Basal Medium Eagle (BME)
With Earle's Salts and L-Glutamine
Without Sodium bicarbonate

Product Code: AT001

Product Description:
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.

AT001 is Basal Medium Eagle with Earle's balanced salts 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:

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>mg/L</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>INORGANIC SALTS</strong></td>
<td></td>
</tr>
<tr>
<td>Calcium chloride dihydrate</td>
<td>265.000</td>
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<tr>
<td>Magnesium sulphate anhydrous</td>
<td>97.720</td>
</tr>
<tr>
<td>Potassium chloride</td>
<td>400.000</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>6800.000</td>
</tr>
<tr>
<td>Sodium dihydrogen phosphate anhydrous</td>
<td>122.000</td>
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<tr>
<td><strong>AMINO ACIDS</strong></td>
<td></td>
</tr>
<tr>
<td>L-Arginine hydrochloride</td>
<td>21.100</td>
</tr>
<tr>
<td>L-Cystine dihydrochloride</td>
<td>15.650</td>
</tr>
<tr>
<td>L-Glutamine</td>
<td>292.000</td>
</tr>
<tr>
<td>L-Histidine hydrochloride</td>
<td>10.500</td>
</tr>
<tr>
<td>L-Isoleucine</td>
<td>26.200</td>
</tr>
<tr>
<td>L-Leucine</td>
<td>26.200</td>
</tr>
<tr>
<td>L-Lysine hydrochloride</td>
<td>36.480</td>
</tr>
<tr>
<td>L-Methionine</td>
<td>7.500</td>
</tr>
<tr>
<td>L-Phenylalanine</td>
<td>16.500</td>
</tr>
<tr>
<td>L-Threonine</td>
<td>23.800</td>
</tr>
<tr>
<td>L-Tryptophan</td>
<td>4.000</td>
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<tr>
<td>L-Tyrosine disodium salt</td>
<td>25.950</td>
</tr>
<tr>
<td>L-Valine</td>
<td>23.400</td>
</tr>
</tbody>
</table>

**VITAMINS**
- Choline chloride: 1.000
- 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: 2.000

**OTHERS**
- D-Glucose: 1000.000
- Phenol red sodium salt: 11.000

Directions:
1. Suspend 9.2gms in 900ml tissue culture grade water with constant, gentle stirring until the powder is completely dissolved. Do not heat the water.
2. Add 2.2gms of Sodium bicarbonate powder (TC230) or 29.3ml of 7.5% Sodium bicarbonate solution (TCL013) 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 the 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 9.2 gms/L.

pH without Sodium Bicarbonate
4.70 - 5.30

pH with Sodium Bicarbonate
7.40 - 8.00

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:
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