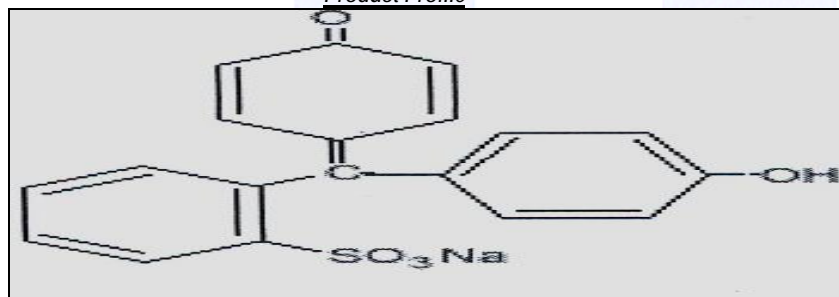


### Product Profile



|   |                               |
|---|-------------------------------|
| Product Name:                               | Phenol Red Solution           |
| Product Catalog Number:                     | 03-100-1                      |
| Unit Size Availability:                     | 100ml                         |
| Formulation:                                | Liquid                        |
| Defined Storage Conditions:                 | 15-30°C (Room Temperature)    |
| Stability: (Under Ideal Handling & Storage) | Please Refer To Product Label |

**Important Note!** Please read the *MSDS* and *Product Profile* carefully in their entirety *before* using this material for possible safety precautions and potential hazards.

#### Product Description:

Most commercial culture media include Phenol Red Sodium Salt as a pH indicator. As cells undergo metabolic processes especially that of glucose, acids and CO<sub>2</sub> (e.g. lactic, pyruvic) are produced with a resultant pH decrease. As a pH indicator, Phenol Red assists in rapidly identifying even superficial changes from neutral to acidic pH values by measuring gradual nutrient depletion. Cultured cells require not only a sterile environment but also unique niche nutrient requirements for growth. In addition, the cell culture environment must maintain its stability to optimally grow and this is where temperature and pH come to the fore. Most cells require pH conditions within a 7.2-7.4 range and controlling pH is a key, essential factor for creating the most optimum cell culture milieu. There are major variations to this optimum, as the ideal pH at culture initiation should be nearer to 7.4, but should not fall below 7.0 during the culture. A pH below 6.8 usually inhibits cell growth. Continuous transformed cell lines (e.g. Hybridoma Cell Lines) prefer more of an acidophilic milieu (7.0-7.4). Thus, the regulation of pH is critical immediately following cell seeding when the new culture is acclimatizing itself to its new environment. This is usually achieved by one of two buffering systems:

- The normal buffer system from the cell culture media constituents analogous to the blood in which the gaseous CO<sub>2</sub> stabilizes with the CO<sub>3</sub>/HCO<sub>3</sub> content
- Improved buffering and pH stability using a zwitterion buffer such as HEPES either in addition to or instead of bicarbonate

In basal media for instance, the salts are primarily responsible for providing physiological pH among other factors such as osmotic pressure and membrane potential. Generally, culture media are buffered to compensate for the cellular production of CO<sub>2</sub> and lactic acid as by-products of cellular metabolism. The traditional role of buffering culture media is with bicarbonate (HCO<sub>3</sub>). As cells grow and generate CO<sub>2</sub> naturally, an accumulation of CO<sub>2</sub> in the headspace will prevent CO<sub>2</sub> diffusing out of the medium with a resultant increase in weakly-dissociated NaHCO<sub>3</sub> producing an excess of H<sup>+</sup> ions and a concomitant decrease in pH. When this CO<sub>2</sub> is dissolved, it forms a buffering system with the bicarbonate. If a situation exists where cell density is low or the cells have entered a lag phase, they may not produce enough CO<sub>2</sub> to maintain optimal pH levels. In order to counter such problems, bicarbonate-buffered media require the use of incubators with a 5-10% CO<sub>2</sub> atmosphere. Some media with lower bicarbonate levels (e.g. MEM) or with higher levels (DMEM) require ~5% CO<sub>2</sub> and ~10% CO<sub>2</sub> respectively to maintain pH. The most important factor being the correct percent CO<sub>2</sub> based upon the particular medium's bicarbonate level, irrespective of cell type.

