Phenol Red Broth Base

**Intended Use:**
A basal medium to which carbohydrates are added for determination of fermentation reactions of pure cultures of microorganisms.

**Composition**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Gms / Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proteose peptone</td>
<td>10.000</td>
</tr>
<tr>
<td>HM peptone B #</td>
<td>1.000</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>5.000</td>
</tr>
<tr>
<td>Phenol red</td>
<td>0.018</td>
</tr>
<tr>
<td>Final pH ( at 25°C)</td>
<td>7.4±0.2</td>
</tr>
</tbody>
</table>

**Directions**
Suspend 16.02 grams in 1000 ml purified / distilled water, mix well. Heat if necessary to dissolve the medium completely. Mix well and dispense in fermentation tubes (tubes containing inverted Durham's tubes). Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Aseptically add filter sterilized or autoclave sterilized carbohydrate solution to sterile basal medium.

**Principle And Interpretation**
Phenol Red Broth Medium is formulated as per Vera (8) and is recommended to determine the fermentation reaction of carbohydrates for the differentiation of microorganisms (1,2,6). Phenol Red Broth Medium with various added carbohydrates serves as a differential medium by aiding in differentiation of various species and genera by their ability to ferment specific carbohydrate, with the production of acid or acid and gas (7). Phenol Red Broth Base is a complete medium without added carbohydrate, which can be used with the addition of 5-10 %, desired carbohydrate. It is used as a negative control for studying fermentations or as a base for the addition of carbohydrates. Proteose peptone and HM peptone B serve as sources for carbon and nitrogen. Sodium chloride is the osmotic stabilizer. Phenol red is the pH indicator, which turns yellow at acidic pH. Gas formation is seen in Durhams tubes. All of the Enterobacteriaceae grow well in this medium. In addition to producing a pH colour shift, the production of mixed acids, notably butyric acids, often results in a pungent, foul odour from the culture medium (5).

**Type of specimen**
Isolated Microorganisms

**Specimen Collection and Handling**
For isolated Microorganisms samples follow appropriate techniques for handling specimens as per established guidelines. After use, contaminated materials must be sterilized by autoclaving before discarding.

**Warning and Precautions :**
Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

**Limitations :**
1. In addition to producing a pH colour shift, the production of mixed acids, notably butyric acids, often results in a pungent, foul odour from the culture medium (1)

**Performance and Evaluation**
Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.
Quality Control

Appearance
Light yellow to pink coloured homogeneous free flowing powder

Colour and Clarity of prepared medium
Red coloured clear solution without any precipitate

Reaction
Reaction of 1.6% w/v aqueous solution at 25°C. pH : 7.4±0.2

pH
7.20-7.60

Cultural Response
Cultural characteristics observed after an incubation at 35 - 37°C for 18 - 24 hours.( longer if necessary)

<table>
<thead>
<tr>
<th>Organism</th>
<th>Inoculum (CFU)</th>
<th>Growth</th>
<th>without carbohydrate, (Acid)</th>
<th>without carbohydrate, (Gas)</th>
<th>with dextrose, (Acid)</th>
<th>with dextrose, (Gas)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Citrobacter freundii ATCC 8090</em></td>
<td>50-100</td>
<td>luxuriant</td>
<td>Negative reaction, no colour change</td>
<td>Negative reaction</td>
<td>Positive reaction, yellow reaction colour</td>
<td>Positive reaction, yellow reaction colour</td>
</tr>
<tr>
<td><em>Escherichia coli ATCC 25922</em> (00013*)</td>
<td>50-100</td>
<td>luxuriant</td>
<td>Negative reaction, no colour change</td>
<td>Negative reaction</td>
<td>Positive reaction, yellow reaction colour</td>
<td>Positive reaction, yellow reaction colour</td>
</tr>
<tr>
<td>#Klebsiella aerogenes ATCC 13048 (00175*)</td>
<td>50-100</td>
<td>luxuriant</td>
<td>Negative reaction, no colour change</td>
<td>Negative reaction</td>
<td>Positive reaction, yellow reaction colour</td>
<td>Positive reaction, yellow reaction colour</td>
</tr>
<tr>
<td><em>Klebsiella pneumoniae ATCC 13883 (00097</em>)*</td>
<td>50-100</td>
<td>luxuriant</td>
<td>Negative reaction, no colour change</td>
<td>Negative reaction</td>
<td>Positive reaction, yellow reaction colour</td>
<td>Positive reaction, yellow reaction colour</td>
</tr>
<tr>
<td><em>Proteus vulgaris ATCC 13315</em></td>
<td>50-100</td>
<td>luxuriant</td>
<td>Negative reaction, no colour change</td>
<td>Negative reaction</td>
<td>Positive reaction, yellow reaction colour</td>
<td>Positive reaction, yellow reaction colour</td>
</tr>
<tr>
<td><em>Salmonella Typhi ATCC 6539</em></td>
<td>50-100</td>
<td>luxuriant</td>
<td>Negative reaction, no colour change</td>
<td>Negative reaction</td>
<td>Positive reaction, yellow reaction colour</td>
<td>Positive reaction, yellow reaction colour</td>
</tr>
<tr>
<td><em>Salmonella Typhimurium ATCC 14028 (00031</em>)*</td>
<td>50-100</td>
<td>luxuriant</td>
<td>Negative reaction, no colour change</td>
<td>Negative reaction</td>
<td>Positive reaction, yellow reaction colour</td>
<td>Positive reaction, yellow reaction colour</td>
</tr>
<tr>
<td><em>Serratia marcescens ATCC 8100</em></td>
<td>50-100</td>
<td>luxuriant</td>
<td>Negative reaction, no colour change</td>
<td>Negative reaction</td>
<td>Positive reaction, yellow reaction colour</td>
<td>Positive reaction, yellow reaction colour</td>
</tr>
<tr>
<td><em>Shigella flexneri ATCC 12022 (00126</em>)*</td>
<td>50-100</td>
<td>luxuriant</td>
<td>Negative reaction, no colour change</td>
<td>Negative reaction</td>
<td>Positive reaction, yellow reaction colour</td>
<td>Positive reaction, yellow reaction colour</td>
</tr>
</tbody>
</table>

Key : (*) Corresponding WDCM numbers.  
(#) Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life
Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal
User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

Please refer disclaimer Overleaf.
Reference


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