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Product Specification

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Technical Information

RPMI-1640

With L-Glutamine, Phenol red, 2gms per litre Glucose and 0.165 moles per litre MOPS buffer Without Sodium bicarbonate

Product Code: AT1180

Application:- Roswell Park Memorial Institute (RPMI) media are a series of media developed by Moore et al for the culture of human normal and neoplastic cells in vitro. RPMI 1640 is the most commonly used medium in the series. A modification of McCoy's 5A medium, the medium was specifically designed to support the growth of human lymphoblastoid cells in suspension culture. Presently the medium is extensively used for a wide range ofanchorage dependant cell lines. The medium needs to be supplemented with 5-20% fetal bovine serum. The medium is also known to support growth of cells in the absence of serum.

AT1180 is RPMI 1640 with L-glutamine, 2gms per litre glucose, 0.165M per litre MOPS buffer. It does not contain sodium bicarbonate. *"MOPS, a zwitterionic buffer does not antagonize antifungal agents at final concentration of 0.165mol/L for pH 7.0. Therefore, this medium is used as a diluent for antifungal agents that are water-soluble as well as water-insoluble. For water- insoluble antifungal agents, that cannot be prepared as stock solutions in water, such as amphotericin B,anidulafungin, itraconazole, ketoconazole, posaconazoleand voriconazole, a dilution series of the agent should be prepared first at 100 times final strength in an appropriate solvent. Each of these non-aqueous solutions should then be diluted tenfold in RPMI-1640 broth". Users are advised to review the literature for recommendations regarding medium supplementation and physiological growth requirements specific for different cell lines.

Composition**

Ingredients	mg/Litre
INORGANIC SALTS	
Calcium nitrate tetrahydrate	100.000
Magnesium sulphate anhydrous	48.840
Potassium chloride	400.000
Sodium chloride	6000.00
Sodium phosphate dibasic anhydrous	800.000
AMINO ACIDS	
Glycine	10.000
L-Arginine hydrochloride	241.000
L-Asparagine	50.000
L-Aspartic acid	20.000
L-Cysteine dihydrochloride	65.200
L-Glutamic acid	20.000
L-Glutamine	300.000
L-Histidine hydrochloride monohydrate	20.960
L-Hydroxyproline	20.000
L-Isoleucine	50.000
L-Leucine	50.000

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L-Lysine hydrochloride	40.000
L-Methionine	15.000
L-Phenylalanine	15.000
L-Proline	20.000
L-Serine	30.000
L-Threonine	20.000
L-Tryptophan	5.000
L-Tyrosine disodium salt	28.830
L-Valine	20.000
VITAMINS	
Choline chloride	3.000
D-Biotin	0.200
D-Ca-Pantothenate	0.250
Folic acid	1.000
Niacinamide	1.000
Pyridoxine hydrochloride	1.000
Riboflavin	0.200
Thiamine hydrochloride	1.000
Vitamin B12	0.005
i-Inositol	35.000
p-Amino benzoic acid (PABA)	1.000
OTHERS	
D-Glucose	2000.00
Glutathione reduced	0.500
MOPS Buffer, Free acid	34500.00
Phenol red sodium salt	5.300

Methodology

1. Suspend 44.9gms in 900ml tissue culture grade water with constant, gentle stirring until the powder is completely dissolved. Do not heat the water.
2. Add 2.0gms of sodium bicarbonate powder (TC1230) or 26.67ml of 7.5% sodium bicarbonate solution (TCL1013) 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.

Material required but not provided :

Tissue culture grade water (TCL1010)
Sodium bicarbonate (TC1230)
Sodium bicarbonate solution, 7.5% (TCL1013)
1N Hydrochloric acid (TCL1003)
1N Sodium hydroxide (TCL1002)
Foetal bovine serum (BA3112/BA12432)

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Quality Control

Appearance

Off-white to Creamish white, homogenous powder.

Solubility

Clear solution at 44.9gms/L.

pH without Sodium Bicarbonate

5.20 -5.80

pH with Sodium Bicarbonate

6.00 -6.60

Osmolality without Sodium Bicarbonate

390.00 -430.00

Osmolality with Sodium Bicarbonate

430.00 -470.00

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 and comparing itwith a control medium through minimum three subcultures.

Endotoxin content

NMT 5EU/ml

Storage and Shelf Life

1. All the powdered media and prepared liquid culture mediashould be stored at 2-8°C. Use before the expiry date. In spiteof above recommended storage condition, certain powderedmedium may show some signs of deterioration /degradationin 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 recommendedsince free base amino acids and salt complexes having low solubility may precipitate in concentrated medium.
3. pH and sodium bicarbonate concentration of the preparedmedium are critical factors affecting cell growth. This is also influenced by amount of medium and volume of culturevessel 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 priorto or after filter sterilization observing sterility precautions. Shelf life of the medium will depend on the nature of supplement added to the medium.

Disclaimer :

- User must ensure suitability of the product(s) in their application prior to use.
- The product conforms solely to the technical information provided in this booklet and to the best of knowledge research and development work carried at **CDH** is true and accurate.
- **Central Drug House Pvt. Ltd.** reserves the right to make changes to specifications and information related to the products at any time.
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- Do not use the products if it fails to meet specifications for identity and performance parameters.