

Code No. 10777

**Anti-Human
Gd-IgA1(KM55) Rat IgG MoAb**

- ① Paraffin-embedded sections of 3 μ m thickness
- ② Deparaffinization with a series of xylene and ethanol concentrations and subsequent rehydration

Xylene	10min.	x2
100% Ethanol	5min.	x3
90% Ethanol	5min.	x1
70% Ethanol	5min.	x1
50% Ethanol	5min.	x1
Rinse with distilled water	5min	
- ③ Antigen retrieval : 0.05% bacterial protease subtilisin A (Sigma-Aldrich, Tokyo, Japan) dissolved in 5mmol/l tris (hydroxymethyl)-aminomethane buffer (pH7.6) room temp. 2hr
***This process is extremely important. If the dose of bacterial protease subtilisin A is not enough, KM55 mAb staining will not be succeeded.**
- ④ Rinse with distilled water.
- ⑤ Block with Protein Block (DAKO, Tokyo, Japan) . room temp. 30min
- ⑥ 1st Ab : Anti-Human Gd-IgA1(KM55) Rat IgG 100 μ g/mL 37°C 60min
- ⑦ Several washes with phosphate-buffered saline and triethanolamine-buffered saline containing 0.05% tween-20.
- ⑧ 2nd Ab: Alexa Fluor 555(or Other colors)-conjugated goat anti-rat IgG antibody (1:1000; Life Technologies, Carlsbad, CA). 37°C 30min
- ⑨ Several washes with phosphate-buffered saline and triethanolamine-buffered saline containing 0.05% tween-20.
- ⑩ Seal with Fluoromount (Diagnostic BioSystems, Pleasanton,CA).

For research use only, not for use in diagnostic procedures.