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NB110-90592 Protocol

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Immunohistochemistry Protocol for Ki67 Antibody (NB110-90592)

[[URL:https://www.novusbio.com/products/ki67-mki67-antibody_nb110-90592]][[Caption:Ki67 Antibody]] Immunohistochemistry

- 1. Slice fresh tissues in 3um sections
- 2. Formaldehyde fixation 50C for 1 hour

3. Dehydrate under 80%, 90%, 90%, 95%, 95%, 100% and 100% ethanol successively under 50C for 1 hour of each concentration

- 4. Xylene I 40C for 1 hour
- 5. Xylene II 40C for 1 hour
- 6. Paraffin I embedding 65 C for 1 hour
- 7. Paraffin II embedding 65 C for 1 hour
- 8. Paraffin III embedding 65 C for 1 hour
- 9. Deparaffinize and rehydrate sections as standard
- 10. Antigen retrieval using 10mM sodium citrate microwave for 20 minutes
- 11. Wash and PBS wash as standard
- 12. Add 100ul primary antibody diluted to 1:3200 separately and incubate at 4C overnight
- 13. Wash in PBS buffer for 3 times 5 minutes

14. Remove excess PBS buffer and incubate sections with biotinylated secondary antibody, incubate for 30 minutes at 30C

- 15. Wash in PBS buffer for 3 times 5 minutes
- 16. Remove excess PBS buffer and incubate sections with ABC at 30C for 30 minutes
- 17. Wash in PBS buffer for 3 times 5 minutes
- 18. Develop with DAB
- 19. Rinse slides in gently running tap water for 10 minutes