

Technical Information

Basal Medium Eagle (BME) (Modified for Autoclaving) With Earle's salts Without L-Glutamine and Sodium bicarbonate

Product Code : AT1004A

Application:- Basal Medium developed by Harry Eagle is a combination of essential nutrients in appropriate concentrations for monolayer cultivation of a wide variety of normal and transformed cells. The medium was initially developed as a result of studies to determine the nutritional requirements of HeLa cells and mouse fibroblast L cells in culture. Although there are many versions of Basal Medium described by Eagle, the name Basal Medium Eagle applies to only the formulation developed for HeLa cells. Basal Medium Eagle when properly supplemented supports growth of variety of diploid or primary mammalian cell cultures. Modifications to the original BME have resulted in other media, including MEM and DMEM.

AT1004A is Basal Medium Eagle with Earle's balanced salts and is modified for autoclaving. Autoclavable media offer a convenient alternative to membrane sterilized liquid medium. It 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	Gms / Litre
INORGANIC SALTS	
Calcium chloride dihydrate	265.000
Magnesium sulphate anhydrous	97.720
Potassium chloride	400.000
Sodium chloride	6800.000
Sodium dihydrogen phosphate anhydrous	122.000
AMINO ACIDS	
L-Arginine hydrochloride	21.100
L-Cystine dihydrochloride	15.650
L-Histidine hydrochloride	10.500
L-Isoleucine	26.200
L-Leucine	26.200
L-Lysine hydrochloride	36.480
L-Methionine	7.500
L-Phenylalanine	16.500
L-Threonine	23.800
L-Tryptophan	4.000
L-Tyrosine disodium salt	25.950
L-Valine	23.400
VITAMINS	
Choline bitartrate	1.800
D-Biotin	1.000
D-Ca-Pantothenate	1.000
Folic acid	1.000
Nicotinamide	1.000

Pyridoxal hydrochloride	1.000
Riboflavin	0.100
Thiamine hydrochloride	1.000
i-Inositol	1.800
OTHERS	
D-Glucose	1000.000
Phenol red sodium salt	6.360
Sodium succinate	100.00
Succinic acid	75.000

Methodology

1. Suspend 9.1gms 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 4.0 before autoclaving..
3. Make up the volume to 960ml (This volume is derived after subtracting 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 15 minutes
5. Remove the medium promptly from the autoclave to avoid extended heating or evaporation .
6. Allow to cool at room temperature
7. Add 29.3ml of 7.5% sodium bicarbonate solution (TCL1013) and 10 ml 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 powder (TC1230)
 Sodium bicarbonate solution, 7.5% (TCL1013)
 L-Glutamine (TC1243)
 200 mM L-Glutamine solution (TCL1012)
 1N Hydrochloric acid (TCL1003)
 1N Sodium hydroxide (TCL1002)
 Fetal bovine serum (BA3112/BA30432)

Quality Control

Appearance

Off-white to Creamish white, homogenous powder.

Solubility

Clear solution at 9.1gms/L

pH without Sodium Bicarbonate

4.30-4.90

pH with Sodium Bicarbonate

7.20 -7.80

Osmolality without Sodium Bicarbonate

230.00 -270.00

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 2-8°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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