

### Technical Information

#### MCDB 131 Medium With Trace elements and L- Glutamine Without Sodium bicarbonate

#### Product Code: AT1133

**Application:-** 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.

AT1133 is MCDB 131 with trace elements and L- glutamine. Users are advised to review the literature for recommendations regarding medium supplementation and physiological growth requirements specific for different cell lines.

#### Composition\*\*

Ingredients	mg/Litre
<b>INORGANIC SALTS</b>	
Ammonium metavanadate	0.0006
Calcium chloride dihydrate	235.200
Cupric sulphate pentahydrate	0.0012
Disodium hydrogen phosphate anhydrous	71.000
Ferrous sulphate heptahydrate	0.278
Magnesium sulphate anhydrous	1204.00
Manganese sulphate	0.0002
Molybdic acid tetrahydrate (ammonium)	0.0037
Nickel chloride hexahydrate	0.0001
Potassium Chloride	298.200
Sodium bicarbonate	1180.000
Sodium chloride	6428.400
Sodium metasilicate nonahydrate	2.842
Sodium selenite	0.0052
Zinc sulphate heptahydrate	0.0003
<b>AMINO ACIDS</b>	
Glycine	2.250
L-Alanine	2.670
L-Arginine hydrochloride	63.210
L-Asparagine monohydrate	15.000
L-Aspartic acid	13.310
L-Cystine hydrochloride monohydrate	35.120
L-Glutamic acid	44.130
L-Glutamine	1461.000
L-Histidine hydrochloride monohydrate	41.920
L-Isoleucine	65.600
L-Leucine	131.200

L-Lysine hydrochloride	182.600
L-Methionine	14.920
L-Phenylalanine	33.040
L-Proline	11.510
L-Serine	31.530
L-Threonine	11.910
L-Tryptophan	4.080
L-Tyrosine disodium salt dihydrate	22.520
L-Valine	117.100
<b>VITAMINS</b>	
Choline chloride	13.960
D-Biotin	0.0073
D-Ca-Pantothenate	11.915
Folinic acid (Calcium)	0.5115
Nicotinamide	6.105
Pyridoxine hydrochloride	2.056
Riboflavin	0.0038
Thiamine hydrochloride	3.373
Vitamin B12	0.0136
myo-Inositol	7.208
<b>OTHERS</b>	
Adenine hydrochloride	0.1716
D-Glucose	1000.000
Phenol red sodium salt	12.420
Putrescine dihydrochloride	0.0002
Sodium pyruvate	110.000
Thioctic acid	0.0021
Thymidine	0.0242

## Methodology

1. Suspend 11.7gms in 900 ml tissue culture grade water with constant, gentle stirring until the powder is completely dissolved. Do not heat the water.
2. Add 1.18gms of sodium bicarbonate powder (TC1230) or 15.73ml of 7.5% sodium bicarbonate solution (TCL1013) for 1 litre of medium and stir until dissolved.
3. 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.
4. Make up the final volume to 1000ml with tissue culture grade water.
5. 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.
6. Aseptically add sterile supplements as required and dispense the desired amount of sterile medium into sterile containers.
7. Store liquid medium at 2-8°C and in dark till use

## Material Required But Not Provided

Tissue culture grade water (TCL1010)  
 Sodium bicarbonate powder (TC1230)  
 Sodium bicarbonate solution, 7.5% (TCL1013)  
 1N Hydrochloric acid (TCL1003)  
 1N Sodium hydroxide (TCL1002)  
 Foetal bovine serum (BA3112/BA12432)

### Quality control

#### Appearance

Off-white to Creamish white, homogenous powder.

#### Solubility

Clear solution at 11.7 gms/L.

#### pH without Sodium Bicarbonate

5.70 -6.30

#### pH with Sodium Bicarbonate

6.90 -7.50

#### Osmolality without Sodium Bicarbonate

235.00 -275.00

#### Osmolality with Sodium Bicarbonate

260.00 -300.00

#### 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 1EU/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.

### Disclaimer :

- User must ensure suitability of the product(s) in their application prior to use.
- The product conforms solely to the technical information provided in this booklet and to the best of knowledge research and development work carried at **CDH** is true and accurate.
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