Van Gieson Staining

Summary and Explanation

Van Gieson Stain is used to differentiate between collagen and smooth muscle in tumors and to demonstrate the increase of collagen in diseases. This method combines two or more anionic dyes and rely on differential binding by tissue components.

The differentiation is determined by a combination of differences in the relative size of the dye molecules, differences in the physical structure of the tissue, and differences in the amino acid composition of tissue Elements.

Principles of the Procedure

Van Gieson's Stain is a mixture of Picric Acid and Acid Fuchsin. It is the simplest method of differential staining of Collagen and other Connective Tissue. When using combined solutions of picric acid and acid fuchsin, the small molecules of picric acid penetrate all of the tissues rapidly, but are only firmly retained in the close textured, red blood cells and muscle. The larger molecules of Ponceau S displace picric acid molecules from collagen fibres, which have larger pores, and allow the larger molecules to enter.

Reagents Provided

Celestin blue Solution	20ml, ready to use
Mayer's Haematoxylin	20ml, ready to use
Curtis Stain	20ml, ready to use

Reagents Required but Not Supplied

EZ-DeWax™ Solution	500ml, concentrate
Super Sensitive TM Wash Buffer, 20X	500ml, concentrate
Mounting Medium	
Positive Control	

Storage and Handling

Store all reagents at 2-8°C. Do not use after expiration dates as indicated on the reagent labels.

Specimen Preparation

Fixation plays an important role in preserving the tissue structures to be visualized using this stain. 10% neutral buffered formalin is the preferred fixative. Ensure that the fixed sections are adequately embedded in paraffin. Cut tissue sections to 4-5 microns.

Automated Staining System Protocol

1. Bring reagents to room temperature.
2. Load the barcode-labeled slides for the appropriate stain into the Xmatrx® Automated Staining System slide racks. Load the RFID Tagged vials into the Xmatrx® Automated Staining System reagent racks.
3. Start Special Stains software on the instrument and check the protocol parameters against the factory default settings listed below. Change any of the parameters as required. Please note that any parameter change, once saved, becomes the default setting until changed again. Use of parameter settings other than factory default requires validation by user.

4. Select “Start Scan” and the instrument will perform the steps listed in the table (if the factory default settings have been selected).

VAN GIESON STAINING PROTOCOL DEFAULT SETTING:

Reagent	No.of Incubations	Incubation Time	No. of After Rinses
Baking	1	15min	0
XDeWax	3	3min	0
Special Stains Wash Solution	3	30sec	0
Celestin Blue	1	5min	2
Haematoxylin	1	5min	4
Curtis stain	1	10min	3
Xmount	1	10min	0

* Alcohol Wash

Quality Control

A positive control slide–one that will display positive staining with this stain–should be included in every run.

Trouble shooting

1. Follow data sheet instructions correctly.
2. Gently invert all reagents prior to use.

Expected Results

Nuclei stain blue; Collagen stain bright red; Cytoplasm, muscle, fibrin and red blood cells stain yellow

Limitations of the procedure

1. The solution weakens after long standing and may be strengthened by adding few drops of fresh Acid Fuchsin.
2. The thickness of the section may affect the intensity of the staining. Necrotic tissue may exhibit non-specific staining.
3. Use tissue, which shows positive structure or element for which it is being tested.

Performance Characteristics

Specificity of Special Stain

Van Gieson is used to differentiate between collagen and smooth muscle in tumors and to demonstrate the increase of collagen in diseases.

Saint-Bio has conducted studies to evaluate the performance of all its Special Stains products. Saint-Bio Special Stains have shown reproducible and consistent results when used within a single run, between runs and between lots. The products have been determined to be stable for the periods specified on the labels either by standard real time or accelerated testing methods. Saint-Bio ensures product quality through 100% quality control for all products released and through surveillance programs.