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Parasitology

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Examination of Thin Blood Films for malaria

1. Rapid Field's stain for thin films

This is a modification of the original Field's stain to enable **rapid** staining of **fixed** thin films. This method is suitable for malaria parasites, Babesia sp., Borrelia sp. and Leishmania sp.

Method.

a. Air dry the film
b. Fix in methanol for 1 minute.
c. Flood the slide with 1 ml of **Field's stain B, diluted 1 in 4 with distilled water.**
d. **Immediately,** add an equal volume of **undiluted** Field's stain A, mix well and allow to stain for 1 minute.
e. Rinse well in tap water and drain dry.

Uses.

This is a useful method for rapid presumptive species identification of malarial parasites. It shows adequate staining of all stages including stippling (mainly Maurer's clefts). However , staining with Giemsa is always the method of choice for definitive species differentiation.

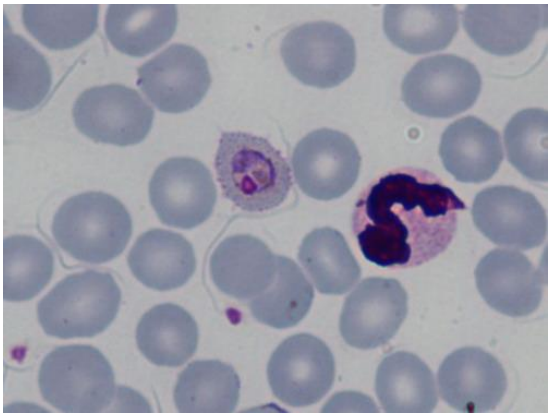
2. Giemsa stain for thin films.

Method.

a. Air dry thin films
b. Fix in methanol for 1 minute
c. Wash in tap water and flood the slide with Giemsa diluted **1 in 10** with buffered distilled water **pH 7.2.** The diluted stain **must be freshly prepared each time.**
d. Stain for 25 - 30 minutes.
e. Run tap water on to the slide to float off the stain and to prevent deposition of precipitate on to the film. Drain dry vertically.
f. Examine the film using the x100 objective.

Notes on the stained film.

a. Examine the tail end of the slide where the red cells are separated into a one-cell-layer thick.
b. An alkaline buffer pH 7.2 is vital for clear differentiation of nuclear and cytoplasmic material and to visualise inclusions such as Schuffner's/James's dots in the red cells. **Acidic buffer is unsuitable.**
c. The red cells are fixed in the thin film so the morphology of the parasitised cells and the parasites can be seen.
d. On a well stained film the chromatin stains red/purple and the cytoplasm blue. Leucocytes have purple nuclei, the red stippling , if present should be clearly visible.



Trophozoite of *Plasmodium ovale*