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### Product Profile

Product Name:	Minimum Essential Medium-Eagle(MEM-E) for	
	Suspension Culture	
Product Catalog Number	01-045-1	
Concentration:	1X	
Unit Size Availability:	(A)500ml (B)100ml	
Formulation:	Clear Red-Colored Solution	
Optimal Storage Conditions:	2-8°C	
Stability: (Under Specified Handling & Storage	Please Refer To Product Label	
Conditions)		

<u>Important Note!</u> Please read the <u>MSDS</u> and <u>Product Profile</u> carefully in their entirety <u>before</u> using this material for possible safety precautions and potential hazards.

### Product Description

Minimum Essential Medium (MEM) is one of the most widely used of all synthetic cell culture media originally developed by Harry Eagle. During early attempts to cultivate normal Mammalian Fibroblasts and certain subtypes of HeLa(i.e. Immortal Cell Line of Cervical Cancer Cells) cells revealed that they had unique niche nutritional requirements that could not be accommodated by Eagle's Basal Medium (BME). Subsequently, studies utilizing these and other cells in culture were indicative that additions or supplements could be made to the original BME medium and optimized or uniquely designed to promote continuous growth of a wider array of more intricate and complex cells and cell lines. The modification of MEM incorporating these essential add-ons including higher concentrations of Amino Acids (AA's) and/or other constituents significantly broadens its applicability. MEM comes highly recommended for a wide array of cell culture applications. MEM has typically been utilized for the cultivation of cells grown in monolayers (i.e. most cell cultures grow as a single thickness cell layer or sheet attached to a substrate) for a wide variety of normal and transformed cell lines as these new modifications have expanded its utility.

### Suspension Culture

Cell cultures are derived from either primary tissue explants or cell suspensions. Most cultures, including primaries, are propagated as a monolayer, anchored to a glass or plastic substrate. Some cultures, principally transformed cells, haemopoietic cells and ascites tumors may be propagated in suspension. Generally speaking, cultures derived from blood cells (e.g. lymphocytes) are grown in suspension. Cells may grow as single cells or in clumps (e.g., EBV transformed lymphoblastoid cell lines). Suspension cultures are fed by dilution into fresh medium. For such cell lines, subculture by dilution is relatively easy. But for lines that grow in clumps, it may be necessary to bring the cells into a single-cell suspension by centrifugation and resuspension by pipetting in a smaller volume before counting. Cells in suspension are either shaken, stirred or grown in vessels identical to those used for anchorage-dependent cells. Cell suspensions are clonally maintained by the routine subculture or transfer of cells in the early stationary phase to a fresh medium. During incubation, the biomass of the suspension cultures not only increases due to cell division but also cell development. This growth and enlargement occurs for a limited period only as the viability of cells after the stationary phase decreases due to the exhaustion of certain nutrients and/or parallel cytotoxic effects within the medium. At this point in time, an aliquot of the cell suspension with uniformly dispersed free cells and cell aggregates is transferred to a fresh, liquid medium of original composition. The timing of this subculture is crucial. The incubation period from culture initiation to the stationary phase is determined primarily by three (3) factors: a).Initial Cell Density, b).The Duration of Lag Phase and c).The Growth Rate of the Cell Line.

While initiating a new suspension culture, it is incumbent to determine optimal cell density which must be proportionate to the volume of the culture medium in order to achieve maximum growth. Cell cultures that are initiated at very low densities will not grow well unless the medium is enriched with the essential metabolites necessary to grow single cells of a small population thereof.

 $\label{thm:continuous} The \ advantage \ of \ growing \ cells \ in \ suspension \ may \ have \ some \ of \ the \ following \ characteristics:$ 

- Simpler Propagation (i.e. subculture requires only dilution, no trypsinization)
- ♣ No Requirement For Increasing Surface Area With Concomitant Increase In Bulk
- ♣ Ease of Harvesting
- Possibility of Achieving of a "Steady-State" Culture if required
- ← Cell Attachment must be discouraged
- Offers the easiest means of scale-up
- Often cell line specific

Cultured cells require a sterile environment and an optimal nutrient supply for growth and viability. Over the years variously defined media have been designed, developed, modified and enriched with a wide spectrum of constituents for supporting a vast range of cell types. Precise media formulations have been specifically developed by optimizing the concentrations of each and every component which performs a uniquely defined function.

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This type of cell culture medium consist of relevant concentrations of Amino Acids (AA's), Energy Sources, Inorganic Salts, and Vitamins among other nutrients. The modification, development and variation of this MEM formulation is a necessary prerequisite for the attachment, spreading and growth of certain cells *in vitro*. To maximize success, the *in vitro* culture conditions are designed to mimic such crucial *in vivo* conditions of nutrition, osmolality, pH and temperature. Optimal and critical nutrient components including such Inorganic Salts (e.g. NaCI, KCI, CaCl<sub>2</sub>), Amino Acids (e.g. Arginine, Histidine, Lysine), energy sources (e.g. Glucose, Glutamine, Pyruvate), and Vitamins(e.g. Nicotinimide, Thiamine, Riboflavin) are part and parcel that culminate in a perfect milieu for growth and viability. Whereas Amino Acids are incorporated into proteins and at a minimum these media must contain the essential Amino Acids, Vitamins are not only needed for cell growth and multiplication but also are important for cell survival and growth rate.

