For Research Use Only.
Not for use in diagnostic procedures.

MBL

MONOCLONAL ANTIBODY

HLA-DR

Code No.	Clone	Subclass	Quantity	Form
K0019-1	LN-3	Mouse IgG2b	200 µL	Liquid

SOURCE: This antibody was concentrated from hybridoma (clone LN-3) supernatant. This hybridoma was established by fusion of mouse myeloma cell p3-NS1-Ag4-1 with Balb/c mouse splenocyte immunized with activated human peripheral blood mononuclear cells.

PRODUCT: 200 µL aliquot of concentrate antibody from the supernatant with preservative (0.1% sodium azide)
*Azide may react with copper or lead in plumbing system to form explosive metal azides. Therefore, always flush plenty of water when disposing materials containing azide into drain.

STORAGE: This antibody solution is stable for one year from the date of purchase when stored at 4°C.

REACTIVITY: This antibody reacts with human HLA-DR antigen.

APPLICATION:
Immunohistochemistry; 1:50 - 1:100
Usually, heat treatment or enzyme treatment is not required. However, heat treatment may enhance staining intensity.
Microwave oven; 2 times for 10 minutes each in citrate buffer (pH 6.5)

Detailed procedure is provided in the following **PROTOCOL**.

SPECIES CROSS REACTIVITY:

Species	Human	Mouse	Rat
Tissues	Tonsil	Not tested	Not tested
Reactivity on IHC	+		

INTENDED USE:
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REFERENCE:

1) Marder, R.J., et al. *Lab. Invest.* **52**, 497-504 (1985)
2) Ioachim, H.L., et al. *Am. J. Surg. Pathol.* **20**, 64-71 (1996)
3) Fullen, D.R., et al. *J. Cutan. Pathol.* **25**, 553-558 (1998)

Clone LN-3 is used in these references.

PROTOCOL:
Immunohistochemical staining for paraffin-embedded sections: SAB method

1) Deparaffinize sections with xylene 3 times for 3-5 minutes each.
2) Wash the slides with Ethanol 3 times for 3-5 minutes each.
3) Wash the slides with PBS 3 times for 3-5 minutes each.
4) Remove the slides from the citrate buffer and cover each section with 3% H_2O_2 for 10 minutes at room temperature to block endogenous peroxidase activity. Wash 3 times in PBS for 5 minutes each.
5) Remove slides from PBS, wipe gently around each section and cover tissues with of Protein Blocking Agent (Ultratech HRP Kit; IMMUNOTECH Cat# IM-2391) for 5 minutes to block non-specific antibody staining. Do not wash.
6) Tip off the blocking buffer, wipe gently around each section and cover tissues with of the anti-HLA-DR monoclonal antibody diluted with PBS containing 1% BSA (1:50 - 1:100).
7) Incubate the section for 1 hour at room temperature.
8) Wash the slides 3 times in PBS for 5 minutes each.
9) Wipe gently around each section and cover tissues with Polyvalent Biotinylated antibody (Ultratech HRP Kit). Incubate for 10 minutes at room temperature. Wash as in step 8).
10) Wipe gently around each section and cover tissues with of Streptavidin-Peroxidase (Ultratech HRP Kit). Incubate for 10 minutes at room temperature. Wash as in step 8).
11) Visualize by reacting for 10-20 minutes with substrate solution containing 7.5mg DAB, 40 µL of 30% H_2O_2 in 150 mL PBS. *DAB is a suspect carcinogen and must be handled with care. Always wear gloves.
12) Wash the slides in water for 5 minutes.
13) Counter stain in hematoxylin for 1 minute, wash the slides 3 times in water for 5 minutes each, and then immerse the slides in PBS for 5 minutes. Dehydrate by immersing in Ethanol 3 times for 3 minutes each, followed by immersing in Xylene 3 times for minutes each.
14) Now ready for mounting.

(Positive controls for immunohistochemistry, Tonsil)

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MBL MEDICAL & BIOLOGICAL LABORATORIES CO., LTD.
URL https://ruo.mbl.co.jp
e-mail support@mbl.co.jp, TEL 052-238-1904