# Novocastra™ Liquid Mouse Monoclonal Antibody Immunoglobulin D

## Product Code: NCL-L-IgD

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| **Intended Use** | FOR RESEARCH USE ONLY. |
| **Specificity** | Delta chain of human Immunoglobulin D |
| **Clone** | DRN1C |
| **Ig Class** | IgG1 |
| **Antigen Used for Immunizations** | Prokaryotic recombinant protein corresponding to 222 amino acids of the N terminus of the Delta heavy chain constant region. |
| **Hybridoma Partner** | Mouse myeloma (p3-NS1-Ag4-1) |
| **Preparation** | Liquid tissue culture supernatant containing 15 mM sodium azide.<br>Volume as indicated on vial label. |
| **Effective on Frozen Tissue** | Not evaluated. |
| **Effective on Paraffin Wax Embedded Tissue** | Yes (using heat induced epitope retrieval with Tris/EDTA based buffer, pH 9.0: see overleaf) |
| **Recommendations on Use** | Immunohistochemistry: Typical working dilution 1:1000. Heat induced epitope retrieval technique using Tris/EDTA-based buffer, pH 9.0. 30 minutes primary antibody incubation at 25 °C. Polymer detection recommended. Technical Note: The use of PBS-based diluents may result in increased background staining. Western Blotting: 1:25–1:100 (ECL™, Amersham Pharmacia Biotech). |
| **Positive Controls** | Immunohistochemistry: Tonsil<br>Western Blotting: Tonsil |
| **Staining Pattern** | Cytoplasmic and Membrane. |
| **Storage and Stability** | 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. |
| **General Overview** | IgD, together with IgM, are the major immunoglobulins expressed on the surface of B cells where it seems they may operate as mutually interacting antigen receptors for the control of lymphocyte activation and suppression. The greater susceptibility of IgD to proteolysis in combination with antigen could well be implicated in such a function. |
| **General References** | Geisberger R, Lamers M and Achatz G. Immunology. 118:429–437 (2006).<br>Preud'homme J, Petit I, Barra A, et al. Molecular Immunology. 37:871–887 (2000).<br>Vladutiu A. Clinical and diagnostic laboratory immunology. 7(2):131–140 (2000). |

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BIOSYSTEMS

# 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.