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Technical Datasheet

Dulbecco's Modified Eagle Medium/ Nutrient Mixture F12 Ham (DMEM/F12, 1:1 mixture)

With L-Glutamine, 2mg/L Riboflavin and 55mg/L Sodium pyruvate
Without HEPES buffer, Trace elements, Pyridoxal hydrochloride and Sodium bicarbonate

Product Code: AT243

Product Description :

Dulbecco's Modified Eagle Medium / Nutrient Mixture F12 Ham DMEM/F12, 1:1 mixture) was originally formulated for rat neuroblastoma cells and MDCK cells. The mixture is extremely nutritious and supports growth of a wide variety of cells including certain epithelial, endothelial and granulosa cells.

AT243 is DMEM/ Nutrient Mixture F-12 Ham with L-glutamine, 2mg/L Riboflavin and 55mg/L Sodium pyruvate. It does not contain HEPES buffer, trace elements and Pyridoxal hydrochloride. 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	154.500
Copper sulphate pentahydrate	0.0013
Disodium hydrogen phosphate, anhydrous	71.020
Ferric nitrate nonahydrate	0.050
Ferrous sulphate heptahydrate	0.417
Magnesium chloride anhydrous	28.640
Magnesium sulphate anhydrous	48.840
Potassium chloride	311.800
Sodium chloride	6996.000
Sodium dihydrogen phosphate anhydrous	54.300
Zinc sulphate heptahydrate	0.432
AMINO ACIDS	
Glycine	18.750
L-Alanine	4.450
L-Arginine hydrochloride	147.500
L-Asparagine monohydrate	7.500
L-Aspartic acid	6.650
L-Cysteine dihydrochloride	31.290
L-Cystine hydrochloride monohydrate	17.560
L-Glutamic acid	7.350
L-Glutamine	365.000
L-Histidine hydrochloride monohydrate	31.480
L-Isoleucine	54.470
L-Leucine	59.050
L-Lysine hydrochloride	91.250
L-Methionine	17.240
L-Phenylalanine	35.480
L-Proline	17.250
L-Serine	26.250
L-Threonine	53.450
L-Tryptophan	9.020
L-Tyrosine disodium salt dihydrate	48.100
L-Valine	52.850
VITAMINS	
Choline chloride	8.980
D-Biotin	0.0035
D-Ca-Pantothenate	2.240
Folic acid	2.660
Niacinamide	2.020
Pyridoxine hydrochloride	0.031
Riboflavin	2.000
Thiamine hydrochloride	2.170
Vitamin B12	0.680
myo-Inositol	12.600
OTHERS	
D-Glucose	3151.000
DL-Thioctic Acid	0.105
Hypoxanthine sodium salt	2.400
Linoleic acid	0.042
Phenol red Sodium Salt	8.630
Putrescine hydrochloride	0.081
Sodium pyruvate	55.000
Thymidine	0.365

Directions :

1. Suspend 12.01gms in 900ml tissue culture grade water with constant and gentle stirring until the powder is completely dissolved. Do not heat the water. .
2. Add 1.2gms of sodium bicarbonate powder (TC230) or 16ml 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 liquid medium at 2-8°C and in dark till use.

Materials 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
White to light pink, homogenous powder
Solubility
Clear solution at 12.01 gms/L
pH without Sodium Bicarbonate
6.00 -6.60
pH with Sodium Bicarbonate
7.20 -7.80
Osmolality without Sodium Bicarbonate
260.00 -300.00
Osmolality with Sodium Bicarbonate
285.00 -325.00
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.
Endotoxin content
NMT 1EU/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. Inspite 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 filter sterilization observing sterility precautions. Shelf life of the medium will depend on the nature of supplement added to the medium.