

Glasgow's Minimum Essential Medium (GMEM)

With L-Glutamine

Without Tryptose phosphate broth and Sodium bicarbonate

Product Code: AT102

Product Description :

Glasgow's Minimum Essential Medium is a modification of Basal medium Eagle (BME). Ian Macpherson and Michael Stoker added tryptose phosphate broth and twice the concentration of amino acids and vitamins to BME. The medium was originally used to culture BHK-21 clone13 cells, used for investigating the genetic factors affecting cell competence.

AT102 is Glasgow's Minimum Essential Medium with L-glutamine. It does not contain tryptose phosphate broth. 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
INORGANIC SALTS	
Calcium chloride dihydrate	265.000
Ferric nitrate nonhydrate	0.100
Magnesium sulphate anhydrous	97.720
Potassium chloride	400.000
Sodium chloride	6400.000
Sodium dihydrogen phosphate anhydrous	109.000
AMINO ACIDS	
L-Arginine hydrochloride	42.000
L-Cystine	24.000
L-Glutamine	292.000
L-Histidine hydrochloride	21.000
L-Isoleucine	52.400
L-Leucine	52.400
L-Lysine hydrochloride	73.100
L-Methionine	15.000
L-Phenylalanine	33.000
L-Threonine	47.600
L-Tryptophan	8.000
L-Tyrosine Disodium salt	52.000
L-Valine	46.800
VITAMINS	
Choline chloride	2.000

D-Ca-Pantothenate	2.000
Folic acid	2.000
Nicotinamide	2.000
Pyridoxal hydrochloride	2.000
Riboflavin	0.200
Thiamine hydrochloride	2.000
i-Inositol	3.600
OTHERS	
D-Glucose	4500.000
Phenol red Sodium Salt	15.000

Directions :

- Suspend 12.6gms in 900ml tissue culture grade water with constant, gentle stirring until the powder is completely dissolved. Do not heat the water.
- Add 2.75gms of sodium bicarbonate powder (TC230) or 36.7ml of 7.5% sodium bicarbonate solution (TCL013) for 1 litre of medium and stir until dissolved.
- 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.
- Make up the final volume to 1000ml with tissue culture grade water.
- 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.
- Aseptically add sterile supplements as required and dispense the desired amount of sterile medium into sterile containers.
- Store liquid medium at 2-8°C and in dark till use.

Material required but not provided :

Tissue culture grade water (TCL010)
Sodium bicarbonate (TC230)
Sodium bicarbonate solution, 7.5% (TCL013)
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 12.6 gms/L.

pH without Sodium Bicarbonate

4.10 -4.70

pH with Sodium Bicarbonate

7.10 -7.70

Osmolality without Sodium Bicarbonate

210.00 -250.00

Osmolality with Sodium Bicarbonate

265.00 -305.00

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. on cell lines is carried out through two subcultures.

Endotoxin content

NMT 5EU/ml

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.

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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



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