

Product Information

Ham's Nutrient Mixture F12

with 1.0 mM L-glutamine, without sodium bicarbonate

CATALOG NO. 56659C

Description

Ham's Nutrient Mixture F12 was originally developed for the serum-free clonal growth of Chinese Hamster Ovary (CHO) cells, lung cells and mouse L-cells. It is frequently used with dialyzed serum, hormones, selenium and other supplements for serum-free cultures. It is the medium of choice for supporting the growth of cells of rodent origin, particularly rabbit and rat, and has proven to be an excellent cloning medium for myeloma and hybridoma cells.

Precautions

Use aseptic technique when handling or supplementing this medium after filtration. This product is for further manufacturing use. THIS PRODUCT IS NOT INTENDED FOR HUMAN OR THERAPEUTIC USE.

Storage

Store dry powder medium at 2 to 8 C. Do not use after expiration date. Store hydrated medium protected from light at 2 to 8 C.

Indications of Deterioration

Dry powder medium should be free flowing. Do not use if powder is caked. Prepared medium should be clear of particulates and flocculent material. Do not use if liquid medium is cloudy or contains precipitate. Other evidence of deterioration may include color change or degradation of physical or performance characteristics.

Formulation

Component (all components measured in mg/L)	
INORGANIC SALTS	
Calcium chloride anhydrous	33.220
Cupric sulfate pentahydrate	0.00249
Ferrous sulfate heptahydrate	0.834
Magnesium chloride anhydrous	57.220
Potassium chloride	223.650
Sodium chloride	7599.000
Sodium phosphate dibasic anhydrous	142.040
Zinc sulfate heptahydrate	0.863
VITAMINS	
Biotin	0.00733
D-calcium pantothenate	0.238
Choline chloride	13.960
Cyanocobalamin	1.357
Folic acid	1.300
i-inositol	18.020
Niacinamide	0.03663
Pyridoxine HCl	0.06171
Riboflavin	0.03764
Thiamine HCl	0.03373
AMINO ACIDS	
L-alanine	8.910
L-arginine HCl	210.700
L-asparagine monohydrate	15.010
L-aspartic acid	13.310
L-cysteine HCl monohydrate	35.120
L-glutamic acid	14.710
L-glutamine	146.200
Glycine	7.510
L-histidine HCl monohydrate	20.960
L-isoleucine	3.936
L-leucine	13.120
L-lysine HCl	36.540
L-methionine	4.476
L-phenylalanine	4.956
L-proline	34.530
L-serine	10.510
L-threonine	11.910
L-tryptophan	2.042
L-tyrosine 2Na dihydrate	7.780
L-valine	11.710
OTHER	
Dextrose anhydrous	1801.600
Hypoxanthine sodium salt	4.770
Linoleic acid	0.08412
DL- α -Lipoic acid	0.2063
Phenol red sodium salt	1.2740
Putrescine 2HCl	0.161
Sodium pyruvate	110.100
Thymidine	0.7266
ADD: Sodium bicarbonate	1176.000
Grams of powder per liter	10.625

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Preparation Instructions

1. Measure 80 - 90% of the final volume of cell culture grade water (Catalog No. 59900C) into an appropriate size mixing vessel. Water temperature should be 15 to 30 C. Do not heat water.
2. Add the dry powder medium to the water. Rinse the original package with a small amount of cell culture grade water to remove all traces of powder and add to the solution. Mix until completely dissolved.
3. For each liter being prepared, add 1.176 g./L of sodium bicarbonate (Catalog No. 90421C) or 15.7 mL/L of sodium bicarbonate solution 7.5% (Catalog No. 59221C). Mix until completely dissolved.
4. While stirring the solution in Step 3, adjust the pH to 6.9 - 7.1 using NaOH 1N (Catalog No. 59223C) or HCl 1N. The pH of bicarbonate buffered solutions usually rises 0.1 - 0.2 units during filtration.
5. Add cell culture grade water to the solution in Step 4 to bring it to the final volume. Keep the vessel closed until the solution is filtered to avoid fluctuations in pH.
6. Sterilize the solution using a membrane filter with a pore size of 0.22 µm or less. A peristaltic pump or an inert gas such as nitrogen can be used to provide positive pressure at 3 - 15 psi. Do not use CO₂ gas.
7. Sterile solutions should be dispensed aseptically into sterile containers. Store protected from light at 2 to 8 C.
8. Supplements, such as antibiotics, can be added to the sterilized solution using aseptic technique. Storage conditions and shelf life of the supplemented product may be affected by the nature of the supplements. Sterile serum should not be refiltered before or after being added to sterile medium because growth promoting capacity may be reduced upon refiltration.

NOTE: Dry powder medium is extremely hygroscopic and must be protected from atmospheric moisture. We recommend that the entire contents of each package be used immediately after opening.

Characteristics

Appearance

Off-white free-flowing powder

Bioburden

≤ 100 CFU/100 mL

Endotoxin

≤ 1.0 EU/mL

Osmolality (as supplied)

240 - 280 mOsm/kg H₂O

pH (as supplied)

6.6 - 7.0

References

1. Ham, R. G., *PNAS* (1965) 53:288.

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Issued December 2006 P56659
1203 1005 0406 0606 0906

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