

Giemsa Stain, 10X

Catalogue #: CD006
Storage: RT
Packing Size: 100mL

Brief Description:

Giemsa's stain is a member of the Romanowski group of stains, which are defined as being the black precipitate formed from the addition of aqueous solutions of methylene blue and eosin, dissolved in methanol. The variants of the Romanowski group differ in the degree of oxidation (polychroming) of the methylene blue stain prior to the precipitation. The stain class was originally designed to incorporate cytoplasmic (pink) staining with nuclear (blue) staining and fixation as a single step for smears and thin films of tissue (spread preparations of omentum). Minor modifications of working stain concentration and staining time have been made over the years for fixed tissue sections. The Romanowski stains are extremely tedious to prepare, and are best purchased as the commercially available pre-made stock stain. Giemsa stain is used to differentiate nuclear and/or cytoplasmic morphology of a variety of cells including blood cells, platelets, RBCs, WBCs, and parasites. The stain must be diluted for use with water buffered to pH 6.8 to 7.2, depending on the specific procedure used. Either should be tested for proper staining reaction before use. The stock is stable for years in a dry environment and the aqueous working dilution of stain is good only for 1 day.

Experimental Procedures

Preparation of working solution before use:

Giemsa Stain Solution, 10X	10 mL
Methanol	10 mL
Distilled water	80 mL

For tissue staining:

- Bring tissue sections to distilled water
- Stain with diluted Giemsa's stain working solution (made from 10X stock before use) for 6 hrs to overnight
- Quickly Rinse tissue sections with distilled water
- Differentiate with 0.5% aqueous acetic acid for 30 sec
- Dip in 95% ethyl alcohol to clear plastic for 2-3 quick dips
- Dehydrate sections rapidly in air-flow cabinet
- Mount if desired.

For blood film staining:

- Preparation of blood films: Using any of the conventional techniques, smear a small drop of blood on a clean

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microscope slide. Allow to air dry.

- Fix by immersing in absolute methanol for 5 min.
- Apply Giemsa's stain working solution for 30-60 min (depending on thickness of blood film) on a horizontally positioned slide.
- Rinse in Wright-Giemsa Buffer Solution (Cat#: CD044) with three changes, 10 dips each.
- Dry the slide in a tilted position; do not blot-dry.
- Mount a coverglass if desired.

Reference:

1. Horobin RW, Walters K, (1987) The Romanowsky-Giemsa effect in blood smears. *Histochemistry* 86:331.
2. Wittekind DH, Gehring T, (1985) On the nature of the Romanowsky-Giemsa staining and the Romanowsky-Giemsa effect. *Histochemical Journal* 17:263.

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