

## ANTI-BCL2 (CLONE: 100/D5)

<b>CATALOG ID</b>	<b>DESCRIPTION</b>
MM003-3D, 6D	3.0mL and 6.0mL RTU
MM003-AA, CC	0.2mL and 1.0 mL Conc.

<b>ALTERNATIVE NAME</b>	Bcl-2
<b>CLONE</b>	100/D5
<b>SPECIES</b>	Mouse
<b>ISOTYPE</b>	Mouse IgG1
<b>TISSUE CONTROL</b>	Non-Hodgkin's Lymphoma & Tonsil
<b>EPITOPE/ IMMUNOGEN</b>	BCL-2
<b>CELL LOCALIZATION</b>	Endoplasmic reticulum, outer mitochondrial membrane & Nuclear membrane
<b>SPECIES REACTIVITY</b>	Human & Mouse
<b>DILUTION RANGE</b>	Assay dependent
<b>DILUENT</b>	Antibody Diluent Standard
<i>Supplied as Buffer with protein carrier &amp; preservative</i>	

### INTENDED USE

BioMarq BCL2 antibody is used for *in vitro* diagnostic use only. This antibody is designed for the specific identification of BCL2 protein in formalin-fixed paraffin-embedded tissue sections. The results using this product should be interpreted by a qualified pathologist in conjunction with the patient's relevant clinical history, other diagnostic tests and proper controls.

### PRODUCT DESCRIPTION

This antibody recognizes a protein of 25-26kDa, identified as the bcl-2  $\alpha$  oncoprotein. It shows no cross-reaction with Bcl-x or Bax protein. Bcl-2 family proteins are important regulators of apoptosis. Studies confirm that normal thyroid follicular cells expressed Bcl-2 and Bak, but not Bax (12, 17, 18), suggesting that Bcl-2 and Bak expression in differentiated normal thyroid follicular cells and apoptotic cells. The determination of prognosis for B-Non-Hodgkin's lymphoma (NHL) is known to be related to the multiple

differences in tumor cell biology. Bcl-2 and Bcl-6 are two markers linked to germinal center B cells. Bcl-2 and Bcl-6 can be used as prognostic marker in NHL.

### PRINCIPLE OF PROCEDURE

Immunohistochemistry (IHC) is a method for detecting antigens or haptens in cells of a tissue section by exploiting the principle of antibodies binding specifically to antigens in biological tissues. The antibody-antigen binding can be visualized in different methods. Enzymes, such as Horseradish Peroxidase (HRP) or Alkaline Phosphatase (AP), are commonly used to catalyze a color-producing reaction. IHC is widely used technique which makes it possible to visualize the distribution and localization of specific cellular components within cells and in proper tissue context. There are numerous IHC methods that can be used to localize antigens. The method selected should include consideration of parameters such as the specimen types and assay sensitivity.

### IHC RECOMMENDED PROTOCOL

**DeParaffinization & Hydration:** DeParaffinization & Hydration is done using two grades of xylene & ethanol. Rinse in distilled water & follow next steps given

**Pretreatment Solution:** Perform heat Retrieval using BioMarq's Epitope Retrieval 1 (Catalog No PS001). (Refer to BioMarq's Epitope Retrieval 1 datasheet for specific instructions).

**Peroxide Block:** Incubate for 10 minutes with BioMarq EP Block (Catalog No BR001).

**Protein Block (Optional):** Incubate for 5-10 minutes at RT with BioMarq Protein Block (Catalog No BR002).

**Primary Antibody:** Incubate with Anti- BCL-2 antibody (Catalog No MM003) for 30-60 minutes at RT.

**Probe:** Incubate for 20 minutes at RT with a BioMarq Histochemistry probe (Catalog No HP001).

**Secondary Antibody:** Incubate for 20 minutes at RT with a BioMarq Polymer HRP antibody (Catalog No SA001).

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**Substrate/Chromogen:** Incubate sections in DAB solution for 5-7 minutes.

**Counterstain:** Stain with BioMarq Hematoxylin solution (Catalog No CS001) for 3-5min.

**Mounting Solution:** Mount the slides with BioMarq XY-Mount (Catalog No MS002) or using BioMarq T-Mount (Catalog No MS003).

### TECHNICAL NOTE

This antibody staining has been standardized with BioMarq **IHC DETECTION KIT** (Catalog No DA001).

Ensure after each step slides are washed with BioMarq Immuno Wash Standard (Catalog No WB001) except peroxide Block step. Follow the instructions in the wash buffer data sheet for 1X solution preparation.

Follow the Antibody specific protocol recommendations provided in the data sheet. If atypical results occur, contact BioMarq Technical Support at 040-29702960.

### STORAGE AND STABILITY

Store at 2-8°C. Do not freeze. Not to be used beyond the expiration date prescribed on label.

### QUALITY CONTROL

For Quality Control purpose, each lot of this antibody is tested by immunohistochemistry using, formalin-fixed, paraffin-embedded **Non-Hodgkin's Lymphoma** biopsy as control tissue. Users can also procure the Qualified Positive Control Slides available from BioMarq for their Quality Control purpose.

### PRECAUTIONS

The material contains 0.05% Sodium azide as preservative. Although the quantity of azide is very small, appropriate care should be taken when handling this material. Specimens should be handled carefully before and after the assay to avoid transmission of infection and disposed of with proper precautions.

Microbial contamination of reagents may yield nonspecific staining.

For detailed safety information related to BioMarq Products, please refer to appropriate safety data sheets (SDS) available online at [www.biomarq.net](http://www.biomarq.net)

### LIMITATIONS

Factors which affect Immunohistochemical staining include the fixation process, Epitope-retrieval method, incubation times, tissue section thickness and detection kit used. Detection systems other than recommended by Biomark when used results may vary due to the varied sensitivity of reagents and recommended incubation times. The recommendations and protocols mentioned in the datasheet are based on exclusive use of BioMarq products. Ultimately, it is the responsibility of the investigator to determine optimal conditions. The clinical interpretation of any positive or negative staining should be evaluated within the context of clinical presentation, morphology and other histopathological criteria by a qualified pathologist.

### REFERENCES

1. Mari Koga et al, Immunohistochemical Analysis of Bcl-2, Bax, and Bak Expression in Thyroid Glands from Patients with Subacute Thyroiditis, The Journal of Clinical Endocrinology & Metabolism, 1999 June.
2. Hanan Mohamed Mahmoud et al, Significance of Bcl-2 and Bcl-6 immunostaining in B-Non Hodgkin's lymphoma , Hematology Reports, 2011 Oct 19 .