



Malaria Stain Kit

General Information

Species of Plasmodium, Babesia, Trypanosoma, Leishmania donovani, and microfilariae can be detected in human blood during various stages of their life cycles. Plasmodium and Babesia can be identified within erythrocytes. Leishmania donovani amastigotes are difficult to detect in the peripheral blood but can be identified within the monocytes in concentrated buffy coats. Trypanosomes and some microfilariae may be found in plasma and in buffy coat concentration.

Microscopic examination of Romanowsky stained peripheral blood smears includes examination of thin and thick films. Microfilariae are best identified at low power magnification. Other blood parasites require examination under oil immersion of thick and thin smears.

Trypanosomes are more frequently identified in the thicker portion of thin peripheral blood smears. Plasmodium and Babesia are detected in thin peripheral blood smears but often are more easily identified in the thinner portion of the thin smear.

With the exception of Lyme disease, diagnosis of most patients with borreliosis is usually by detection of spirochetes in the peripheral blood of febrile patients. During the febrile period of relapsing fever, large numbers of spirochetes often circulate in peripheral blood and can be detected by light or dark field microscopy wet preparations made from a drop of blood mixed with a small drop of sodium citrate. Romanowsky stains are used to stain thick and thin films for examination. Spirochetes in the peripheral blood of patients with mild infection are more readily detected by a buffy coat concentration technique.

Packaging

Catalog #	Volume
MA0303001	Malaria Stain Kit, 500mL, 1 kit
MA0303002	Malaria Stain Kit, Thin Prep Solution A, 500mL
MA0303003	Malaria Stain Kit, Thick Prep Solution B, 125mL
MA0303004	Malaria Stain Kit, Thick Prep Solution C, 1000mL
MA0303005	Malaria Stain Kit, 250mL, 1 kit
MA0303006	Malaria Stain Kit, Thin Prep Solution A, 250mL
MA0303007	Malaria Stain Kit, Thick Prep Solution B, 60mL
MA0303008	Malaria Stain Kit, Thick Prep Solution C, 500mL

Store all solutions at Room Temperature.

Staining Procedure

Staining of thin peripheral blood smears:

- Place 50 mL of Solution A in a coplin jar. Then place 50 mL of distilled water in each of two sequentially placed coplin jars.

- Solution A is good for 30 slides or a maximum of 1 month if used.

A). Dip slide in Solution A for 10 to 15 seconds.

B). Dip slide in distilled water for 15 to 30 seconds. For darker stained leukocytes, dip slide in distilled water for one minute or more.

C). Rinse by dipping in distilled water, drain for several minutes, and allow to air dry.

D). Examine a minimum of 300 fields of the thin film under oil immersion. Examine the entire smear under low power if microfilariae are suspected. The sheath of *W. bancrofti* may not be visible.

Staining of thick peripheral blood smears:

- Do not fix the thick film. Lyse the RBC's by covering the smear with distilled water for 10 minutes.

Drain the water and allow the smear to air dry horizontally.

A). Add 1 drop of solution B to each milliliter of solution C. about 15 to 20 mL is required to stain 2 slides.

B). Place 2 thick smears, face down, across a watch glass.

C). Run freshly prepared diluted solution B under the slides until the undersurface is covered.

D). Stain for 30 to 60 minutes. The longer time of 60 minutes is recommended.

E). Rinse slides by dipping into clean distilled water, drain for several minutes, and allow to air dry.

F). Examine thick film for 5 to 10 minutes, covering a minimum of 100 fields. Examine the entire smear under low power if microfilariae are suspected. The sheath of *W. bancrofti* may not be visible.

Comments

- 1). If Plasmodium organisms are present, the cytoplasm will stain blue and the nuclear material will stain red to purple red.
- 2). Schuffner's stippling and other inclusions in the erythrocytes infected with Plasmodium spp. will stain red.
- 3). Nuclear and cytoplasmic colors that are seen in the malarial parasites will also be seen in the trypanosomes and in any intracellular leishmania that are present.
- 4). Infection with malaria cannot be ruled out by examination of one peripheral blood sample. Multiple samples may be required and should be collected every 6 to 12 hours after the first sample until a diagnosis is made or infection is no longer suspected. The optimal time for drawing blood to detect periodic microfilariae is between 10 p.m. and 4 a.m. When collecting a sample by venipuncture, EDTA is the preferred anticoagulant. Heparin or sodium citrate may be used if trypanosomes or microfilariae are suspected.
- 5). The thick smear provides more sample than the thin smear and is more likely to have increased detection rates of low levels of parasitemia. Erythrocytes are lysed during the staining process, and this provides a somewhat transparent film leaving only the parasites, platelets, and leukocytes.
- 6). If the thick smear is prepared from anti-coagulated blood that is more than one hour old, morphology of the parasites may not be typical, and care must be taken so blood film does not wash off the slide during the staining process.
- 7). All glass slides should be dipped in alcohol and polished with a lint-free cloth to remove any grease or dirt.
- 8). Examine the entire smear under low power if microfilariae are suspected. The sheath of *W. bancrofti* may not be visible.