This procedure is available in kit form from:
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cat# cy018

ACRIDINE ORANGE FLUORESCENT TECHNIQUE

SOLUTIONS:

1. Acridine Orange Stock Solution
2. Sorensen's Phosphate Buffer pH 6.0
3. Calcium Chloride 0.1M Solution
4. Acetic Acid 1% Aqueous

STAINING PROCEDURE:

1. Hydrate rapidly through graded Ethyl Alcohols – 80%, 70%, 50%, to distilled water.
2. Rinse briefly in Acetic Acid 1% Aqueous.
3. Wash in distilled water.
4. Stain in Acridine Orange Working Solution (1 part Acridine Orange Stock to 9 parts Sorensen's Phosphate Buffer pH 6.0) for 3 minutes.
5. Transfer to Sorensen's Phosphate Buffer pH 6.0 for at least 1 minute to remove excess dye. If batches of slides are processed, they may remain in buffer for several hours while they are examined successively.
6. Differentiate 1 to 2 minutes in Calcium Chloride 0.1M Solution until nuclei (especially of leukocytes) show bright green fluorescence. The time for differentiation can be standardized by trial.
7. Rinse ith Sorensen's Phosphate Buffer pH 6.0 by using a polyethylene wash bottle.
8. Mount wet, using a few drops of Sorensen's Phosphate Buffer pH 6.0 under a coverslip, and examine.

REMARKS: Note: The time of the procedure is approximately 6 to 7 minutes. After microscopic examination, slides can be destained by placing them in 50% Ethyl Alcohol and then restained by the Papanicolaou Technique or other method.

RESULTS:

Mature superficial squamous cells.....Green fluorescence
Intermediate Squamous Cells..... Brownish
Parabasal and Basal Cells.....Brown or Reddish Brown
Sheets of Atrophic Squamous Cells .....Brownish-Orange to Red
Normal Endocercial Cells ..... Reddish Brown
Malignant Cells.....Generally Flaming Orange to Red with Greenish – Yellow Nuclei