

Document N	umber: SASoM/METHOD/119.v1
Title:	Histology – Masson's Trichrome Staining
Version:	v1
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Effective from:	28/04/2020	
Valid to:	27/04/2022	

SOP History		
Number	Date	Reason for Change
v1	28/04/20	Original

1.0 Purpose -

This SOP describes the current procedure for carrying out Periodic acid-Schiff Staining on FFPE sections in Laboratory 248/249 at the St Andrews School of Medicine (SASoM).

2.0 Scope -

This SOP applies to all staff in the SASoM carrying out Periodic acid-Schiff Staining on FFPE sections in Laboratory 248 at the St Andrews School of Medicine (SASoM).

3.0 Responsibilities -

All staff performing Periodic acid-Schiff Staining on FFPE sections in this manner are responsible for ensuring that the methods are followed in accordance with this SOP.

All staff must have read and signed the relevant risk assessment documents before performing this procedure.



4.0 Procedure –

The Masson's trichrome stain staining method is used for distinguishing cells from surrounding connective tissue. It is particularly useful to visualise collagenous tissue connective fibres in tissue such as skin and heart. It can also be used to differentiate between collagen and smooth muscle in tumours where an increase of collagen can often be seen in diseases such as cirrhosis of the liver. It is a routine stain for liver and kidney biopsies.

Control Tissue: Skin, lung, stomach, intestine, uterus





De-wax & Re-hydrate

Always allow any excess fluid to drain from the slide rack before proceeding to the next solution.

- 1. Dewax-Xylene 1
- 2. Dewax-Xylene 2
- 3. Dewax-Xylene 3
- 4. Rehydration-100% Alcohol
- 5. Rehydration-100% Alcohol
- 6. Rehydration-80% Alcohol
- 7. Rehydration-50% Alcohol
- 8. Wash in running water

5 minutes 2 minutes

5 minutes

5 minutes

- 2 minutes
- 2 minutes
 - 2 minutes
- 2 minutes

Staining

- 1. After hydration in water, place the slides in 'Celestine Blue' for 5 minutes.
- 2. Wash in water.
- 3. Stain in haematoxylin, differentiate and blue up
- 4. Wash in water.
- 5. Stain in 'Ponceau Fuchsin' solution for 5 minutes
- 6. Wash in water.
- 7. Differentiate in 1% Phosphomolybdic acid until collagen staining is decolourised (between 3-5minutes).
- 8. Wash in water.
- 9. Counterstain in 1% Light Green (made up in 1% acetic acid) for 1-2minutes.
- 10. Wash in water.
- 11. Rinse in 1% acetic acid for 1 minute.
- 12. Wash, dehydrate, clear and mount.

5.0 Personal protection -

A Howie coat must be worn at all times. Gloves as specified in the appropriate COSHH RA.

6.0 Spillages -

Always clean up any spills immediately after use, only you know what you have spilt and are aware of its hazard. Spillages should be mopped up with paper towel, disinfected with 70% ethanol and finally washed with detergent.

7.0 Training –

All staff should undergo training in this technique before performing the procedure.



8.0 Related documents -

8.1 Risk assessments –

RA20229 (Histology - Martius, Masson's and Periodic acid-Schiff Staining)

9.0 Approval and sign off –

Author:			
Name:	Peter Mullen		
Position:	Research Fellow		
Signature:		Date:	
Management Appr	oval:		
Name:	Peter Mullen		
Position:	SOP Administrator		
Signature:		Date:	
QA release by:			
Name:	Alex MacLellan		
Position:	QA Manager		
Signature:		Date:	

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STANDARD OPERATING PROCEDURE

Please sign below to indicate you have read this S.O.P and understand the procedures involved.

NAME	POSITION HELD	SIGNATURE	DATE