

DULBECCO'S MODIFIED EAGLE'S MEDIUM

With L-Glutamine and 1000 mg/L Glucose, Without L-Cystine, L-Methionine and Sodium Bicarbonate

Product Number **D3916** Storage Temperature 2-8°C

Product Description

Many modifications of Eagle's Medium have been developed since the original formulation appeared in the literature. Among the most widely used of these modifications is Dulbecco's Modified Eagle's Medium (DME). DME is a modification of Basal Medium Eagle (BME) that contains a higher concentration of amino acids and vitamins, as well as additional supplementary components. The original DME formula contains 1000 mg/L of glucose and was first reported for culturing embryonic mouse cells. A further modification with 4500 mg/L glucose has proved to be optimal in cultivating certain cell types.

DULBECCO'S MODIFIED EAGLE'S MEDIUM, Product No. D3916 is one of the cell culture media available from Sigma. The selection of a nutrient medium is strongly influenced by 1] type of cell, 2] type of culture [monolayer, suspension, clonal] and 3] degree of chemical definition necessary. It is important to review the literature for recommendations concerning medium, supplementation and physiological parameters required for a specific cell line.

Components	<u>g/L</u>
Calcium Chloride•2H ₂ O	0.265
Ferric Nitrate•9H ₂ O	0.0001
Magnesium Sulfate (anhydrous)	0.09767
Potassium Chloride	0.4
Sodium Chloride	6.4
Sodium Phosphate Monobasic (anhydrous	s) 0.109
L-Arginine•HCl	0.084
L-Glutamine	0.584
Glycine	0.030
L-Histidine•HCI•H ₂ O	0.042
L-Isoleucine	0.105
L-Leucine	0.105
L-Lysine•HCl	0.146
L-Phenylalanine	0.066
L-Serine	0.042
L-Threonine	0.095
L-Tryptophan	0.016
L-Tyrosine•2Na•2H ₂ O	0.10379
L-Valine	0.094
Choline Chloride	0.004

ProductInformation

Folic Acid	0.004
myo-Inositol	0.0072
Niacinamide	0.004
D-Pantothenic Acid (hemicalcium)	0.004
Pyridoxal•HCl	0.004
Riboflavin	0.0004
Thiamine•HCI	0.004
D-Glucose	1.0
Phenol Red•Na	0.0159
Pyruvic Acid•Na	0.11

Precautions and Disclaimer

REAGENT

For In Vitro Diagnostic Use

Preparation Instructions

Powdered media are extremely hygroscopic and should be protected from atmospheric moisture. The entire contents of each package should be used immediately after opening. Preparing a concentrated solution of medium is not recommended as precipitates may form.

Supplements can be added prior to filtration or introduced aseptically to sterile medium. The nature of the supplement may affect storage conditions and shelf life of the medium.

- Measure out 90% of final required volume of water. Water temperature should be 15-20°C.
- 2. While gently stirring the water, add the powdered medium. Stir until dissolved. Do NOT heat.
- Rinse original package with a small amount of water to remove all traces of powder. Add to solution in step 2.
- 4. To the solution in step 3, add 3.7 g sodium bicarbonate or 49.3 ml of sodium bicarbonate solution [7.5%w/v] for each liter of final volume of medium being prepared. Stir until dissolved.
- 5. While stirring, adjust the pH of the medium to 0.1-0.3 pH units below the desired pH since it may rise during filtration. The use of 1N HCl or 1N NaOH is recommended.
- 6. Add additional water to bring the solution to final volume.

- 7. Sterilize immediately by filtration using a membrane with a porosity of 0.22 microns.
- 8. Aseptically dispense medium into sterile container.

Storage/Stability

Store the dry powdered medium at 2-8°C under dry conditions and liquid medium at 2-8°C in the dark. Deterioration of the powdered medium may be recognized by any or all of the following: [1] color change, [2] granulation/clumping, [3] insolubility. Deterioration of the liquid medium may be recognized by any or all of the following: [1] pH change, [2] precipitate or particulate matter throughout the solution, [3] cloudy appearance [4] color change. The nature of supplements added may affect storage conditions and shelf life of the medium. Product label bears expiration date.

Procedure

Materials Required but Not Provided Water for tissue culture use [W3500] Sodium Bicarbonate [S5761] or Sodium Bicarbonate Solution, 7.5% [S8761] 1N Hydrochloric Acid [H9892] 1N Sodium Hydroxide [S2770] Medium additives as required

Product Profile

Appearance	off-white powder
Moisture content	≤ 2.0%
Solubility cl	ear solution at 1x concentration
pH at RT [without sodium b	6.7 ± 0.3
pH at RT [with sodium bica	7.6 ± 0.3 rbonate]
Osmolality	$240 \text{ mOsm/kg} \text{H}_{2}\text{O} + 5\%$

Osmolality	240 mOsm/kg H ₂ O	± 5%
[without sodium bicar	bonate]	

Osmolality 311 mOsm/kg $H_2O \pm 5\%$ [with sodium bicarbonate]	
Endotoxin	\leq 1.0 EU/ml at 1x
Amino Acid Analysis by HPLC	Analysis has confirmed that amino acids are present at concentrations consistent with t he formula.
Key Element Analysis by ICAP	Analysis has confirmed that key elements are present at concentrations consistent with the formula.

Biological Performance Characteristics

Biological performance is assessed using an appropriate cell line(s). Growth studies are carried through 2 subculture generations. Cells are counted and growth is plotted as a logarithmic function of time in culture. Seeding efficiencies, doubling time, and final cell densities are determined. During the testing period cultures are examined microscopically for atypical morphology and evidence of cytotoxicity. Test results are available upon request.

References

- 1. Dulbecco, R. and Freeman, G.(1959). Plaque Production by the Polyoma Virus. Virology. 8, 396-397.
- 2. Smith,J.D., Freeman,G., Vogt,M. and Dulbecco, R.(1960). The Nucleic Acid of Polyoma Virus. 12, 185-196.
- Morton, H..J., (1970). A Survey of Commercially Available Tissue Culture Media.In Vitro. 6, 89.
- Rutzky, L.P. and Pumper, R.W., (1974). Supplement to a Survey of Commercially Available Tissue Culture Media(1970). In Vitro. 9, 468.

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