CDH

Product Specification

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Technical Information

Iscove's Modified Dulbecco's Medium (IMDM)

With L-Glutamine and 25mM HEPES Buffer
Without Sodium bicarbonate

Product Code: AT1070

Application: - Iscove's Modified Dulbecco's Medium is an enriched modification of Dulbecco's Modified Eagle's Medium where in serum can be partially or totally replaced by chemically defined substances. The medium contains additional amino acids, sodium selenite, sodium pyruvate, vitamins and inorganic salts. Potassium nitrate is substituted by ferric nitrate. IMDM was the first medium utilizing HEPES buffer. The medium when appropriately supplemented supports good growth of precursor cells of erythrocytes and macrophages. The medium also supports good growth of T and B- lymphocytes and a variety of hybrid cells under serum free or reduced serum conditions.

AT1070 is Iscove's Modified Dulbecco's Medium with L-glutamine and 25mM HEPES buffer. HEPES, a zwitterionic buffer having a pKa of 7.3 at 37ºC 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 / Litre
INORGANIC SALTS	
Calcium chloride dihydrate	219.000
Magnesium sulphate anhydrous	97.720
Potassium chloride	330.000
Potassium nitrate	0.076
Sodium chloride	4505.000
Sodium dihydrogen phosphate, anhydrous	109.000
Sodium selenite	0.0173
AMINO ACIDS	
Glycine	30.000
L-Alanine	25.000
L-Arginine hydrochloride	84.000
L-Asparagine	25.000
L-Aspartic acid	30.000
L-Cysteine dihydrochloride	91.240
L-Glutamic acid	75.000
L-Glutamine	584.000
L-Histidine hydrochloride monohydrate	42.000
L-Isoleucine	104.800
L-Leucine	104.800
L-Lysine hydrochloride	146.200
L-Methionine	30.000
L-Phenylalanine	66.000
L-Prolin	40.000
L-Serine	42.000
L-Threonine	95.200
L-Tryptophan	16.000

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L-Tyrosine Disodium Salt	104.200
L-Valine	93.600
VITAMINS	
Choline chloride	4.000
D-Biotin	0.013
D-Ca-Pantothenate	4.000
Folic acid	4.000
Nicotinamide	4.000
Pyridoxal hydrochloride	4.000
Riboflavin	0.400
Thiamine hydrochloride	4.000
i-Inositol	7.200
OTHERS	
D-Glucose	4500.000
HEPES Buffer	5958.000
Phenol red Sodium Salt	15.000
Sodium Pyruvate	110.00

Methodology

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. Add 3.024gms of Sodium bicarbonate powder (TC1230) or 40.32ml 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 (TC1230)
Sodium bicarbonate solution, 7.5% (TCL1013)
1N Hydrochloric acid (TCL1003)
1N Sodium hydroxide (TCL1002)
Foetal bovine serum (BA3112/BA30432)

Quality Control

Appearance
Off-white to Creamish white, homogenous powder.

Solubility
Clear solution at 17.7gms/L

pH without Sodium Bicarbonate
5.20-5.80

pH with Sodium Bicarbonate
6.70-7.30

Osmolality without Sodium Bicarbonate
210.00-250.00

Osmolality with 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 2-8ºC. 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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- Do not use the products if it fails to meet specificatons for identity and performens parameters.