

Hanks Balanced Salt Solution Composition

Composition	Concentration (mM)
NaCl	137
KCl	5.4
Na ₂ HPO ₄	0.25
KH ₂ PO ₄	0.44
CaCl ₂	1.3
MgSO ₄	1.0
NaHCO ₃	4.2
C ₆ H ₁₂ O ₆	5.5

Hanks Balanced Salt Solution (HBSS) is a widely used isotonic salt solution that provides a nutrient-rich environment for cells in culture. It is critical for maintaining cell viability and function during various laboratory procedures, including cell transport, dissociation, and storage. This article delves into the composition of HBSS, its utility in biological research, and its preparation and applications.

Overview of Hanks Balanced Salt Solution

Hanks Balanced Salt Solution was developed by Dr. Harry Hanks in the 1960s. It was designed to mimic the ionic composition of extracellular fluid, providing an optimal environment for cell cultures. HBSS is frequently used in various applications, including:

- Cell culture maintenance
- Tissue dissociation
- Transport of cells and tissues
- Washing cells during various cell-based assays

The balanced salt solution is composed of a variety of salts, buffers, and nutrients that collectively support cell growth and maintenance. Understanding its composition is essential for researchers working with in vitro cell systems.

Composition of Hanks Balanced Salt Solution

The composition of Hanks Balanced Salt Solution can vary depending on specific formulations, but a standard HBSS typically includes the following components:

1. Salts

Salts in HBSS provide essential ions that maintain osmotic balance and support cellular functions. The primary salts included in HBSS are:

- Sodium Chloride (NaCl): 8.0 g/L
- Maintains osmotic balance and provides sodium ions essential for cellular activities.
- Potassium Chloride (KCl): 0.4 g/L
- Supplies potassium ions, which are crucial for maintaining the resting membrane potential of cells.
- Calcium Chloride (CaCl₂): 0.1 g/L
- Essential for cell adhesion and signaling, calcium ions play a vital role in various cellular processes.
- Magnesium Sulfate (MgSO₄): 0.1 g/L
- Provides magnesium ions, which are important co-factors for enzymatic reactions within the cell.
- Sodium Bicarbonate (NaHCO₃): 0.2 g/L
- Acts as a buffer, helping to maintain physiological pH in the solution.

2. Buffers

Buffers are critical for maintaining the pH of the solution within a physiological range. In HBSS, the following buffers are commonly used:

- HEPES (4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid): Optional, typically 10-25 mM
- A zwitterionic buffer that helps maintain pH stability, especially when cells are exposed to atmospheric CO₂ levels.
- Sodium Bicarbonate (NaHCO₃): As mentioned earlier, it helps maintain pH levels, particularly in buffered solutions.

3. Nutrients

While HBSS does not contain amino acids or vitamins, it can be supplemented with these components as needed. Some formulations may include:

- Glucose: 1.0 g/L
- Provides an energy source for cells, especially in prolonged culture conditions.
- Amino Acids and Vitamins: Optional
- These may be added to provide additional nutrients for specific cell types.

4. pH and Osmolality

The pH of HBSS is generally adjusted to 7.2-7.4, which is optimal for most mammalian cells. The osmolality is typically around 300 mOsm/kg, making it isotonic with physiological fluids, preventing cell lysis or shrinkage when cells are suspended in the solution.

Preparation of Hanks Balanced Salt Solution

Preparing HBSS in the laboratory requires careful measurement and mixing of the components to ensure the desired concentrations are achieved. Here's a general protocol for preparing 1L of HBSS:

Materials Needed

- Analytical balance
- pH meter
- Magnetic stirrer
- Sterile glass or plastic containers
- Distilled water
- Sodium Chloride, Potassium Chloride, Calcium Chloride, Magnesium Sulfate, Sodium Bicarbonate, and optional components like Glucose and HEPES.

Preparation Steps

1. Weigh the Dry Ingredients: Accurately weigh the required amounts of each salt and any optional components.
2. Dissolve in Water: Add the salts to approximately 800 mL of distilled water and stir until completely dissolved.
3. Adjust the pH: Use a pH meter to check the pH of the solution. If necessary, adjust the pH to 7.2-7.4 using either HCl or NaOH.
4. Make Up the Volume: Once the pH is adjusted, add distilled water to bring the total volume to 1 L.
5. Sterilize the Solution: Filter the solution through a 0.22 μm filter to sterilize it. Alternatively, it can be autoclaved if all components are heat stable.
6. Storage: HBSS can be stored at 4°C for short-term use. For long-term storage, it is recommended to aliquot the solution and freeze it at -20°C.

