

Medium 199 with Earle's salts

Product No. A1325

Description

Powder mixture to prepare Powder mixture to prepare Medium 199 with Earle's salts according to Morgan, J.F. et al. (1950) *Proc. Soc. Exp. Biol. Med.* **73**, 1 and Morgan, J.F. et al. (1955) *J. Nat. Cancer Inst.* **16**, 557

with L-Glutamine
without Sodium hydrogen carbonate

Hygroscopic!
Storage: 2-8°C

Instructions

General Information: Powdered media and salts are very hygroscopic and must be stored under dry conditions. After opening the package the whole contents must be dissolved at once.

Reconstitute the powdered form of media to produce 1X liquid medium, as the different amino acids may precipitate at higher concentrations. They potentially can form salts which are of low solubility in concentrated solutions. If supplements are needed, they can be added before filtration (unsterile) or after filtration (sterile).

Use bidistilled or deionized, pyrogen-free water to reconstitute powder media.

Preparing sterile filtered liquid medium

- 1.) Add water to the required quantity of powdered medium (use approx. 90 % of the required amount of water so as to adjust the pH later). Flush out any remaining powder from the container. Stir until completely dissolved. The temperature of the water should be between 15-30°C
- 2.) When the powder is completely dissolved, **add Sodium hydrogen carbonate (NaHCO₃) 2,200 g per liter of final medium** and dissolve completely as well.
- 3.) Adjust to the desired pH value (physiological optimum is pH 6.8 - 7.2) with 1 M HCl or 1 M NaOH while stirring.

Note: The pH should be approx. 0.2 units lower than the target pH, since pH will rise slightly during filtration when CO₂ leaks out.

- 4.) After adjusting the pH, add water to the appropriate final volume and mix well. Filter immediately under sterile conditions.
- 5.) Store the medium at 2-8°C protected from light.

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Composition:

Components		mg/L final medium:
Inorganic salts	Calcium chloride x 2H ₂ O	264,92
	Potassium chloride	400,00
	Magnesium sulfate dried	139,53
	Sodium acetate x 3H ₂ O	82,95
	Sodium chloride	6800,00
	Sodium dihydrogen phosphate x H ₂ O	140,00
	Iron(III) nitrate x 9H ₂ O	0,72
Other Components	Phenol red	10,00
	Tween 80	4,90
	L-Glutamine	100,00
	Adenine sulfate	10,00
	Adenosine 5'-monophosphoric acid disodium salt x 2H ₂ O (AMP)	0,20
	Adenosine 5'-triphosphoric acid disodium salt (ATP)	1,00
	Cholesterol	0,20
	D(+)-Glucose anhydr.	1000,00
	L-Glutathione red.	0,05
	Guanin x HCl	0,30
	Hypoxanthine	0,30
	D(-)-Ribose	0,50
	Thymine	0,30
	Xanthine	0,30
Amino acids	L-Alanine	25,00
	L-Arginine x HCl	70,00
	L-Aspartic acid	30,00
	L-Cysteine x HCl x H ₂ O	0,10
	L-Cystine	20,00
	L-Glutamic acid	67,00
	Glycine	50,00
	L-Histidine x HCl x H ₂ O	21,88
	L-Hydroxyproline	10,00
	L-Isoleucine	20,00
	L-Leucine	60,00
	L-Lysine x HCl	70,00

	L-Methionine	15,00
	L-Phenylalanine	25,00
	L-Proline	40,00
	L-Serine (non animal origin)	25,00
	L-Threonine	30,00
	L-Tryptophan	10,00
	L-Tyrosine	40,00
	L-Valine	25,00
Vitamins	4-Amino benzoic acid	0,05
	L(+)-Ascorbic acid powdered	0,05
	D(+)-Biotin	0,01
	Vitamin D2	0,10
	D-Calcium pantothenate	0,01
	Choline chloride	0,50
	Folic acid	0,01
	myo-Inositol	0,05
	Menadione	0,01
	Nicotinic acid	0,03
	Nicotinamide	0,03
	Pyridoxal x HCl	0,03
	Pyridoxine x HCl	0,03
	Riboflavin	0,01
	DL-alpha-Tocopherol phosphate disodium salt	0,01
	Thiamine x HCl	0,01
	Vitamin A acetate	0,14
	Uracil	0,30
Deoxyribonucleoside	2-Deoxy-D-ribose	0,50