

FUJIFILM**Wako**Code No. 012-26723 (10 μ L)
016-26721 (50 μ L)**Anti Iba1, Monoclonal Antibody (NCNP24)**
抗 Iba1, モノクローナル抗体 (NCNP24)**[Background]**

Iba1 is a protein highly expressed in microglia and macrophages with a molecular weight of about 16.7 kDa¹⁾. The protein is a commonly known microglial marker in the nervous system. This item is a mouse monoclonal antibody that reacts with Iba1²⁾.
For research use only. Not for use in diagnostic procedure.

[Description]

[Purification]	Purified from the hybridoma supernatant by Protein A-affinity chromatography
[Reactivity]	Reacts with Iba1
[Antigen]	Synthetic peptide corresponding to the C-terminus of Iba1
[Clone No.]	NCNP24
[Species cross reactivity]	Mouse, rat and marmoset (Other species have not been tested)
[Host]	Mouse
[Isotype]	IgG1 \cdot κ
[Concentration]	Indicated on the label.
[Formulation]	1 \times TBS (pH 7.4) aqueous solution with 50% glycerol containing 0.05% sodium azide.

[Applications]

Immunohistochemistry (mouse, rat and marmoset frozen section *, DAB**) 1 : 500~2,000

Immunohistochemistry (rat frozen section *, fluorescent**) 1 : 100

*Paraffin section is not stained.

**For fluorescent staining, amplify the signal using biotinylated anti-mouse IgG and streptavidin-conjugated fluorescent dye (see recommended protocol). When using a secondary antibody labeled with a fluorescent dye, the positive signal weakens.

Optimal concentration should be determined by each laboratory for each application.

[Storage]

Store at -20°C .

Avoid repeated freeze and thaw.

[Package]

012-26723	10 μ L
016-26721	50 μ L

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[Recommended protocol (frozen section, DAB)]

1. Mouse and/or rat are perfusion-fixed with 4% paraformaldehyde, replaced sucrose, and prepared 50 μ m brain section by microtome.
2. Wash : 0.3% TritonX-100 in PBS, 5 min \times 3
3. Block of endogenous peroxidase : 3% hydrogen peroxide in 80% methanol, 20 min, -20°C
4. Wash : 0.3% TritonX-100 in PBS, 5 min \times 3
5. Blocking : 1% BSA, 0.3 % TritonX-100 in PBS, 2 hours, room temperature.
6. Primary antibody : Anti-Iba1 Mouse Monoclonal Antibody (NCNP24) (1 : 500), 1% BSA, and 0.3% TritonX-100 in PBS, Overnight, 4°C
7. Wash : PBS, 5 min \times 3
8. Secondary antibody : biotinylated anti-mouse IgG (1 : 200), 1% BSA, and 0.3% TritonX-100 in PBS, 2 hours, room temperature
9. Wash : PBS, 5 min \times 3
10. ABC : VECTASTAIN ABC kit (Vector Laboratory), 2 hours, room temperature
11. Wash : PBS, 5 min \times 3
12. DAB solution (10 mg/15 mL TBS)
13. Wash : PBS, 5 min \times 3
14. Mount

[Recommended protocol (frozen section, fluorescent)]

1. Rat is perfusion-fixed with 4% paraformaldehyde, replaced sucrose, and prepared 50 μ m-brain section by microtome.
2. Wash : 0.3% TritonX-100 in PBS, 5 min \times 3
3. Blocking : 1% BSA and 0.3% TritonX-100 in PBS, 2 hours, room temperature
4. Primary antibody : Anti Iba1, Mouse Monoclonal Antibody (NCNP24) (1 : 100), 1% BSA, and 0.3% TritonX-100 in PBS, overnight, 4°C
5. Wash : 0.3% TritonX-100 in PBS, 5 min \times 3
6. Secondary antibody : biotinylated anti-mouse IgG (1 : 200), 1% BSA, and 0.3% TritonX-100 in PBS, 2 hours, room temperature
7. Wash : 0.3% TritonX-100 in PBS, 5 min \times 3
8. Streptavidin Alexa Fluor 488 (1 : 1,000), 1% BSA, and 0.3% TritonX-100 in PBS, 2 hours, room temperature
9. Wash : 0.3% TritonX-100 in PBS, 5 min \times 3
10. Mount

[References]

- 1) Imai, Y., Iyata, I., Ito, D., Ohsawa, K. and Kohsaka, S. : *Biochem. Biophys. Res. Commun.*, **224**, 855 (1996).
- 2) Daniel Lee, C. Y. *et al.* : *Neuron*, **97**, 5, 1032 (2018).

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