

# Novocastra™ Liquid Mouse Monoclonal Antibody Geminin

## Product Code: NCL-L-Geminin

<b>Intended Use</b>	FOR RESEARCH USE ONLY.
<b>Specificity</b>	Human Geminin.
<b>Clone</b>	EM6
<b>Ig Class</b>	IgG2a
<b>Antigen Used for Immunizations</b>	A prokaryotic recombinant protein containing the amino acids 5-209 of the mature geminin protein.
<b>Hybridoma Partner</b>	Mouse myeloma (p3-NS1-Ag4-1).
<b>Preparation</b>	Liquid tissue culture supernatant containing 15 mM sodium azide. Volume as indicated on vial label.
<b>Effective on Frozen Tissue</b>	Not fully evaluated.
<b>Effective on Paraffin Wax Embedded Tissue</b>	Yes (using heat induced epitope retrieval with Citrate-based buffer, pH 6.0: see overleaf).
<b>Recommendations on Use</b>	Immunohistochemistry: Typical working dilution 1:40. Citrate-based buffer, pH 6.0. 30 minutes primary antibody incubation at 25 °C. Polymer detection recommended. Western blotting: Not recommended.
<b>Positive Controls</b>	Immunohistochemistry: Tonsil.
<b>Staining Pattern</b>	Nuclear.
<b>Storage and Stability</b>	Store liquid antibody at 4 °C. Under these conditions, there is no significant loss in product performance up to the expiry date indicated on the vial label. Prepare working dilutions on the day of use.
<b>General Overview</b>	Geminin is a protein of 209 amino acids thought to be involved in the control of DNA replication via the interaction with Cdt1. Geminin is not found in the G1 phase of the cell cycle, but is first expressed in the G1 to S transition phase, with expression levels rising through the rest of the cell cycle and levels reaching a maximum during mitosis. Geminin is reported to be expressed in proliferating lymphocytes and epithelial cells eg germinal centers in tonsil, as well as in colon, spermatocytes, seminiferous tubules of the testes and within the basal layers of the squamous epithelium of the skin and breast.
<b>General References</b>	Wohlschlegel J A, Lutok J L, Weng A P, et al.. American Journal of Pathology. 161(1): 267–273 (2002). Wohlschlegel J A, Dwyer B T, Dhar S K, et al.. Science. 290 (5500): 2309–2312 (2000).



# Instructions for Use

## Heat Induced Epitope Retrieval Combined With Polymer Detection For Immunohistochemical Demonstration On Paraffin Sections

1. Cut and mount sections on slides coated with a suitable tissue adhesive.
2. Deparaffinize sections and rehydrate to distilled water.
3. Place sections in 0.5% hydrogen peroxide/methanol for 10 minutes (or use other appropriate endogenous peroxidase blocking procedure). Wash sections in tap water.
4. Heat 1500 mL of the recommended epitope retrieval solution (Citrate based pH 6.0 - Epitope Retrieval Solution unless otherwise indicated overleaf) in a stainless steel pressure cooker until boiling. Cover but do not lock lid.
5. Position slides into metal staining racks (do not place slides close together as uneven staining may occur) and lower into pressure cooker ensuring slides are completely immersed in epitope retrieval solution. Lock lid.
6. When the pressure cooker reaches operating temperature and pressure (after about 5 minutes) start a timer for 1 minute (unless otherwise indicated on the data sheet).
7. When the timer rings, remove pressure cooker from heat source and run under cold water with lid on. **DO NOT OPEN LID UNTIL THE INDICATORS SHOW THAT PRESSURE HAS BEEN RELEASED.** Open lid, remove slides and place immediately into a bath of tap water.
8. Wash sections once using fresh Tris-Buffered Saline (TBS, pH 7.6) buffer for 5 minutes.
9. Place sections in diluted normal serum (eg NCL-G-SERUM) for 10 minutes.
10. Incubate sections with primary antibody.
11. Wash twice, each time using fresh TBS buffer for 5 minutes.
12. For visualization of the bound primary antibody, follow instructions supplied with the Polymer Detection System.
13. Counterstain with hematoxylin (if required), dehydrate and mount.

*\* (In most applications, Phosphate Buffered Saline, pH 7.6, can be used instead of TBS, pH 7.6).*

### Safety Note

To ensure the correct and safe use of your pressure cooker, PLEASE READ THE MANUFACTURER'S INSTRUCTIONS.