Applications of Hanks Balanced Salt Solution

Hanks Balanced Salt Solution finds extensive use in various biological and medical applications. Some of the notable applications include:

1. Cell Culture

HBSS is frequently used in maintaining and washing cells during culture. Its isotonic nature ensures

that cells remain healthy and viable. It is particularly useful in:

- Rinsing Cells: Before trypsinization or other dissociation methods.
- Diluting Cells: When preparing cell suspensions for various assays.

2. Tissue Dissociation

HBSS is often used in the dissociation of tissues into single-cell suspensions. Its balanced ionic composition prevents cellular stress during the enzymatic digestion of tissues.

3. Cell Transport

In laboratory settings, HBSS is commonly used for the transport of cells and tissues due to its isotonicity. It helps maintain cell viability during transportation to various experimental setups or analyses.

4. Experimental Assays

Various assays, including cell viability and cytotoxicity assays, utilize HBSS as a medium for incubating cells. The solution provides a suitable environment for cellular interaction with experimental agents.

Conclusion

Understanding the composition and applications of Hanks Balanced Salt Solution is crucial for researchers and lab technicians in the fields of cell biology, biochemistry, and related disciplines. Its carefully balanced ionic environment and nutrient content make it an indispensable tool for maintaining cell viability and functionality in vitro. As research continues to evolve, the use of HBSS remains pertinent, affirming its role as a cornerstone in biological research methodologies.

Frequently Asked Questions

What is Hank's Balanced Salt Solution (HBSS) used for?

Hank's Balanced Salt Solution is primarily used in cell culture and biological research as a nutrient solution that maintains the osmotic balance and provides essential ions and nutrients for cells.

What are the main components of Hank's Balanced Salt

Solution?

HBSS typically contains sodium chloride, potassium chloride, calcium chloride, magnesium sulfate, sodium bicarbonate, glucose, and sodium phosphate, all of which help maintain the physiological pH and osmotic balance.

Is Hank's Balanced Salt Solution isotonic?

Yes, Hank's Balanced Salt Solution is isotonic, which means it has a similar osmotic pressure to that of physiological fluids, making it suitable for maintaining cell viability during culture and experimentation.

Can Hank's Balanced Salt Solution be used for washing cells?

Yes, HBSS is often used to wash cells prior to trypsinization or other treatments in cell culture, as it helps remove serum components and maintains cell integrity.

What is the significance of glucose in Hank's Balanced Salt Solution?

Glucose serves as an energy source for cells in Hank's Balanced Salt Solution, supporting metabolic activity and overall cell health during culture procedures.

Are there variations of Hank's Balanced Salt Solution?

Yes, there are several formulations of Hank's Balanced Salt Solution, including those with or without calcium and magnesium, and some that are adjusted for specific cell types or experimental conditions.

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Howard considered Hanks for the role of the main character's wisecracking brother, which eventually went to John Candy. Instead, Hanks landed the lead role and the film went on to become a surprise box office success, grossing more than \$69 million.

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Chet Hanks - Biography - IMDb

Chet Hanks was born on August 4, 1990 in Los Angeles, California, USA. He is an actor, known for Indiana Jones and the Kingdom of the Crystal Skull (2008), Larry Crowne (2011) and Fantastic Four (2015).

Colin Hanks - Biography - IMDb

Biography Colin Lewes Hanks is an American actor. He was born in Sacramento, California, to actors Samantha Lewes and Tom Hanks. Colin is best-known for his work as "Jack Bailey" in the series, The Good Guys (2010) and as "Alex Whitman" in Roswell (1999).

Cast Away (2000) - Full cast & crew - IMDb

Dan Plum stunts Jon Roseman stunt double: Tom Hanks Dennis Scott stunts Jennifer Watson-Johnston

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Cast Edit (in credits order) verified as complete Tom Hanks Josh Elizabeth Perkins Susan

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Explore the precise composition of Hanks Balanced Salt Solution and its importance in cell culture. Learn more about its applications and formulation details!

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