🌐 TS Spurrs - unstained FFPE on Thermanox coverslips (TM - 013) V.4

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[1]CSIRO ACDP
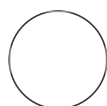
◯ sandra.crameri

ABSTRACT

Processing unstained FFPE sections on Thermanox coverslips to Spurrs resin block for EM.

GUIDELINES

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

VERSION 4

MAY 09, 2023

DOI:

Protocol Citation: sandra.crameri 2023. TS Spurrs - unstained FFPE on Thermanox coverslips (TM - 013). **protocols.io**
https://dx.doi.org/10.17504/protocols.io.3byl4b7rjvo5/v4Version created by sandra.crameri

Protocol status: Working
We use this protocol and it's working

Created: May 09, 2023

Last Modified: May 09, 2023

PROTOCOL integer ID:
81628

HEADER

1 SAN:

SPEC No:

OPERATOR & STEPS:

OPERATOR & STEPS:

Dewax FFPE sections · 15m

2 ⊗ xylene **Contributed by users** or X3B ⏱ 00:05:00 · 5m

3 ⊗ xylene **Contributed by users** or X3B ⏱ 00:05:00 · 5m

4 ⊗ xylene **Contributed by users** or X3B ⏱ 00:05:00 · 5m

Rehydrate · 15m

5 [M] 100 % volume ⊗ Ethanol **Contributed by users** ⏱ 00:05:00 · 5m

6 [M] 95 % volume ⊗ Ethanol **Contributed by users** ⏱ 00:05:00 · 5m

7 [M] 70 % volume | ⊗ Ethanol **Contributed by users** | 🕐 00:05:00 | 5m

Conventional - with reduced times for 5um thick FFPE secti... 2h 50m

8 [M] 2.5% % volume | 20m

⊗ Glutaraldehyde aqueous solution 25% EM Grade ampoules **ProSciTech Catalog #C002** in [M] 0.1 Molarity (M) 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 🕐 00:30:00 | 30m

11 [M] 70 % volume | ⊗ Ethanol **Contributed by users** | 🕐 00:10:00 | 10m

12 [M] 95 % volume | ⊗ ethanol **Contributed by users** | 🕐 00:05:00 | 5m

13 [M] 100 % volume | ⊗ ethanol **Contributed by users** | 🕐 00:05:00 | 5m

14 [M] 100 % volume dry   ⊗ ethanol **Contributed by users**   ⏱ 00:05:00   `5m`

15 [M] 100 % volume Spurrs: [M] 100 % volume dry   ⊗ ethanol **Contributed by users** in 1:1 ratio ⏱ 00:30:00   `30m`

16 [M] 100 % volume Spurrs ⏱ 00:30:00   `30m`

17 [M] 100 % volume Spurrs ⏱ 00:30:00   `30m`

18 Oven polymerise 🌡 65 °C   ⏱ Overnight