

Product Profile

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| Product Name: | Dulbecco's Modified Eagle Medium (DMEM):/Nutrient Mixture F-12(Ham's) (1:1)without L-Glutamine, with Sodium Bicarbonate 1.2gm/l,with Hepes 15mM,with Sodium Pyruvate 55mg/l |
| Product Catalog Number | 01-170-1 |
| Unit Size Availability: | (A)500ml ;(B)100ml |
| Concentration: | 1X |
| Formulation: | Clear Solution |
| Specified Storage Conditions: | 2-8°C |
| Stability: (Under Specified Handling & Storage) | Please Refer to the Product Label |

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

Product Description:

Dulbecco's Modified Eagle Medium (DMEM):/Nutrient Mixture F-12(Ham's) (1:1) without L-Glutamine, with Sodium Bicarbonate 1.2gm/l,with Hepes 15mM,with Sodium Pyruvate 55mg/l a.k.a. DMEM F-12 (1:1), was originally developed for a vast array of cell lines in a serum-free milieu that contained growth-promoting factors and hormones specific for a particular cell line. SF Media have been designed with specific cell requirements in mind and therefore, DMEM F-12(1:1) has shown effective utility by demonstrating the impact of variegated growth factors and hormones on target tissues. This medium serves as a foundation to grow cells in a Serum-Free environment or in very low serum conditions. This formulation has proven useful in reducing serum requirements for a wide variety of cell lines and applications with the inclusion of Hepes buffer that compensates for the loss of buffering capacity which is incurred by the elimination of serum. DMEM F-12 is a 1:1 mixture of DMEM and Ham's F-12 Nutrient Mixture that finds broad applicability in a serum-free culture milieu of not only normal Mouse and Chicken cell but also transformed cells which are indicative of cells whose growth characteristics have been altered in some manner.

Dulbecco's Modified Eagle's Medium is a very common Mammalian tissue cell culture medium. DMEM is Dulbecco's modification of Eagle's medium(BME) and is considered the most commonly utilized(e.g. MEM & RPMI) and less complex medium in contrast to the enriched media like Ham's F-12 or CMRL among others which are utilized not only for more specialized cell types but also as the basis for some of the more unique Serum-Free Media(SFM) formulations. DMEM is available with a High Glucose formulation and higher concentrations of Amino Acids (Essential & Non-Essential) and vitamins in addition to other ancillary constituents. The original or standard DMEM formulation contains 1000mg/L of glucose and was first reported for culturing Mouse Embryonic Cells MEC's. A higher glucose level (4500mg/l) has proven to be optimal for the cultivation of other cell types. These more complex media not only meet the minimum requirements for cell growth and proliferation but also are part and parcel of a much wider array of factors culminating in a final medium that segues with the essential cell-niche requirements demanded for optimal results.

The *series of Ham's Nutrient Mixtures of which Nutrient Mixture F-12(Ham)* is a part, was originally designed and developed specifically for supporting the growth of several clones of Chinese Hamster Ovary (CHO), HeLa clones and Mouse L-Cells. Both Nutrient Mixtures were developed with the intention that serum supplementation may be used as an option in serum-containing media or in a serum-free growth environment depending upon the individual cell culture. Ham's F-12 was designed and developed specifically for the growth of Primary Rat Hepatocytes (PRHs) and Rat Prostate Epithelial Cells (RPECs).F-12, as the medium of choice for a Clonal Toxicity Assay (CTA) has been utilized with Chinese Hamster Ovary (CHO) cells.

Most common types of media consists of an isotonic, buffered basal nutrient enriched environment which provides an energy source, inorganic salts, vitamins, amino acids as well as additional constituents(e.g. supplements) according to the demands of a particular cell line. This relatively more complex medium formulation provides the optimal cell-culture environment in which *in vitro* culture conditions mimic those of the *in vivo* environment including basic nutritional requirements, osmotic pressure, physiological pH, temperature among other considerations. At a minimum, it consists of the foundation medium components that are all part and parcel of a pre-tested complete media to assist the cells in meeting their metabolic demands.