Unlike the balanced salt solutions that form the basis of many complex media formulations and are utilized to maintain cells for the short term in a viable condition, MEM may be modified and further enriched to promote the growth and viability of other cells in culture. MEM may be utilized for a broad and variegated spectrum of cell lines when properly supplemented.

These variegated components that constitute MEM-Eagle have been developed in order to fulfill the basic cell requirements for some of the basic and essential ions including: calcium, phosphate, potassium and sodium and therefore contain various amounts of  $CaCl_{l}$ , KCI, NaCl, and others . The key constituents of salts are the ions which function in osmolality whereas others such as Calcium and Magnesium are known, among other functions, to serve as cofactors for and support cell attachment and aggregation. Glucose and L-Glutamine serve as a major carbon and energy source and Phenol Red may serves as a pH indicator in specific cell culture media. MEM also contains Sodium Bicarbonate which has an intimate relationship with and plays a major role with  $CO_2$  by helping to maintain optimal physiological pH.

Some Predominant Characteristics of MEM – Eagle, for Suspension Culture include:

- ♦ Liquid Formulation
- ♦ <u>With Sodium Bicarbonate(NaHCO<sub>3</sub>)</u>
- ♦ With Phenol Red(C<sub>19</sub>H<sub>13</sub>NaO<sub>5</sub>S) as pH indicator
- ♦ <u>Without</u> L-Glutamine
- ♦ Sterile-Filtered(0.1µ)
- Cell Culture-Tested

### Storage & Stability

The product should be stored at 2-8°C. The medium should be warmed to room temperature prior to use. The product should not be left in the light for prolonged periods as it is light-sensitive. When stored in the dark under ideal conditions, the product is stable until the expiry date.

### Instructions /Procedure:

- 1) Take a bottle from the proper storage conditions at 2-8°C and read the label.
- 2) Ensure that the cap of the bottle is tight.
- 3) Gently swirl the solution in the bottle.
- 4) Wipe the outside of the bottle with a disinfectant solution such as 70% ethanol.
- 5) Using aseptic/sterile technique under a laminar-flow culture hood, work according to established protocols.

## **Quality Control:**

Test	Specification
Appearance:	Clear Solution
Cell Culture Test::	Pass Test
Endotoxins:	Test & Record
Osmolality:	300-320 mOsm/Kg
pH:	7.1-7.6
Sterility:	Sterile



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# Auxiliary Products:

Product Name	Catalog Number	Storage Temperature
Basal Medium Eagle (BME), Earle's Salts Base, without L-Glutamine,	01-015-5	2-8°C
without Sodium Bicarbonate 10X		
Minimum Essential Medium Eagle (MEM-E), Earle's Salts Base,	01-025-1	2-8°C
without L-Glutamine		
Minimum Essential Medium Eagle(MEM-NEAA), Earle's Salts Base,	01-040-1	2-8°C
with Non-Essential Amino Acids, without L-Glutamine		
Minimum Essential Medium Alpha(MEM-A) with 4.5g/l D-	01-043-1	2-8°C
Glucose(High Glucose) with L-Glutamine, without Ribonucleosides		
and Deoxyribonucleosides		
Medium-M-199(Earle's), Earle's Salts Base, with L-Glutamine	01-080-1	2-8°C
Medium-M-199 10X Conc., (Earle's), Earle's Salts Base, with L-	01-080-5	2-8°C
Glutamine, without Sodium Bicarbonate	04.040.4	2002
Ribonucleosides and Deoxyribonucleosides for MEM-Alpha, 500X	01-343-1	-20°C
Conc,  Earle's Balance Salt Solution 10X Conc., without Sodium Bicarbonate	02-010-5	Doors Townsonshine
Earle's Balance Salt Solution Tox Conc., without Socium Bicarbonate	02-010-5	Room Temperature (15-30°)
Earle's Balance Salt Solution without Phenol Red	02-011-1	Room Temperature
Edile's Dalance Salt Solution without Phenor Reu	02-011-1	(15-30°)
Earle's Balance Salt Solution without Phenol Red, without Sodium	02-011-5	Room Temperature
Bicarbonate	02-011-3	(15-30°)
L-Glutamine Solution 29.2mg/ml in Saline	03-020-1	-20°C
L-Alanyl-L-Glutamine Solution(A Dipeptide Substitute)	03-022-1	-20°C
Penicillin-Streptomycin Solution,10,000 units/ml Penicillin G Sodium	03-031-1	-20°C
Salt,10mg/ml Streptomycin	00 001 1	20 0
Sterile Culture-Grade Water	03-055-1	Room Temperature
		(15-30°)
Serum-Free Cell Freezing Medium	05-065-1	2-8°C
Note: For a list of other Antibiotics, Serum, Reagents and		
Supplements, please refer to our Product Catalog/Product Profiles,		
Product Guides and Internet Site.		

# References:

- 1) 2) 3) 4)
- Current Edition Merck Index
  Biological Industries(BI) Specifications
  Darling, D.C. and Morgan, S.J. <u>Animal Cells: Culture and Media</u>, New York: John Wiley & Sons,1994
  Lackie, J. M. <u>The Dictionary of Cell & Molecular Biology</u>, <u>Aca</u>demic Press: London, 2007



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