A Phenol Red Solution changes from a yellowish color to a reddish color within a pH range of 6.8-8.4. A yellowish colored solution is indicative of a pH of 6.4 or below and a reddish color is indicative of a pH of 8.2 and above. Cell culture media turns acidic as it becomes exhausted as the acid end products from carbohydrate fermentation lower the pH causing the Phenol Red to turn from a reddish-purple hue (alkaline) to a yellowish hue (acid). The pH status of the medium is continually monitored as indicated by the color. The breakdown of carbohydrates during fermentation may occur especially by contamination by microorganisms such as bacteria and yeasts yielding incompletely oxidized products. Some forms of fermentation may occur in the absence of oxygen in which case ATP is generated in reaction pathways when organic compounds act as both acceptors and donors of electrons. Factors affecting pH stability of the cell culture medium include:

- ✚ Glucose Concentration
- ✚ Headspace Above the Medium to Prevent CO<sub>2</sub> Loss and an Increase in Hydroxyl Ions
- ✚ Buffer Capacity & Type

Phenol Red, under certain circumstances, may not only act as a weak estrogen with human breast cancer cells in culture but also may interfere with luminescence assays. Lipophilic impurities, not the Phenol Red Dye *per se* accounts for this estrogenic activity. A change in color is usually indicative therefore that the culture medium must be changed or replenished appropriately. When Phenol Red is added as a component to cell culture media, it may be autoclaved with no untoward effects.

*Some of the Predominant Characteristics of Phenol Red Solution include:*

- pH Indicator
- Liquid Formulation
- Easy-To-Use
- For Cell Culture Applications

#### *Instructions/Procedure*

For optimal cell-line performance, the supplementation and use of the Chemical Raw Material, *Phenol Red Solution* for cell-culture media is usually added *prior to* using the liquid cell-culture media *per se*. Supplements may be added *prior to* filtration or *introduced aseptically* to sterile medium. Sterile solutions may be prepared by sterile filtration utilizing a 0.2µm filter. Solutions are stable @ 37° for 4-5 days. For long-term storage, aqueous stock solutions must be stored at 15-30°C. The nature of the supplement may not only affect storage conditions but the shelf-life of the medium as well. The biochemical raw material product should be stored at 15-30°C. The contents 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.

#### *Storage & Stability*

This product should be stored under specified conditions (15-30°C) and used within two (2) years. Powdered media are extremely hygroscopic and should be protected from atmospheric moisture. Do not use after the expiration date as specified on the label. Deterioration of the powdered medium may be recognized by any or all of the following factors, among others including: 1). Clumping and/or granulation and 2). Color change.

As the selection of a nutrient medium or supplementation thereof is strongly influenced, among others, by many factors, of note are three major considerations:

- ◇ Cell Type
- ◇ Type of Culture(e.g., Clonal, Monolayer, Suspension)
- ◇ Degree of Chemical Definition

It is recommended to review the extensive literature concerning cell-culture media and its supplementation and the physiological parameters required for each specific cell-line as per their essential requirements.

#### *Quality Control*

| Test        | Specification      |
|-------------|--------------------|
| Appearance: | Red-Colored Liquid |
| Sterility:  | Sterile            |

#### *Auxiliary Products*

| Product Name  | Catalog Number | Storage Temperature       |
|---|----------------|---------------------------|
| Dulbecco's Phosphate Buffered Saline(DPBS) without Calcium and Magnesium  | 02-023-1       | Room Temperature (15-30°) |
| Amphotericin B 250 micrograms/ml  | 03-028-1       | -20°C                     |
| Amphotericin B 2500 micrograms/ml   | 03-029-1       | -20°C                     |
| Penicillin-Streptomycin Solution  | 03-031-1       | -20°C                     |
| Penicillin-Streptomycin 10X Solution  | 03-031-5       | -20°C                     |
| Penicillin-Streptomycin Nystatin Solution   | 03-032-1       | -20°C                     |
| Trypan Blue Solution  | 03-102-1       | Room Temperature (15-30°) |
| Penicillin G Sodium Salt Cell Culture-Tested Biochemical  | 41-501-        | 2-8°C                     |
| Phenol Red Sodium Salt Cell Culture-Tested Biochemical  | 41-902-        | Room Temperature (15-30°) |
| <b>Note:</b> For a list of Serum, other Antibiotics, or Biological Industries' Products Reagents and Supplements, please refer to our Product Catalog/Product Profiles/ Product Guides and Internet Site. |                |                           |

#### *References:*

- 1) 14th Edition Of Merck Index, pps.1224,1514
- 2) Current Editions USP/E Ph
- 3) Biological Industries(BI )Specifications
- 4) Martindale, *The Extra Pharmacopeia*, 28th Edition, Royal Pharmaceutical Society: London, England pps.1076-1086.
- 5) Barile, Frank A. *Clinical Toxicology: Principles and Mechanisms*, CRC Press: Boca Raton, Florida, 2004.
- 6) Homburger, Freddy, Hayes, John A. and Pelikan, Edward W. *A Guide To General Toxicology*, Karger Press: Basel, Switzerland, 1984, pps.101-102
- 7) Hansel, Donna E. and Dintzis. *Pathology*, Lipponcott, Williams & Wilkins Press: Baltimore, Maryland, 2006