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## $\label{eq:protocols} \textbf{Protocols}, \textbf{Publications}, \textbf{Related Products}, \textbf{Reviews and more:}$

www.novusbio.com/NBP1-77459

## NBP1-77459 Protocol

Immunohistochemistry-Paraffin Embedded Sections protocol specific for Heme Oxygenase 1 antibody (NBP1 -77495)

HO-1/HMOX1/HSP32 Antibody: https://www.novusbio.com/products/ho-1-hmox1-hsp32-antibody\_nbp1-77459 Immunohistochemistry-Paraffin Embedded Sections

## Antigen Unmasking:

Bring slides to a boil in 10 mM 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 deionized water three times for 5 minutes each.
- 2. Wash sections in wash buffer for 5 minutes.
- 3. Block each section with 100-400 ul blocking solution for 1 hour at room temperature.
- 4. Remove blocking solution and add 100-400 ul diluted primary antibody. 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 diluted secondary antibody. 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 deionized water.
- 12. Counterstain sections in hematoxylin.
- 13. Wash sections in deionized water two times for 5 minutes each.
- 14. Dehydrate sections.
- 15. Mount coverslips.