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Paraffin section tissue preparation

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Ashley Seifert¹

¹University of Kentucky



Ashley Seifert

University of Kentucky

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Protocol status: Working

We use this protocol and it's working

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Disclaimer

Note that any protocol involving animals should be reviewed and approved by your Institutional Animal Care and Use Committee (IACUC) before use.

Abstract

This protocol describes the procedure used by the Seifert Lab to prepare paraffin-embedded tissue sections.

Protocol materials

✕ Eppendorf tubes 1.5 mL uncolored **Eppendorf Catalog #022363204**

✕ Epredia 10% Neutral Buffered Formalin **Thermo Fisher Scientific Catalog #5701**

✕ 1X PBS (Phosphate-buffered saline)

✕ 70% Ethanol

✕ Formula R paraffin **Leica Biosystems Catalog #3801450**



- 1 For each sample, prepare a

⊗ Eppendorf tubes 1.5 mL uncolored **Eppendorf Catalog #022363204** with

⊗ Eppredia 10% Neutral Buffered Formalin **Thermo Fisher Scientific Catalog #5701**
- 2 Harvest tissue from animal and immerse in 10% NBF in prepared tube
- 3 Incubate

⌚ Overnight

 at

🌡️ 4 °C

 on a rocker (16-18 hr fix)
- 4 Remove the 10% NBF
- 5 Fill tube with

⊗ 1X PBS (Phosphate-buffered saline)
- 6 Wash at

🌡️ 4 °C

 on rocker for

⌚ 00:10:00
- 7 Drain and repeat steps 5 and 6 two more times for a total of 3 washes
- 8 Drain PBS and add

⊗ 70% Ethanol
- 9 Wash at

🌡️ 4 °C

 on rocker for

⌚ 00:10:00
- 10 Drain and repeat two more times for a total of 3 washes
- 11 After third wash, drain and replace with fresh 70% Ethanol
- 12 Store at

🌡️ 4 °C

 until ready for processing



- 13 Process for  Formula R paraffin **Leica Biosystems Catalog #3801450** embedding using a

Equipment

Histos 5

Tissue processor

Milestone

61577

https://www.milestonemedsrl.com/us/products/tissue-processing/?_gl=1*18i3dwh*_up*MQ..*_gs*MQ..&gclid=EAlaIQobChMIke623eqTigMVtszCBB3u3RJ2

- 14 Section samples at  5 μm