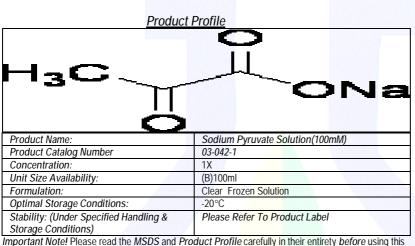


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<u>Important Note!</u> Please read the <u>MSDS</u> and <u>Product Profile</u> carefully in their entirety <u>before</u> Product Description:

Sodium Pyruvate like Glutamine and Glucose is considered an important constituent of most media and is now recognized as an easily accessible additional carbohydrate energy source. Pyruvate, an anion of pyruvic acid, is the end product of glycolysis in which organisms break down glucose into lactic acid in the absence of molecular oxygen for the purpose of obtaining chemical energy. The glucose is converted to pyruvate with the production of Adenosine Triphosphate (ATP). Glycolysis is just one of several pathways used by different species to degrade glucose anaerobically. In the mitochondria of aerobic organisms, pyruvate is converted to Accetyl-CoA which in turn is completely oxidized to Carbon Dioxide (CO₂). Acetyl CoA is not only the initiator for the Krebs cycle, it is also formed during the metabolism of Fats, certain Amino Acids and also is utilized in the biosynthesis of a variety of larger molecules. The Krebs cycle doesn't consume energy; it produces it most efficiently. The cycle is fed by pyruvic acid from this anaerobic glycolysis pathway. Glycolysis releases energy and part of that energy goes to the conversion of ATP where it is stored. ATP provides the energy which drives cellular metabolic reactions and is considered the most important high-energy compounds in cells. Approximately 70% of the energy in the ATP comes from Carbohydrate Metabolism.

In essence, cells in culture have two primary paths of energy (ATP) production:

- Conversion of Glucose to Lactate(or full oxidation to CO₂)
- Oxidation of L-Glutamine to CO₂ and Ammonia

During aerobic metabolism, the pyruvic acid is integrated into the Krebs cycle and oxidized to CO₂. However when oxygen is not present in sufficient quantities, Pyruvate is metabolized into lactate which can have deleterious effects on cells in culture. Culture media are buffered to compensate for the cellular production of CO₂ and lactic acid as untoward metabolic by-products.

In Mammalian cell culture, the growth of cells is dependant upon the presence of a carbon and energy source. In most of the commonly used cell culture media, glucose and glutamine are those major energy sources. Whereas Glucose is essential for continuous cell attachment to the microcarrier and cell rate of growth, L-Glutamine provides a significant portion of the energy to maintain cell growth. Media containing Glucose should be supplemented with pyruvate for cellular growth especially under conditions of low density. A variety of other compounds including pyruvate appear in complex media especially when serum is reduced and may assist in cloning and in maintaining specialized cells. Recent research has indicated that different pathways for Glutamine Metabolism are possible resulting in not only different energy output, but also however, with resultant ammonia accumulation. These by-products can limit cell growth and product formation. Reducing ammonia accumulation by replacing of Glutamine with Pyruvate supported cell growth without adaptation for at least 19 passages without a reduction in growth rate of different adherent commercial cell lines(e.g.MDCVK,BHK21,CHO-K1) in both serum-containing and Serum-Free Media.⁴

We now should be able to see part of the relationship between the CO₂, Lactic Acid, Glucose, Glutamine and Pyruvate and proper physiological pH and the crucial roles each one plays in cell culture.

Cultured cells require a sterile environment and an optimal nutrient supply for growth and viability. Over the years variously defined media have been designed, developed, modified and enriched with a wide spectrum of supplementation for supporting a vast range of cell types. Precise media formulations have been specifically developed by optimizing the concentrations of each and every component which performs a uniquely defined function.

Biological Industries, Kibbutz Beit Haemek 25115 Israel Telephone: 972-4-9960-595 Fax: 972-4-9968-896 Web Site: www.bioind.com E-M

<u>E-Mail: info@bioind.com</u>

Biological Industries(BI)

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Some Predominant Characteristics of Sodium Pyruvate Solution include:

- Frozen Formulation
- 100mM Concentration
- Sterile-Filtered
- Cell-Culture Tested
- Endotoxin-Tested

Instructions:

The product should be stored at -20°C. The medium should bethawed to room temperature prior to use. The product should not be left in the light for prolonged periods as it is light-sensitive. When stored in the dark under ideal conditions, the product is stable until the expiry date. Recommended Dilution: 1:100 For Most Cell Cultures

Procedure:

- Take a bottle from the proper storage conditions at -20°C, read the label., thaw and aliquot. 1)
- 2) Ensure that the cap of the bottle is tight.
- 3)
- Gently swirl the solution in the bottle. Wipe the outside of the bottle with a disinfectant solution such as 70% ethanol. 4)

5) Using aseptic/sterile technique under a laminar-flow culture hood, work according to established protocols.

Quality Control:	
Test	Specification
Cell Culture Test	Test & Record
Cell Line	Vero
Endotoxins	Test and Record
Osmolality	180-2 <mark>00 mOs</mark> m/Kg
рН	5.0-7.0
Sterility	Sterile

Auxiliary Products

Auxiliary Floducis.		
Product Name	Catalog Number	Storage Temperature
Basal Medium Eagle (BME), Earle's Salts Base, without L-Glutamine, without Sodium Bicarbonate 10X	01-015-5	2-8°C
Minimum Essential Medium Eagle (MEM-E), Earle's Salts Base, without L-Glutamine	01-025-1	2-8°C
Medium-M-199(Earle's), Earle's Salts Base, with L-Glutamine	01-080-1	2-8°C
Medium-M-199 10X Conc., (Earle's), Earle's Salts Base, with L- Glutamine, without Sodium Bicarbonate	01-080-5	2-8°C
Earle's Balance Salt Solution 10X Conc., without Sodium Bicarbonate	02-010-5	Room Temperature (15-30°)
Earle's Balance Salt Solution without Phenol Red	02-011-1	Room Temperature (15-30°)
Earle's Balance Salt Solution without Phenol Red, without Sodium Bicarbonate	02-011-5	Room Temperature (15-30°)
L-Glutamine Solution 29.2mg/ml in Saline	03-020-1	-20°C
L-Alanyl-L-Glutamine Solution(A Dipeptide Substitute)	03-022-1	-20°C
Penicillin-Streptomycin Solution,10,000 units/ml Penicillin G Sodium Salt,10mg/ml Streptomycin	03-031-1	-20°C
Sterile Culture-Grade Water	03-055-1	Room Temperature (15-30°)
Serum-Free Cell Freezing Medium	05-065-1	2-8°C
<u>Note</u> : For a list of other Antibiotics, Serum, Reagents and Supplements, please refer to our Product Catalog/Product Profiles, Product Guides and Internet Site.		

References:

1) **Current Edition Merck Index**

- 2) Biological Industries(BI) Specifications
- Darling, D.C. and Morgan, S.J. Animal Cells: Culture and Media, New York: John Wiley & Sons, 1994 3)
- Genzel, Yvonne, Ritter, Joachim B. et. al. "Substitution of Glutamine By Pyruvate To Reduce Ammonia Formation..." Cell Culture 4) Engineering IX.

Biological Industries, Kibbutz Beit Haemek 25115 Israel Telephone: 972-4-9960-595 Fax: 972-4-9968-896

Web Site: www.bioind.com

E-Mail: info@bioind.com