B-156          PRICE’S GIEBMSA STAIN

METHOD

Fixation:  10% Buffered Neutral Formalin or Zenker’s. Use no fixation technique that will destroy erythrocytes.

Sections:  Cut paraffin @ 4 microns

Staining:
1. Deparaffinize with two changes at 2 minutes each in xylene. Rinse in absolute alcohol for two changes, 2 minutes each.
2. Rinse in 95% Alcohol for two changes after a one minute immersion. If necessary to further remove mercuric chloride crystals, use an Alcoholic Iodine Solution (B-156-4), which will be further removed in successive alcohols.
3. Rinse through 80%, 70%, 50%, alcohols for one change, 1 minute each.
4. Rinse in distilled water for 15 seconds.
5. Buffer in the Phosphate Buffer Solution, (B-156-2) for 30 minutes.
6. Stain in the Giemsa Working (B-156-1A) overnight.
   To prepare Giemsa Working:
   - Giemsa Stock (B-156-1)…………………3 parts
   - Phosphate Buffer pH 7.0 (B-156-2)….97 parts
7. Rinse in the buffer solution.
8. Rinse in Acetic Acid, 0.2% (B-156-3), for one minute, absolute alcohol for two changes, 15 seconds each. This step should take only 90 seconds!!!
9. Dip in xylene for two changes, two minutes each.

Cells:

<table>
<thead>
<tr>
<th>Color</th>
<th>Stain</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blue</td>
<td>Malarial parasites</td>
</tr>
<tr>
<td>Blue</td>
<td>Tissue Nuclei</td>
</tr>
<tr>
<td>Dark Blue</td>
<td>Bacteria</td>
</tr>
<tr>
<td>Black</td>
<td>Malarial Pigment</td>
</tr>
<tr>
<td>Blue</td>
<td>Schistosomic egg shells</td>
</tr>
<tr>
<td>Pale Pink</td>
<td>Collagen, etc.</td>
</tr>
<tr>
<td>Pink-Rose</td>
<td>Erthrocytes</td>
</tr>
</tbody>
</table>

References:
  Publications, c. 1968, p. 127