CDH

Product Specification

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Technical Information

Dulbecco's Modified Eagle Medium (DMEM)
(Modified for Autoclaving)
With 1gm Glucose per litre and Sodium pyruvate
Without L-Glutamine and Sodium bicarbonate

Product Code:AT1065A

Application: Dulbecco's Modified Eagle Medium is one of the most widely used modification of Eagle's medium. DMEM is a modification of Basal Medium Eagle (BME) that contains four fold concentration of amino acids and vitamins. Additionally, the formulation also includes glycine, serine and ferric nitrate. The original formulation contains 1000mg glucose per litre and was originally used to culture embryonic mouse cells. AT1065A is DMEM low glucose and is modified for autoclaving. Autoclavable medium offers a convenient alternative to membrane sterilized liquid medium and is modified to include heat stable components to ensure that product efficacy is maintained after autoclaving. L-glutamine is heat labile, hence has been omitted from the formulation. 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	265.000
Ferric nitrate nonahydrate	0.100
Magnesium sulphate anhydrous	97.720
Potassium chloride	400.000
Sodium chloride	6400.000
Sodium dihydrogen phosphate anhydrous	109.000
AMINO ACIDS	
Glycine	30.000
L-Arginine hydrochloride	84.000
L-Cysteine dihydrochloride	62.570
L-Histidine hydrochloride monohydrate	42.000
L-Isoleucine	105.000
L-Leucine	105.000
L-Lysine hydrochloride	146.000
L-Methionine	30.000
L-Phenylalanine	66.000
L-Serine	42.000
L-Threonine	95.000
L-Tryptophan	16.000
L-Tyrosine Disodium Salt	103.790
L-Valine	94.000
VITAMINS	
Choline chloride	4.000

D-Ca-Pantothenate	4.000
Folic acid	4.000
Nicotinamide	4.000
Pyridoxal hydrochloride	4.000
Riboflavin	0.400
Thiamine	4.000
i-Inositol	7.200
OTHERS	
D-Glucose	1000.00
Phenol red Sodium Salt	9.000
Sodium pyruvate	110.000
Sodium succinate	100.000
Succinic acid	75.000

Methodology

1. Suspend 9.6gms in 900ml tissue culture grade water with constant, gentle stirring until the powder is completely dissolved.
2. Adjust the pH to 4.0 before autoclaving
3. Make up the volume with tissue culture grade water. Subtract the volumes of 7.5% Sodium bicarbonate solution. and 200mM L-glutamine solution from the final volume.
4. Autoclave medium at 121°C at 15psi for 15minutes.
5. Remove the medium promptly from the autoclave to avoid extended heating or evaporation.
6. Allow to cool at room temperature.
7. Add 49.3ml of 7.5% Sodium bicarbonate solution (TCL1013) and 20ml of 200mM L- glutamine solution (TCL1012) to the final volume of the medium being prepared.
8. If necessary, adjust the pH using sterile 1N NaOH (TCL1002) or 1N HCl (TCL1003).
9. Store liquid medium at 2-8°C and in dark till use.

Material required but not provided

Tissue culture grade water (TCL1010)
Sodium bicarbonate solution, 7.5% (TCL1013)
1N Hydrochloric acid (TCL1003)
1N Sodium hydroxide (TCL1002)
200mM L-Glutamine solution (TCL1012)
Foetal bovine serum (BA3112/BA30432)

Quality Control

Appearance
Off-white to Creamish white, homogenous powder.

Solubility
Clear solution at 9.6gms/L

pH without Sodium Bicarbonate
4.50-5.10

pH with Sodium Bicarbonate
7.60-8.20

Osmolality without Sodium Bicarbonate
230.00-270.00

Osmolality with Sodium Bicarbonate

290.00-330.00

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.