



May 09, 2023

TS Spurrs - FFPE block - 1mm3 cube cut out - tissue (TM - 013)

DOI

<https://dx.doi.org/10.17504/protocols.io.kxygx968wg8j/v1>

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Protocol Citation: sandra.crameri 2023. TS Spurrs - FFPE block - 1mm3 cube cut out - tissue (TM - 013). **protocols.io**
<https://dx.doi.org/10.17504/protocols.io.kxygx968wg8j/v1>

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Protocol status: Working

We use this protocol and it's working

Created: May 09, 2023



Last Modified: May 09, 2023

Protocol Integer ID: 81632

Keywords: ts spurr, paraffin block, face to spurr, spurr, ffpe block, resin block, 1mm3 cube, ffpe, resin block for em, tissue, tm, piece, resin

Abstract

Processing unstained paraffin block that has had a 1mm3 piece dissected out of the face to Spurr's resin block for EM.

Guidelines

All step times are the minimum time required. Longer steps are acceptable.

Protocol materials

⊗ xylene

⊗ Ethanol

⊗ Ethanol

⊗ Ethanol

⊗ Glutaraldehyde aqueous solution 25% EM Grade ampoules **ProSciTech Catalog #C002**

⊗ Osmium Tetroxide **ProSciTech**

⊗ ethanol



HEADER

1 SAN:

SPEC No:

OPERATOR & STEPS:

OPERATOR & STEPS:

Dewax FFPE sections

15m

2  xylene or X3B  Overnight at  60 °C

5m

3  xylene or X3B  00:05:00 at  Room temperature

5m

4  xylene or X3B  00:05:00 at  Room temperature

5m

Rehydrate

15m

5 [IM] 100 % volume  Ethanol  00:10:00

10m

6 [IM] 95 % volume  Ethanol  00:10:00

10m

7 [IM] 70 % volume  Ethanol  00:10:00

10m

Conventional - with reduced times for 5um thick FFPE section

2h 50m



- 8 [M] 2.5% % volume 20m
⊗ Glutaraldehyde aqueous solution 25% EM Grade ampoules **ProSciTech Catalog #C002**
in [M] 0.1 Mass Percent Sorenson's phosphate buffer pH 07.2 300 mosmol/kg
⌚ 00:20:00 .
- 9 Wash buffer ⌚ 00:05:00 5m
- 10 [M] 1 Mass / % volume ⊗ Osmium Tetroxide **ProSciTech** in buffer ⌚ 01:00:00 1h
- 11 [M] 70 % volume ⊗ Ethanol ⌚ 00:10:00 10m
- 12 [M] 95 % volume ⊗ ethanol ⌚ 00:10:00 10m
- 13 [M] 100 % volume ⊗ ethanol ⌚ 00:10:00 10m
- 14 [M] 100 % volume dry ⊗ ethanol ⌚ 00:10:00 10m
- 15 [M] 100 % volume Spurrs: [M] 100 % volume dry ⊗ ethanol in 1:1 ratio 00:30:00 30m
- 16 [M] 100 % volume Spurrs ⌚ 01:00:00 1h
- 17 [M] 100 % volume Spurrs ⌚ 01:00:00 1h
- 18 Oven polymerise 🔥 65 °C ⌚ Overnight