

# Reticulin Stain Kit

IN VITRO DIAGNOSTIC DATASHEET

This staining technique is used to demonstrate reticulin fibres.

INTENDED USE: IN VITRO DIAGNOSTIC USE

**DESCRIPTION:** This staining technique is a metal impregnation technique used to demonstrate reticulin fibres providing contrast enabling even the finest of fibres to be seen. Reticulin fibres have poor affinity for silver solutions but treatment with potassium permanganate produces sensitised sites where silver can be deposited. This deposition of silver ions requires an optimum pH of 9.0. Post treatment with formaldehyde solution causes deposition of the silver ions as silver metal. Excess silver is removed by treatment with sodium thiosulphate.

#### **SPECIMEN COLLECTION:**

Fixation in routine Buffered Formalin solution is satisfactory. Paraffin sections at 5 microns.

REAGENTS:	150 TEST
POTASSIUM PERMANGANATE (0.5%)	1x 50ml
SULPHURIC ACID (3%)	1x 50ml
OXALIC ACID(1%)	1x 50ml
FERRIC AMMONIUM SULPHATE (2.5%)	1x 50ml
SILVER NITRATE (10% aqueous solution)	1x 50ml
AMMONIA	1x 50ml
SODIUM HYDROXIDE (3%)	1x 50ml
FORMALDEHYDE (10% v/v) (Unbuffered)	1x 50ml
SODIUM THIOSULPHATE (5% aqueous)	1x 50ml

<sup>\*</sup> Number of TEST calculated according to 330 microliter per slide.

CATALOG NO: PLKit102-150

**PROCEDURE TIME:** Approximate 30 minute.

## **PREPARATION**

## **Stock Solution**

Pipette 5ml of Silver nitrate solution into a chemically clean flask. Add 1ml Ammonia solution, mix well until the precipitate formed just dissolves (this point is reached when a faint opalescence is seen). Add 5ml Sodium hydroxide solution mix and then add 0,5 ml Ammonia solution. Store in Refrigerator.







## **Working Ammoniacal Solution**

Mix 1 part Stock solution with 2 part Distiled water.

## **Preparation of Acidified Potassium Permanganate:**

Mix 4,75 ml of Potassium permanganate solution with 0,25 ml of Sulphuric Acid. Use 5 times and discard.

#### **PROCEDURE**

- 1. Deparaffinise the test and a known positive control section in Xylene and bring to tap water.
- 2. Oxidise the sections with acidified potassium permanganate for 10 minutes.
- 3. Wash well in tap water.
- 4. Decolourise with oxalic acid solution for 1-2 minute.
- 5. Wash sections well in tap water water.
- 6. Mordant sections in Ferric ammonium sulphate solution for 10 minutes.
- 7. Wash well with several changes of distilled water.
- 8. Treat with Ammoniacal working silver solution for 3 minutes.
- 9. Wash well in several changes of distilled water (the section must be transluscent)
- 10. Reduce in 10% Formalin solution for 1-2 min, check microscopically and if under impregnated repeat steps 8-10.
- 11. Wash in distilled water. Treat with sodium thiosulphate for 5 minutes.
- 12. Dehydrate through two changes 90% Ethanol.
- 13. Complete dehydration in two changes of absolute alcohol.
- 14. Clear in Xylene

#### **RESULTS:**

Reticulin fibres: Black

Nuclei: Black

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 ).

