ZN Stain Kit, Methylene Blue

IN VITRO DIAGNOSTIC DATASHEET

This kit is used for the staining of Mycobacterium tuberculosis and other acid fast organisms

INTENDED USE : IN VITRO DIAGNOSTIC USE

DESCRIPTION : TB Stains are use to identify acid fast bacilli (Mycobacterium tuberculosis) in smears and tissue sections. The cell walls of Mycobacteria are not readily penetrated by aniline dyes solutions so in order to stain Mycobacteria the stain incorporates Basic Fuchsin (a red dye) and phenol to make a powerful stain. Heat can be applied to "force "the stain into the bacillus (standard Ziehl-Neelsen method). Once stained the bacillus is very resistant to decol ourisation retaining the stain even when the rest of the preparation has been decolourised. The contrast stain(s) used following the bacillus staining are weak solutions of either Methylene Blue or Malachite Green to give a pale blue or green background against which the stained(red) bacilli will be easily visible.

FIXATION

Smears: Prepare smears on cleaned slides and air dry for 5-10 minutes. Fix in absolute ethanol for 10 min.
Tissues: A standard formaldehyde based fixative provides satifactory results. Tissues should be processed and embedded in paraffin wax and cut at 5 microns

REAGENTS :	**150 TEST**
Carbol Fuchsin ZN	1x 50ml
TB Differentiator	1x 50ml
Methylene Blue Loeffler	1x 50ml

**** Number of TEST calculated according to 330 microliter per slide.***

CATALOG NO : **PLKit24-150**

PROCEDURE TIME : Approximate 40 minute.

PROTOCOL :

1. Deparaffinise sections and take to water through graded alcohols. For smears flood slides with DI water.
2. Place sections/smears in a 50 ml coplin jar (or slide mailer 20 ml) of carbol fuchsin.
   Place in an oven at 37 decrees C, for smears 20 minutes, sections 30 minutes.
   Note : AVOID STAINING MULTIPLE SAMPLES IN STAINING JARS/SLIDE MAILERS TO REDUCE RISK OF CROSS CONTAMINATION OF SLIDES DURING HEATING. ALWAYS THROW AWAY USED SOLUTIONS AND WASH COPLIN JARS. THROW AWAY USED SLIDE MAILERS.
3. Remove from staining solution and place on staining rack.
4. Wash well with deionised water for 3-5 minutes.
5. Differentiate with TB Differentiator until preparation is colourless or pale pink 20-30 seconds.
6. Wash well in tap water for 5 minutes. Rinse in distilled water.
7. Counterstain with Methylene Blue for 15-20 seconds. Dilute Loeffler's blue up to 1 in 10 with distilled water (depending upon intensity of colour desired).
   Note : COUNTERSTAIN SHOULD BE PALE BLUE AND NOT OVERSTAIN THE ACID FAST BACILLI.
8. Wash well in distilled water and blot dry.
9. Take smears/sections rapidly through absolute alcohols, clear and mount.
   Note : IF IMMERSION OIL WILL BE USED FOR VISUALISATION, SMEARS MUST AIR DRIED FOR 20-30 MIN.

RESULTS :

Acid fast bacilli (M.tuberculosis) :	Magenta / Red
Cells and background material, other organisms :	Blue

STORAGE AND STABILITY : This product is stable for 36 months when stored in +15 /+25 C

TROUBLESHOOTING : Please contact Patolab Technical Support by e-mail (patolab@patolab.com.tr).

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