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TS Spurrs - Cell Monolayer on Thermanox coverslips (TM - 013)

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Protocol status: Working

We use this protocol and it's working

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Keywords: cell monolayer on thermanox coverslip, thermanox coverslip, cell monolayer, resin block for em, resin block, ts spurr, resin

Abstract

Processing a cell monolayer on Thermanox coverslips to Spurr's resin block for EM.

Guidelines

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

Protocol materials

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

⊗ Osmium Tetroxide **ProSciTech**

⊗ Ethanol

⊗ ethanol



HEADER

1 SAN:

SPEC No:

OPERATOR & STEPS:

OPERATOR & STEPS:

Conventional - with reduced times for monlayer thickness

2h 50m

- 2 [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 .
- 3 Wash buffer ⌚ 00:05:00 5m
- 4 [M] 1 Mass / % volume ⚗ Osmium Tetroxide **ProSciTech** in buffer ⌚ 00:30:00 30m
- 5 [M] 70 % volume ⚗ Ethanol ⌚ 00:10:00 10m
- 6 [M] 95 % volume ⚗ ethanol ⌚ 00:05:00 5m
- 7 [M] 100 % volume ⚗ ethanol ⌚ 00:05:00 5m



- 8 [M] 100 % volume dry ethanol ⌚ 00:05:00 5m
- 9 [M] 100 % volume Spurrs: [M] 100 % volume dry ethanol in 1:1 ratio ⌚ 00:30:00 30m
- 10 [M] 100 % volume Spurrs ⌚ 00:30:00 30m
- 11 [M] 100 % volume Spurrs ⌚ 00:30:00 30m
- 12 Oven polymerise 🌡 65 °C ⌚ Overnight