

RAPID MALARIA STAIN [JSB I & II]

KIT FOR STAINING MALARIA PARASITES



PRINCIPLE

Polychromated Methylene Blue and Eosin stains specifically to basophilic and acidophilic cellular elements to demonstrate blood cells and hemoparasites.

STAINS COMPOSITION

- RAPID-MALARIA JSB I** 500 mL
Methylene blue 4.3 mMol/L
Sulphuric Acid 1%
Potassium Dichromate 0.5 gms
Phosphate Buffer 0.1 mMol/L
Contains polychromating materials, preservatives and stabilizers.
- RAPID-MALARIA JSB II** 500 mL
Eosin 4.3 mMol/L
Phosphate 0.1 mMol/L
Contains preservatives and stabilizers.
- EASYFIX** 50 mL
Peripheral Smear Fixative

Preparation of Working Reagent

All Reagents are ready to use.

STORAGE AND STABILITY

All reagents are stable between 25 to 35°C for 60 months. RAPID-MALARIA FIXATIVE must be stored tight capped away from heat and fire. Rapid-MALARIA JSB STAIN may be exhausted on prolonged use over 150 smears. It is advised to discard Rapid-MALARIA JSB Stain as and when the smear gets lighter stain.

SAMPLE

Peripheral blood samples on clean glass slides may be collected upon febrile episodes from susceptible patients. K₃ EDTA may be used without substantial change in staining results.

THIN SMEAR

A drop of blood can be spread with a fine edge spreader. Dry the smear at air and fix it with FIXATIVE or rectified spirit within 4 hours. Fixed smear can be stored for longer period without much variation in staining result, however, it is preferable to stain all smears as early as possible.

THICK SMEARS

In mass or conclusive screening it is preferable to prepare thick smears by collecting 1-2 drop of blood on a clean glass slide and spread it to the shape of a coin. Dry at air, add few drops of distilled water to the slide to cover the smears and wait for 1-2 minutes to dehaemoglobinize the smear. Carefully remove water and dry at air. Fix the smear

LIMITATIONS

The performance of stain must be periodically checked by known positive and known negative samples. The accuracy of reporting Malarial parasite is subject to the professional experience of each person as well as the use of a good optical system that could make clear magnification of the parasite from the smear.

PROCEDURE

Fill up two Coplin jars or wide mouth bottles with Rapid-MALARIA STAIN - I and II

A . FOR THIN SMEARS

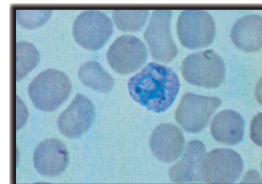
- Spray RAPID-MALARIA JSB STAIN FIXATIVE to thin smears and dry at air.
- Dip fixed smear to RAPID-MALARIA JSB STAIN - II for 3-5 seconds and wash in running tap water.
- Dip smear in RAPID-MALARIA JSB STAIN - I for 35-45 seconds. Wash in running tap water. Dry at air and see under oil immersion objective for malarial parasites. Staining time in each stain may be increased or decreased according to individual staining result.

B. FOR THICK SMEARS DEHAEMOGLOBINIZING

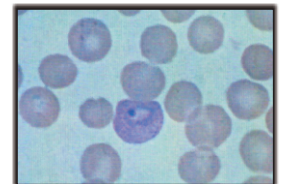
- Arrange air dried unfixed thick smears in a slide rack keeping the smear side up.
- Add few drops of distilled water to the slide to cover the smears and wait for 1-2 minutes to dehaemoglobinize the smear. Carefully remove water and dry at air and fix smear by spraying fixative. Dry at air.
- Dip fixed smear to RAPID-MALARIA JSB STAIN - II for 5-6 seconds and wash in running tap water.
- Dip smear in RAPID-MALARIA JSB STAIN - I for 35-45 seconds. Wash in running tap water. Dry at air and see under oil immersion objective for malarial parasites.

RESULTS

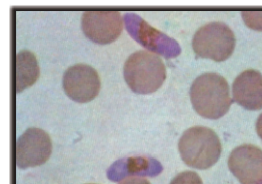
Leucocyte nuclei	- varying shades of dark blue
Leucocyte Cytoplasm	- pale or light blue
RBC	- pale reddish
Malaria Parasite	- Varying shades of blue
Scifner's Dot	- dark bluish red.
Platelets	- pale or dark blue.



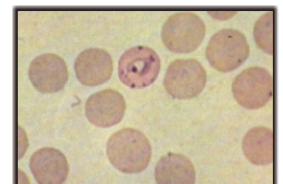
Pl. vivax trophozoites



Pl. Vivax trophozoites



Pl. Falciparum Gametocyte



Pl. falciparum trophozoite

BULK STAINING PROCEDURE

Using staining tanks number of slides can be stained with relatively less time.

A. THIN SMEAR

- Label all smears with appropriate patient identification marks.
- Fix thin smear by immersing or spraying with Rapid-MALARIA JSB STAIN Fixative and arrange in a slide holder.
- Fill Rapid-MALARIA JSB STAIN II in a suitable staining jar and dip the smears in the stain for 3-5 seconds.
- Remove and dip in another jar filled with water for 10-20 seconds. Agitate the slide holder to remove excessive stain from the smear.
- Remove the water by keeping the slides holder over a filter paper.
- Fill Rapid-MALARIA JSB STAIN - I in a suitable staining jar and dip the smears in the stain for 35-45 seconds.
- Remove and dip in another jar filled with water for a minute. Agitate the slide holder to remove excessive stain from the smear. Dry the smears at room temperature or in an oven thermostatically controlled to 50-60°C.

B. THICK SMEAR

- Label all smears with appropriate patient identification marks and arrange in a slide holder.
- Dehaemoglobinize by dipping in a jar containing distilled water for 1-2 minutes. Carefully remove from water.
- Proceed to Bulk staining procedure thin smear step No. 2.

NOTE : Unfixed thick smears may run out from the slides therefore, all steps of Dehamoglobinization and washing must be without much agitation.

BIBLIOGRAPHY

- Mac.Neal, J.A.M.Med.Assoc. 78, 1112 (1922).
- Biological Stain 9th Ed. The Williams & Wilkins Co. Balti More, MD 1977 P. 424.
- J.Med Res. 7, 138 (1902)
- "Histopathological technic and pract histochem The Blackston Co., New York, N.Y. 1954.

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