Dulbecco's Modified Eagle Medium (DMEM):/Nutrient Mixture F-12(Ham's) (1:1) without L-Glutamine, with Sodium Bicarbonate 1.2gm/l,with Hepes 15mM,with Sodium Pyruvate 55mg/l contains no growth-promoting factors or antimicrobials. The type of medium recommended usually is dependent upon the type and character of the cells in culture. Supplementation is the process in which specific additions or supplements (e.g. growth factors, serum, fatty-acids, buffers, hormones) compliment a typical basal or balanced salt solution medium or more complex media such as RPMI 1640.

These more complex media not only meet the minimum requirements for cell growth and proliferation but also are part and parcel of a much wider array of factors culminating in a final medium that segues with the essential cell-niche requirements demanded for optimal results.

Function of Hepes

Hepes increases the buffer strength of the medium and raises the maximum buffering range. Hepes, in comparison to the sodium bicarbonate buffer system is more suitable as a buffer when a physiological pH range of 7.2-7.6 is desired and may be used with or without a CO₂ atmosphere, whereas the bicarbonate buffering system requires the use of a CO₂ incubator. Hepes buffer compensates for the loss of buffering capacity which is incurred by the elimination of serum or in a serum-deficient setting.

Function of L-Glutamine

The addition of L-Glutamine, a precursor of glutamate, is one of the most readily available sources of energy for many rapidly dividing cell-types for use *in vitro* and is a key component and essential amino acid that is required in many cell-culture media formulations and in virtually all mammalian cells in culture. Also adding Sodium pyruvate serves as an additional and easily accessible carbohydrate energy source for cells in culture. Along with D-glucose, these balanced energy sources serve as carbon skeletons for cell anabolic processes in addition to nucleic acid metabolism and protein production while limiting the potential cumulative build-up effects of toxic levels of ammonia.

Function of Sodium Bicarbonate

To maximize success in cell culture, the *in vitro* culture conditions are created to mimic the *in vivo* conditions of osmolality, pH, temperature and nutrition. Ions such as HCO_3^- and Na^+ among others are the major contributors to the osmolality of cell culture media. HCO_3^- levels are determined by the concentration of CO_2 in the incubator (i.e. in contact with the growth medium). Sodium Bicarbonate (NaHCO_3) and CO_2 buffering is probably the most popular system used which requires a CO_2 level of 5-10 % (i.e. dependent on the media utilized) and 100% humidity.

Culture media are often buffered to compensate for the cellular production of CO_2 and Lactic Acid as by-products of metabolism. Traditionally, Basal Cell Culture media have been buffered by HCO_3^- (Bicarbonate). As cells grow, CO_2 evolves; the dissolved CO_2 forms a buffering system with the bicarbonate. However, if cell density is low or the cells have entered into the so-called "Lag Phase," they may not produce sufficient CO_2 to maintain optimal pH and to counter these potential problems, Bicarbonate-Buffered media require the use of incubators with a 5-10% CO_2 atmosphere. Media with low levels of bicarbonate (HCO_3^-) on the one hand, such as MEM(@1.5-2.2g/l) require ~5% CO_2 . DMEM with higher levels of bicarbonate (i.e., 3.7g/L) on the other, require 10% CO_2 in order to maintain the correct pH level. The most important factor in utilizing the correct percent CO_2 is based upon the medium's bicarbonate level to maintain physiological pH, which is irrespective of cell type.

DMEM F-12 (1:1) contains numerous important basic constituents in a ready-to-use formulation, which includes a typical and wide variety of elements, among others:

- ◆ Amino Acids
- ◆ Vitamins
- ◆ Inorganic Salts
- ◆ Phenol Red
- ◆ Trace Elements

Some Predominant Characteristics of **DMEM F-12 (1:1)** includes:

- § Liquid Formulation
- § With Hepes(15mM)
- § With 1.2gm/Sodium Bicarbonate(NaHCO_3)
- § With D-Glucose
- § With Phenol Red($\text{C}_{19}\text{H}_{13}\text{NaO}_5\text{S}$) as a pH indicator
- § With Sodium Pyruvate(55mg/l)
- § Without L-Glutamine
- § Promotes Cell Performance and Productivity
- § More Uniform & Consistent Media Performance
- § Sterile-Filtered(0.1 μ), Cell-Culture-Tested

Storage, Handling, Stability Precautions and Disclaimer:
For *in vitro* diagnostic use only.

DMEM F-12 (1:1) is stable when stored under defined conditions at 2-8°C. The product is light-sensitive and therefore should not be left in the light. When stored in the dark under ideal conditions, the product is stable until the expiry date.

