Atlas Medical

CE

ATLAS LEISHMAN STAIN

IVD *For in -vitro diagnostic use only*

Store at Room Temperature

Intended For Use

Atlas Leishman's stain is used for staining of blood and bone marrow. It is generally used to differentiate and identify leucocytes, malaria parasites, and trypanosomas (unicellar parasitic protozoa). It is based on a mixture of methylene blue and eosin.

Introduction

Leishmaniasis is a parasitic disease caused by several species of genus Leishmania (protozoa) and transmitted by the bite of sand flies.Sand flies are primarily infected by animal reservoir, but humans are also a reservoir for some Leishmania

Materials Provided

- Leishman stain
- Distilled water (buffered to pH 6.6 - 6.8)

Technique A - for blood smears

1. Spread thin smears of the blood to be examined on slide and leave for few minutes to dry in the air.
2. Fix the smears. Immerse the in solution A for 1 minute, in a stoppered or corked staining vessel.
3. Transfer to a 1:3 mixture of solution A and solution B and allow this diluted stain to act for 5-10 minutes.
4. Rinse the slides in solution B then immerse them in this solution until the smears appear pink to the naked eye (this usually takes 1-2 minutes but see note i below).
5. Blot and air dry; mount, if desired in a synthetic neutral medium such as DPX.

Result

As with Wright stain but in lighter tones.

Notes and Observations

- If the differentiation in step 4 (above) is slow or difficult, try drying the smears, rinsing in xylene, drying again and re immersion in solution B for a few minutes whilst agitating the solution. If this does not work then try differentiating in distilled water of a lower pH (but not below pH 6.5).
- For malarial parasites solution B should be pH 7.2 to give maximum visibility to Schuffner's dots.

Technique B - Throat exudates, Bacterial smears

1. prepare thin smear of the material to be stained.
2. Stain for 2 minutes with solution A.
3. Immerse in solution B for 1 minute.
4. Blot dry.

Result

Bacteria stain dark blue; capsules light blue, mucus red.

Technique C - for thick blood smears

A routine method of searching for blood parasites which are believed to be scanty.

1. Place a large drop of blood on a slide and spread rapidly with a thin rod or pin to about 20mm diameter.
2. Thoroughly dry in incubator for about 1-2 hours, keeping slide horizontal.
3. Remove haemoglobin by placing slide face downwards in a dish of cold distilled water.
4. Fix in acid alcohol (alcohol 50ml, hydrochloric acid 10 drops) for 10-15 minutes.
5. Rinse with water and blot dry.
6. Stain as an ordinary blood smear (Technique A above).

References

1. WHO: *Basic Malaria Microscopy; Part 1*. Geneva, Switzerland: World Health Organization,WHO/MAL/20001091; 2010.
2. WHO: *Regional Guidelines for the Management of Severe Falciparum Malaria in Large Hospitals*. New Delhi: World Health Organization, Regional office for South-East Asia; 2006.
3. Bejon P, Andrews L, Hunt-Cooke A, Sanderson F, Gilbert SC, Hill AV: Thick blood film **e**xamination for *Plasmodium falciparum* malaria has reduced sensitivity and underestimates parasite density.

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REF	Product Reference No.	IVD	For in-vitro diagnostic use.
	Caution.		Store at
	Read product insert before use.	Σ	Number of tests in the pack.
LOT	Lot (batch) number.		Manufacturer.
	Expiry date.		Manufacturer telephone number.
	Manufacturer fax number.		