



VitroView™ Safranin O Stain Kit for Cartilage
SKU#: VB-3028

Description:

Safranin O is a cationic dye which binds specifically to polyanions. It detects the amount of proteoglycan present in cartilages. The intensity of Safranin O staining is proportional to the proteoglycan content in the cartilage tissue. It is used to detect any variations in articular disease as a marker for glycosaminoglycan depletion in osteoarthritis. Fast Green counterstains the non-collagen sites and provides a clear contrast to the Safranin O staining.

This VitroView™ Safranin O Stain kit is used for the detection of cartilage, mucin, and mast cell granules on formalin-fixed, paraffin-embedded tissue sections, and may be used for frozen sections as well. The cartilage and mucin will be stained orange to red, and the nuclei will be stained black. The background is stained bluish green.

Kit Contents:

VB-3028-1	Weigert's Hematoxylin Solution A	-----125 ml
VB-3028-2	Weigert's Hematoxylin Solution B	-----125 ml
VB-3028-3	Acid Ethanol	-----250ml
VB-3028-4	Fast Green (FCF) Solution	-----250ml
VB-3028-5	Acetic Acid Solution	-----250ml
VB-3028-6	Safranin O Solution	-----250 ml

Storage

Room temperature.

Protocol

Safranin O Stain for paraffin sections

1. Decalcified paraffin sections cut at 4 microns
2. Deparaffinize in xylene I for 6 minutes and II for 6 minutes.
3. Rehydrate
 - a. Ethanol 100% (2 minutes)
 - b. Ethanol 100% (2 minutes)
 - c. Ethanol 95% (2 minutes)
 - d. Ethanol 95% (2 minutes)
 - e. Ethanol 70% (2 minutes)
4. Rinse in DI water (2 minutes).
5. Prepare Weigert's working hematoxylin: mix Weigert's Hematoxylin Solution A and B at 1:1 ratio.
6. Weigert's working hematoxylin for 10 minutes.
Note: Discard after use.
7. Blue in running tap water for 5 minutes.
8. Destain quickly in Acid ethanol (2-3 dips)

9. Wash in tap water for 2 minutes, rinse once for 1 min in distilled water.
10. Stain with fast green (FCF) solution for 5 minutes.
11. Rinse quickly with acetic acid solution for 2-3 dips.
12. Stain in safranin O solution for 5 minutes.
13. Dehydrate with 2 changes of 100% Ethanol (2 minute per change).
14. Clear with 2 changes of xylene (5 minute per change)
15. Mount cover slip onto glass slide with Permount or some other suitable organic mounting medium.

Safranin O Stain Stain for frozen sections

1. Frozen section are fixed in acetone at -20 °C for 15 min, or 80% Methanol at 4°C for 5 min, or 10% neutralized formalin for 10 min.
2. PBS 1× 10 min.
3. Rinse in DI water (2 minutes).
4. Follow step 4-15 above.

Expected Results:

Nuclei-----Black
Cytoplasm-----Gray green
Cartilage, mucin, mast cell granules-----Orange to red

Note: This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

Precautions: Handle with care. Avoid contact with eyes, skin and clothing. Do not ingest. Wear gloves.