ProductInformation

HANKS' BALANCED SALTS [HBSS]
Without Sodium Bicarbonate and Phenol Red

Product Number **H1387**
Storage Temperature 2-8°C

Product Description

The search for a synthetic medium to replace serum for maintaining cells in vitro began in the late nineteenth century and continues to this day. Ringer, Locks and Tyrode substituted physiological salt solutions augmented with glucose for serum and thereby laid the foundation for the development of defined media.

As biochemical and analytical techniques have improved, more of the components in serum such as vitamins, hormones, and amino acids have been identified and incorporated into physiological salt solutions, reducing, and in some cases eliminating the concentrations of animal sera required as a medium supplement.

Although there have been many modifications to the original formulas in efforts to produce fully defined media, salt solutions still play an important role in tissue culture. A salt solution's basic function, to maintain the pH and osmotic balance in the medium and to provide the cells with water and essential inorganic ions, is as valuable today as when it was first developed a century ago.

HANKS' BALANCED SALTS [HBSS], Product No. H1387 is one of the balanced salts from Sigma. The selection of a balanced salt 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	g/L
Calcium Chloride•$2H_2O$	0.185
Magnesium Sulfate (anhydrous)	0.09767
Potassium Chloride	0.4
Potassium Phosphate Monobasic (anhydrous)	0.06
Sodium Chloride	8.0
Sodium Phosphate Dibasic (anhydrous)	0.04788
D-Glucose	1.0

Precautions and Disclaimer

REAGENT
For In Vitro Diagnostic Use

Preparation Instructions

Powdered salts 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.

1. 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.
3. 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 0.35 g sodium bicarbonate or 4.7 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 salts 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.

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	clear solution at 1x concentration
pH at RT [without sodium bicarbonate]	6.6 ± 0.3
pH at RT [with sodium bicarbonate]	7.4 ± 0.3
Osmolality [without sodium bicarbonate]	276 mOsm/kg H2O ± 5%
Osmolality [with sodium bicarbonate]	286 mOsm/kg H2O ± 5%
Key Element Analysis by ICAP	Analysis has confirmed that key elements are present at concentrations consistent with the formula.
Endotoxin	≤ 1.0 EU/ml at 1x

Biological Performance Characteristics
The ability of this balanced salt solution to maintain the structural integrity of cells in culture was evaluated using appropriate cell lines. There was no evidence of cytotoxicity or uncharacteristic cell morphology.

References

1. Hanks, J. (1976) Hanks' Balanced Salt Solution and pH Control. Tissue Culture Association Manual. 3, 3.
2. Hanks, J.H. and Wallace, R.E. (1949). Relation of Oxygen and Temperature in the Preservation of Tissues by Refrigeration. Proc. Soc. Exp. Biol. Med. 71, 196-200.

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