



Technical Datasheet

Krebs-Ringer Phosphate HEPES Buffer

With 20mM HEPES buffer, Calcium and Magnesium Without Phenol red

Product Code: TS1152

Product Description:

All media used in tissue culture have a basis of a synthetic mixture of inorganic salts known as a physiological or balanced salt solution (BSS). All the physiological salt solutions have been derived from the salt solution originally described by Sydney Ringer (1885). The first balanced salt solution to be developed specifically for supporting the metabolism of mammalian cells was Tyrode's solution. Since then many modifications have been done to obtain better buffering salt solutions and to prevent calcium precipitation.

The function of a salt solution is:

- To maintain the medium within physiological pH range.
- To maintain intracellular and extra cellular osmotic balance.
- Modified with a carbohydrate, such as glucose serves as an energy source for cell metabolism.

TS1152 is Krebs-Ringer phosphate HEPES buffer with 20mM HEPES buffer, Calcium and Magnesium. It does not contain phenol red.

Composition:

Ingredients	
OTHERS	
Calcium chloride dihydrate 147.0)10
HEPES buffer 4766.	000
Magnesium sulfate anhydrous 120.3	370
Potassium chloride 350.3	385
Potassium dihydrogen phosphate 680.0	000
Sodium chloride 7947.	000

Directions:

- 1. Suspend 14.0 gms in 900ml tissue culture grade water with constant, gentle stirring until the powder is completely dissolved. Do not heat the water.
- 2. Add 1.260 gms of Sodium bicarbonate powder or 16.9ml of 7.5% Sodium bicarbonate solution

(TCL013) for 1 litre of solution 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 solution 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 dispense the desired amount of sterile solution into sterile containers.
- 7. Store the liquid solution at ambient temperature and in dark till use.

Material required but not provided:

Tissue culture grade water (TCL010) Sodium bicarbonate (TC230) Sodium bicarbonate Solution (TCL013) 1N Hydrochloric acid (TCL003) 1N Sodium hydroxide (TCL002)

Quality Control:

Appearance

Off-white to creamish white homogenous powder

Solubility

Clear solution at 14.0 gms/L

pН

4.80 - 5.20

Osmolality (mOsm/Kg H₂O)

300.00 -340.00

Toxicity test

Passes

Endotoxin content

NMT 1EU/ml



Storage and Shelf Life:

- 1. All powdered salt mixtures and prepared salt solutions should be stored at ambient temperature. Use before the expiry date. In spite of above recommended storage condition certain powdered salts may show some signs of deterioration /degradation in certain instances. This can be indicated by change in colour, in appearance and presence change particulate matter and haziness after dissolution.
- Preparation of concentrated solutions is not recommended as salt complexes having low solubility may precipitate in concentrated solutions.
- 3. If desired, sterile supplements can be added to the sterile solution observing all sterility precautions. Shelf life of the solution will depend on the nature of supplements added to the solution.

Disclaimer: Revision: 04/2022

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