




Saffron du Gatinais solution  
 Technical information  
 Msds code 09-318  
 Product code 09-318  
 Stability of product properly conserved 24 month.  
 Pack 1000 ml

Produce in Italy by:  
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in case of emergency UE number		112
in case of emergency UK number		999
en cas d'urgence Suisse number		145

**Tissue sample**

Formol sublimate fixation is preferred, although 10% formalin variants are acceptable.  
 Paraffin sections at 3 µm are preferred.

**Method Movat pentachrome**

Bring sections to water via xylene and ethanol.  
 Optionally, place into picro-mercuric chloride for 1 hour.  
 Wash well with warm water to remove picric acid.  
 Rinse with 3% acetic acid.  
 Place in alcian blue at 60°C for 10 minutes  
 Rinse with distilled water.  
 Place in working Verhoeff's solution for 6 minutes.  
 Wash with warm tap water for 6 minutes.  
 Place in the plasma stain for 3 minutes  
 Rinse with distilled water.  
 Place in the polyacid for 15 minutes.  
 Rinse with 1% acetic acid.  
 Dehydrate thoroughly with absolute ethanol 3 changes.  
 Place in the saffron du Gatinais stain for 5-6 minutes.  
 Dehydrate, clear  
 Mount with DdMount

**Results**

Nuclei	black
Elastic fibres	black
Fibrin – mature	red
Muscle	red
Collagen	yellow
Ground substance	blue-green

**Notes**

Sections should be treated with picro-mercuric chloride if formol sublimate or similar fixation was not used initially. The ferric chloride should be the hexahydrate crystals. Sections thicker than 4 µm may need the elastic stain differentiated by treating with 1% aqueous ferric chloride for 20-30 seconds, then washing well with tap water. Although the iodine content of the Verhoeff elastic stain component may remove any mercury pigment, in methods such as this some technologists prefer to apply the iodine-thiosulphate sequence before staining.

HPS stain is haematoxylin saffron phloxine

This combination of three stains is used mostly to demonstrate collagen in connective tissues. Haematoxylin, a basic dye, stains acidic structures such as DNA, purple. Phloxine, an acidic dye, stains basic structures including most proteins, pink. Saffron stains collagen yellow.

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### Hemalum-Phloxine-Saffron Stain (H.P.S.)

#### Principle

Trichome staining: nuclear and cytoplasm staining is achieved with hemalum and phloxine while saffron demonstrates connective tissue collagen.

#### Hemalum-Phloxine-Saffron stain

#### Procedure

Use a wax-free hydrated section  
Nuclear staining with Mayer's haemalum 3 to 5 minute  
If necessary, differentiate in hydrochloric acid  
Place in Scott water for 2-3 minute  
Wash with water  
Stain in 3 % aqueous Phloxine solution for 2 minutes  
Rinse with water,  
Differentiate in 70 then 95 % alcohol  
Absolute alcohol,  
Stain the slide with saffron in alcohol for 5 to 8 minutes  
Pass, very quickly (critical for successful staining) through absolute alcohol, then xylene  
Pass through 2-xylene bath  
Mount with DdMount

#### Note

Staining time depends on tissue type. Phloxine can be replaced by eosin.

#### Results

Nuclei	blue
Cytoplasm	pink
Muscle fibres, RBC	bright red
Collagen	golden yellow
Elastic fibres	pink

#### Reference

Ganter r., jolles g., *histochimie normale et pathologique*, ed. Gauthier villars - paris (1970)

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