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NB110-59729 Protocol

Immunohistochemistry-Paraffin protocol for LOX Antibody (NB110-59729)

LOX Antibody: https://www.novusbio.com/products/lox-antibody_nb110-59729 Antigen Unmasking:

Bring slides to a boil in 10mM sodium citrate buffer (pH 6.0) then maintain at a sub-boiling temperature for 10 minutes. Cool slides on bench top for 30 minutes.

Staining:

- 1. Wash sections in dH2O three times for 5 minutes each.
- 2. Wash section in wash buffer (1X PBS/0.1% Tween-20 (1X PBST)) for 5 minutes.
- 3. Block each section with 100-400 ul blocking solution (1X PBST, 5% goat serum) for 1 hour at room temperature.
- 4. Remove blocking solution and add 100-400 ul primary antibody diluted in 1X PBST, 5% goat serum to each section. Incubate overnight at 4C.
- 5. Remove antibody solution and wash sections in wash buffer three times for 5 minutes each.
- 6. Add 100-400 ul biotinylated secondary antibody, diluted in 1X PBST, 5% goat serum. Incubate 30 minutes at room temperature.
- 7. Remove secondary antibody solution and wash sections three times with wash buffer for 5 minutes each.
- 8. Add 100-400 ul Streptavidin-HRP reagent to each section and incubate for 30 minutes at room temperature.
- 9. Wash sections three times in wash buffer for 5 minutes each.
- 10. Add 100-400 ul DAB substrate to each section and monitor staining closely.
- 11. As soon as the sections develop, immerse slides in dH2O.
- 12. Counterstain sections in hematoxylin.
- 13. Wash sections in dH2O two times for 5 minutes each.
- 14. Dehydrate sections.
- 15. Mount the coverslips.