L-Glutamine

(From non-animal source)
Cell Culture Tested

Product Code: TC243

Product Description:

Molecular Weight: 146.14
Molecular Formula: $H_2NCOCH_2CH_2CH(NH_2)CO_2H$
CAS No: 56-85-9
Synonym: (S)-2, 5-Diamino-5-oxopentanoic acid,
L-Glutamic acid 5-amide

L-Glutamine is polar, hydrophilic, essential α-amino acid coded by codons CAA and CAG. It is chemically acidic in nature. It is an aminated analog of L-Glutamate.

L-Glutamine and L-Glutamate are inter-convertible amino acids. L-Glutamine is synthesized from ammonium and glutamate with the help of enzyme glutamine synthetase. This reaction consumes energy in form of ATP. On the other hand, catabolism of L-Glutamine leads to formation of glutamate and ammonium. This reaction is catalyzed by mitochondrial enzyme glutaminase. Ammonium ions generated in this reaction are converted to urea under *in vivo* conditions. However, under *in vitro* environment ammonium is not converted to urea. It accumulates in extracellular environment and can be toxic for cells. When L-Glutamine is present as an amino acid residue in proteins or peptides it is stable; however when it is present in its free form, it is unstable.

Owing to instability of L-Glutamine in liquid media and accumulation of toxic ammonia in culture system, concentration of L-Glutamine should be optimized for every type of cell culture process.

In cell culture systems, L-Glutamine serves as a crucial component of culture media. It is used extensively in wide range of media including classical media, serum-free media, insect cell culture media and media for hybridoma culturing.

It plays many important roles in cell culture. Some of them are mentioned below:

- **Energy source**

L-Glutamine serves as an alternative source of energy for rapidly dividing cells and cells that use glucose inefficiently. L-Glutamine is readily available amino acid for energy production when carbohydrate energy source is not available or deprived in culture medium.

- **Reservoir of nitrogen for synthesis of proteins, nucleic acid and other nitrogenous compounds:**

L-Glutamine is converted to ammonium and pyroglutamate inside the cells. Cells utilize these ammonium ions for formation of amine of glutamate or amide of glutamine. These two amino acid derivatives serve as primary reservoirs of nitrogen for synthesis of proteins, vitamins, nucleic acids and other nitrogenous compounds such as NAD, NADH.

- **Precursor of glutamate:**

L-Glutamic acid generated from catabolism of L-Glutamine serves as a precursor for synthesis of alpha amino acids.

- **Growth-limiting amino acid:**

L-Glutamine acts as a growth-limiting amino acid in culture media and hence is required at a 5- to 20-fold greater concentration than other amino acids.

Directions:

Preparation instructions:

L-Glutamine is soluble in water. It is insoluble in alcohol and ether. L-Glutamine solutions cannot be autoclaved as it is temperature-sensitive. They are sterilized by filtering

through a sterile membrane of porosity 0.22 micron or less.

Recommended concentration of L-Glutamine ranges from 0.5 to 10mM depending on type of medium. Commonly used concentrations of L-Glutamine in some of the animal cell culture media are mentioned in the table:

Name of the Medium	Concentration of L-Glutamine (mM)
AME's Medium	0.5
BME	2
DMEM	4
MEM	2
Leibovitz's L-15	2.054
DMEM-F12	2.5
McCoy's 5A	2
MCDB	4 - 10
IMDM	4
GMEM	2
RPMI	2.054

Insect cells require higher concentration of amino acids as compared animal cells. Hence L-Glutamine concentration for insect cell culture media ranges from 4 to 12.3mM.

To avoid decomposition of L-Glutamine during storage sterile stock solutions of L-Glutamine should be added in the medium just prior to use.

Quality Control:

Appearance
White odorless crystalline powder having a slightly sweet taste

Solubility
33.3mg soluble in 1ml water

pH (2.5% in water at 25°C)
4.00 - 6.00

Chloride (Cl)
NLT 0.02%

Heavy metals (as Pb)
NLT 0.001%

Loss on drying
NLT 0.2% (at 80°C for 3 hours)

Ammonium (NH_4)
NLT 0.1%

Iron (Fe)
NLT 0.001%

Arsenic (As)
NMT 0.001%

Specific rotation [alpha]
C=4 in water at 20°C+6.3° to +7.3°

Sulphate (SO4)
NLT 0.02%

Residue on ignition (as sulphate)
NLT 0.1%

Assay (On dry basis)
NLT 99%

Fourier Transform Infrared Spectrometry (FTIR)
Standard Pattern

Ninhydrin Positive Substances
Conforms

Residual Solvents USP467
Absent

Cell Culture Test
Passes

Storage and Shelf Life:

Store L-Glutamine powder at 10-30 °C.
Shelf life is 48 months.
L-Glutamine powder is more stable than L-Glutamine solution. L-Glutamine solution is unstable at physiological pH in liquid media. The rate and extent of degradation of L-Glutamine in liquid media depends on pH, temperature, presence of bicarbonate, phosphate and duration of storage. In fixed phosphate concentration, degradation increases when pH rises from 4.3 to 10.
Increase in temperature above frozen range (above -20°C) as well as prolonged storage of Glutamine solution at 2-8°C leads to degradation of L-Glutamine. L-Glutamine solution remains stable for 2 weeks at 2-8°C.
Stock solutions should be aliquoted in sterile bottles and stored at -20°C.
Use before expiry date given on the product label.

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