

Wright-Giemsa Staining

Reagents Needed:

Wright-Giemsa Stain RICCA CHEMICAL COMPANY Cat. No. 9380
Buffer Solution, Giordano, pH 6.4 (M/15), for Wright Staining RICCA CHEMICAL COMPANY Cat. No. 1450

Recommended Method:

1. Prepare and air dry smears (blood films) of capillary or fresh venous blood, or bone marrow, on slides or coverslips in the usual manner.
2. Soon after drying, apply a measured number of drops of undiluted Wright-Giemsa Stain, covering slides completely, with the smears facing upward.
3. Allow 1 - 3 minutes staining time for blood smears, or 5 minutes staining time for bone marrow.
4. Gently add Giordano Buffer of the same quantity as the stain used and mix by blowing gently on the surface. Do not allow stain-buffer mixture to spill off slides.
5. Leave the diluted stain on the slide for twice the undiluted stain time (from step 3).
6. Keeping slides facing upward, flood off the stain and wash well with purified Water until the thin portions of the stained film appear pink to the naked eye.
7. If necessary, remove the stain on the back of the slides by cleaning with alcohol-moistened gauze.
8. Allow slides to air dry by resting an edge on a blotter.
9. Slides may be mounted under a coverslip using Permount.

Satisfactory Staining Results:

A well-stained smear will appear pink macroscopically.

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

Unsatisfactory Staining Results:

Precipitation: should not occur. May be due to insufficient or incorrect washing, allowing the stain to dry on the slide, a dirty slide, dust, or an overconcentrated stain. Use only new, precleaned slides and coverslips.

Excessively blue erythrocytes and dark blue structureless nuclei: May be due to insufficient washing, an overly thick film, overstaining, or excessive alkalinity (high pH) of water, buffer, or stain.

Excessively red erythrocytes and pale gray-blue nuclei: May be due to inadequate staining, prolonged washing, or excessive acidity (low pH) of water, buffer, or stain.

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