

# Product Information

## Dulbecco's Modified Eagle's Medium/ High Modified

with 4500 mg/L dextrose, with 4.0 mM L-glutamine, with 110 mg/L sodium pyruvate,  
without sodium bicarbonate

CATALOG NO. 56499C

### Description

Dulbecco's Modified Eagle's Medium (DMEM) is a modification of Basal Medium Eagle (BME) that contains four-fold concentrations of the amino acids and vitamins. The original formulation contained 1000 mg/L of dextrose and was used to culture embryonic mouse cells. Since then, it has been modified in several ways to support primary cultures of mouse and chicken cells, as well as a variety of normal and transformed cells. Additionally, the dextrose level has been raised to 4500 mg/L, contributing to the name "DMEM/High."

### 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)	
<b>INORGANIC SALTS</b>	
Calcium chloride anhydrous	200.000
Ferric nitrate nonahydrate	0.100
Magnesium sulfate anhydrous	97.670
Potassium chloride	400.000
Sodium chloride	6400.000
Sodium phosphate monobasic monohydrate	125.000
<b>VITAMINS</b>	
D-calcium pantothenate	4.000
Choline chloride	4.000
Folic acid	4.000
i-inositol	7.000
Niacinamide	4.000
Pyridoxal HCl	4.000
Riboflavin	0.400
Thiamine HCl	4.000
<b>AMINO ACIDS</b>	
L-arginine HCl	84.000
L-cystine 2HCl	62.570
L-glutamine	584.000
Glycine	30.000
L-histidine HCl monohydrate	42.000
L-isoleucine	104.800
L-leucine	104.800
L-lysine HCl	146.200
L-methionine	30.000
L-phenylalanine	66.000
L-serine	42.000
L-threonine	95.200
L-tryptophan	16.000
L-tyrosine 2Na dihydrate	103.790
L-valine	93.600
<b>OTHER</b>	
Dextrose anhydrous	4500.000
Phenol red sodium salt	15.930
Sodium pyruvate	110.000
ADD: Sodium bicarbonate	3700.000
Grams of powder per liter	13.485

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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 3.70 g/L of sodium bicarbonate (Catalog No. 90421C) or 49.3 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  $\mu\text{m}$  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 CO<sub>2</sub> 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 as sterile supplements 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

$\leq 100$  CFU/100 mL

### Endotoxin

$\leq 1.0$  EU/mL

### Osmolality (as supplied)

240 - 280 mOsm/kg H<sub>2</sub>O

### Osmolality (with NaHCO<sub>3</sub>)

315 - 355 mOsm/kg H<sub>2</sub>O

### pH (as supplied)

6.0 - 6.4

### pH (with NaHCO<sub>3</sub>)

7.5 - 7.9

## References

1. Eagle, H., *Science* (1959) 130:432.
2. Dulbecco, R. and Freeman, G., *Virology* (1979) 8:396.
3. Smith, J. D., Freeman, G., Vogt, M., and Dulbecco, R., *Virology* (1960) 12:185.
4. Morton, H. J., *In Vitro* (1970) 6:89.

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