As with any other liquid media formulations, deterioration of liquid media may be recognized by any of the following characteristics, among others including: (a). color change, (b). presence of clumping/flocculent debris/ granulation/ particulates/ precipitates or sediments (c). Insolubility, (d). And/or decrease in expected performance parameters. Any material described above should not be used and therefore discarded.

Instructions/Procedure:

- 1) Take a bottle from the defined storage conditions at 2-8°C and read the label. Warm to Room Temperature (15-30° C) prior to use.
- 2) Ensure that the bottle cap is tight and swirl the bottle for homogeneity.
- 3) Wipe the outside of the bottle with a disinfectant solution such as 70% ethanol.
- 4) Using aseptic/sterile technique under a laminar-flow culture hood, work according to established protocols.
- 5) Antibiotics may be added if desired.

Quality Control:

| Test | Specification |
|---------------|-----------------|
| Appearance: | Clear Solution |
| Cell Culture: | Test and Record |
| Cell Line: | Hybridoma |
| Endotoxins: | Test and Record |
| Osmolality: | 288-318 mOsm/kg |
| pH : | 7.1-7.5 |
| Sterility: | Sterile |

Auxiliary Products

| Product Name | Catalog Number | Storage Temperature |
|---|----------------|---------------------|
| Nutrient Mixture F-10(Ham's) ,with L-Glutamine | 01-090-1 | 2-8°C |
| Nutrient Mixture F-10 (Ham's),10X Conc., with L-Glutamine, without Sodium Bicarbonate | 01-090-5 | 2-8°C |
| RPMI 1640, with L-Glutamine | 01-100-1 | 2-8°C |
| RPMI 1640, without D-Glucose, without L-Glutamine | 01-101-1 | 2-8°C |
| RPMI 1640, without Phenol Red, without L-Glutamine | 01-103-1 | 2-8°C |
| RPMI 1640, without L-Glutamine | 01-104-1 | 2-8°C |
| RPMI 1640 10XConc., without L-Glutamine, without Sodium Bicarbonate | 01-104-5 | 2-8°C |
| Dulbecco's Phosphate Buffered Saline(DPBS) without Calcium and Magnesium | 02-023-1 | 15-30°C(RT) |
| 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 | 03-031-1 | -20°C |
| Sodium Bicarbonate Solution (7.5%) | 03-040-1 | 15-30°C (RT) |
| Sodium Bicarbonate Solution (5%) | 03-041-1 | 15-30°C (RT) |
| Sodium Pyruvate | 03-042-1 | -20°C |
| Water, Cell Culture Grade | 03-055-1 | 15-30°C (RT) |
| Fetal Bovine Serum | 04-001-1 | -20°C |
| Fetal Bovine Serum(Qualified for Human Embryonic Stem Cells) | 04-002-1 | -20°C |
| Adult Bovine Serum | 04-003-1 | -20°C |
| Donor Horse Serum | 04-004-1 | -20°C |
| Fetal Bovine Serum(TET System Approved) | 04-005-1 | -20°C |
| Porcine Serum | 04-006-1 | -20°C |
| European Grade Fetal Bovine Serum | 04-007-1 | -20°C |
| Rabbit Serum | 04-008-1 | -20°C |
| Donor Goat Serum | 04-009-1 | -20°C |
| Fetal Bovine Serum(Dialyzed) | 04-011-1 | -20°C |
| Special Newborn Calf Serum(Less than 10 days of age) | 04-102-1 | -20°C |
| Fetal Bovine Serum(Heat-Inactivated) | 04-121-1 | -20°C |
| Special Newborn Calf Serum(Heat-Inactivated) | 04-122-1 | -20°C |
| Adult Bovine Serum(Heat Inactivated) | 04-123-1 | -20°C |
| Note: For a list of other Antibiotics, Serum or other Reagents and Supplements, please refer to our Product Catalog/Product Profiles/Guides and Internet Site. | | |

References:

- 1) Biological Industries (BI) Specifications
- 2) Darling, D.C. and Morgan S.J. *Animal Cells: Culture and Media*, John Wiley & Sons, New York, 1994
- 3) Lackie, J. M. *The Dictionary of Cell & Molecular Biology*, Academic Press: London, 2007

Biological Industries