

Giemsa Staining

Reagents Needed:

Giemsa Stain (Stock) RICCA CHEMICAL COMPANY Cat. No. 3250
Buffer Solution, Giordano, pH 6.4 (M/15), for Wright Staining RICCA CHEMICAL COMPANY Cat. No. 1450

Thin Film for Blood Smears

Recommended Method:

1. Dilute 1 mL of Giemsa Stain Stock with 10 mL - 20 mL high purity Water.
2. Prepare and immediately air dry smears (blood films) of capillary or fresh venous blood without anticoagulants on slides or coverslips in the usual manner.
3. Fix dried smears in absolute Methyl Alcohol for 5-7 minutes. Air dry.
4. Soon after fixing, immerse dried blood films in the diluted Giemsa Stain for 15 to 50 minutes. For intensive staining of material for Donovan bodies, spirochetes, the undulating membranes of trypanosomes, etc., leave in the diluted stain for 1-4 hours.
5. Wash well with neutral high purity water.
6. Allow to air dry and examine.

Satisfactory Staining Results:

Erythrocytes: orange-pink to rose

Neutrophils: deep blue-violet nuclei, purple to lilac granules, pink cytoplasm

Eosinophils: deep blue-violet nuclei, orange to pink granules

Basophils: deep blue-violet nuclei, deep blue to violet granules

Mast Cells: deep blue-violet nuclei, deep blue-violet granules

Lymphocytes: deep blue-violet nuclei, light blue cytoplasm

Monocytes: light bluish-purple nuclei, pale gray-blue cytoplasm

Platelets: central red-purple granule surrounded by a light blue halo

Plasmodia (Malarial parasites): red chromatin dot (nucleus); blue cytoplasm; brown, greenish-brown or black intraparasitic pigment

Thick Film Smears for Malarial Parasites – Accelerated Procedure

Recommended Method:

1. Mix 4 mL Giemsa Stain Stock, 3 mL Acetone, 2 mL Giordano Buffer, and 31 mL high purity water.
2. Prepare thick films by spreading 3-5 drops of fresh non-anticoagulated blood over a circle of about 15 mm diameter on a scrupulously clean slide. The smear must not be too thick or it will flake off. It must allow newsprint to shine through.
3. Allow unfixed thick films to dry 1 hour.
4. Stain unfixed, dried thick films for 5 - 10 minutes in the diluted Giemsa stain.
5. Rinse in distilled water, if necessary. Note: Exposure to water should be very brief to avoid detachment and loss of films.
6. Allow to air dry and examine.

Satisfactory Staining Results:

The Plasmodia (Malarial parasite) chromatin in the nucleus stains clear red and the Plasmodia cytoplasm stains clear blue.

These are typical staining procedures. These reagents may be suitable for other staining procedures. Consult staining reference books or standard operating procedures for other suitable uses of these products.