biotin	0.00733
D-Ca-Pantothenate	0.477
Folinic acid (Calcium)	0.00512
Niacinamide	6.110
Pyridoxine hydrochloride	0.0617
Riboflavin	0.113
Thiamine hydrochloride	0.337
Vitamin B12	0.136
myo-Inositol	18.020
<b>OTHERS</b>	
Adenine hydrochloride	1.720
D-Glucose	1441.000
HEPES buffer	7149.000
Linoleic acid	0.0841
Phenol red sodium salt	1.242
Putrescine dihydrochloride	0.00161
Sodium pyruvate	55.000
Thioctic acid	0.00206
Thymidine	0.0727

## Directions :

1. Suspend 17.7gms in 900ml tissue culture grade water with constant, gentle stirring until the powder is completely dissolved. Do not heat the water.
2. 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.
3. Make up the final volume to 1000ml with tissue culture grade water.
4. 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.
5. Aseptically add sterile supplements as required and dispense the desired amount of sterile medium into sterile containers.
6. Store liquid medium at 2-8°C and in dark till use.

## Material required but not provided :

Tissue culture grade water (TCL010)  
1N Hydrochloric acid (TCL003)  
1N Sodium hydroxide (TCL002)  
Foetal bovine serum (RM1112/RM10432)

## Quality Control:

### Appearance

Off-white to Creamish white, homogenous powder.

### Solubility

Clear solution at 17.7 gms/L.

### pH without Sodium Bicarbonate

5.10 -5.70

### Osmolality without Sodium Bicarbonate

280.00 -320.00

### Cultural Response

The growth promotion capacity of the medium is assessed qualitatively by analyzing the cells for the morphology and quantitatively by estimating the cell counts and comparing it with a control medium through minimum three subcultures.

### Endotoxin Content

NMT 5EU/ml

## Storage and Shelf Life:

1. All the powdered media and prepared liquid culture media should be stored at 2-8°C. Use before the expiry date. In spite of above recommended storage condition, certain powdered medium may show some signs of deterioration /degradation in certain instances. This can be indicated by change in colour, change in appearance and presence of particulate matter and haziness after dissolution.
2. Preparation of concentrated medium is not recommended since free base amino acids and salt complexes having low solubility may precipitate in concentrated medium.
3. pH and sodium bicarbonate concentration of the prepared medium are critical factors affecting cell growth. This is also influenced by amount of medium and volume of culture vessel used (surface to volume ratio). For example, in large bottles, such as Roux bottles pH tends to rise perceptibly as significant volume of carbon dioxide is released. Therefore, optimal conditions of pH, sodium bicarbonate concentration, surface to volume ratio must be determined for each cell type. We recommend stringent monitoring of pH. If needed, pH can be adjusted by using sterile 1N HCl or 1N NaOH or by bubbling in carbon dioxide.
4. If required, supplements can be added to the medium prior to or after filter sterilization observing sterility precautions. Shelf life of the medium will depend on the nature of supplement added to the medium.

Revision : 1 / 2011

## Disclaimer :

User must ensure suitability of the product(s) in their application prior to use. Products conform solely to the information contained in this and other related HiMedia™ publications. The information contained in this publication is based on our research and development work and is to the best of our knowledge true and accurate. HiMedia™ Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are not intended for human or animal diagnostic or therapeutic use but for laboratory, research or further manufacturing use only, unless otherwise specified. Statements contained herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.