



Qty: 100 µg/200 µl

## Mouse anti-Ankyrin G

For Research Use Only

Catalog No. 33-8800

Lot No. See product label

## Mouse anti-Ankyrin G

### FORM

This monoclonal antibody is supplied as a 200 µl aliquot at 0.5 mg/ml in phosphate buffered saline (pH 7.4) containing 0.1% sodium azide. The antibody is Protein-A purified from mouse ascites.

**CLONE:** 4G3F8

**ISOTYPE:** IgG<sub>1</sub>-kappa

### IMMUNOGEN:

A synthetic peptide derived from the spectrin-binding domain of the human ankyrin G protein.

### SPECIFICITY

This monoclonal antibody is specific for the ankyrin G protein and does not cross-react with related ankyrin isoforms. The antibody reacts with all splice forms of Ankyrin G containing a spectrin-binding domain, and identifies a band between ~200 – 300 kDa on Western blots. Ankyrin G is a marker of axon initial segments, nodes of Ranvier, sites of cell-cell contact on epithelial cells, and intracellular membranes of some cells.

### REACTIVITY

Sample	ELISA (Native)	Immuno fluorescence (frozen) <sup>(1,4)</sup>	Immuno- precipitation (native) <sup>(5)</sup>	Western Blotting
Human	+++ <sup>1</sup>	+++	+++	+++
Mouse	++	+	+++	+
Rabbit	ND	++	ND	ND
Rat	++	ND	ND	++
Dog	ND	+++	ND	+++
Immunogen	+++	ND	ND	ND

\* confirmed with HeLa cell lysates.

### USAGE

Working concentrations for specific applications should be determined by the investigator. Appropriate dilutions will be affected by several factors, including secondary antibody affinity, antigen concentration, sensitivity of detection method, temperature and length of incubations, etc. We recommend the following ranges as starting points for this product.

**ELISA:** 0.1-1.0 µg/ml  
**Western blotting:** 1-3 µg/ml  
**Immunofluorescence** (frozen sections)<sup>(1,4)</sup>: 5-10 µg/ml  
**Immunoprecipitation** (native)<sup>(5)</sup>: 5 µg/ IP reaction

- For more information on immunofluorescence and immunoprecipitation assays with Invitrogen's Ankyrin G monoclonal antibody, please see the cited references.
- For Western blot analysis of rat brain, mouse brain, or MDCK cell lysates, add 0.3M sucrose to incubation buffer for best results.
- This antibody is sensitive to fixation conditions. The following conditions are recommended: 1% paraformaldehyde for perfusion and 4 hours of post-fixation (after perfusion). For Sciatic nerve 2% paraformaldehyde is recommended for 2 hours @ 4°C.

(cont'd)

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PI338800

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- For Immunofluorescence, the following procedure is recommended:
  - Permeabilize for 15 min. at 4°C in PBS containing 0.5% Triton X-100, 0.2% Tween-20, and 10 mg/ml BSA.
  - Wash with Wash Buffer (PBS containing 0.2% Tween-20) 3 times at 4°C.
  - Apply Mouse anti-Ankyrin G antibody in blocking buffer (PBS containing 0.2% Tween-20, and 10 mg/ml BSA).
  - Wash with Wash Buffer (PBS containing 0.2% Tween-20) 3-5 times at 4°C.
  - Apply secondary antibody for 2 hours at 4°C (a Biotin-conjugated secondary antibody is recommended).
  - Wash with Wash Buffer (PBS containing 0.2% Tween-20) 3-5 times at 4°C.
  - Apply FITC-conjugated Streptavidin for 2 hours at 4°C.
  - Wash with Wash Buffer (PBS containing 0.2% Tween-20) 3-5 times at 4°C.
  - Mount slides

## STORAGE

Store at 2-8°C for up to one month. Store at -20°C for long-term storage. Avoid repeated freezing and thawing.

## BACKGROUND

Ankyrin G is found in several tissues including brain where, due to tissue-specific RNA processing, it is expressed as two different isoforms, localized to axon initial segments and nodes of Ranvier. Ankyrin G is also found at sites of epithelial cell-cell contact and in the intracellular membranes of some cells. Ankyrin G isoforms contain a unique stretch of sequence highly enriched in serine and threonine residues immediately following a globular head domain. Ankyrin G is thought to participate in the maintenance and targeting of ion channels and cell adhesion molecules to the nodes of Ranvier and to axon initial segments.

## REFERENCES

- Mathis C, et al. *Development* 128:4881-4890, 2001.
- Kordeli E, et al. *J Biol Chem* 270(5):2352-2359, 1995.
- Kordeli E, et al. *J Cell Sci* 111:2197-2207, 1998.
- Martin S, et al. *J. Neurosci* 21(20) : 8119-8128, 2001.
- Malhotra JD, et al. *J Biol chem.* 277(29): 26681-26688, 2002

## RELATED PRODUCTS

<u>Product</u>	<u>Clone/PAD*</u>	<u>Catalog No.</u>
Ms x Ankyrin B	2.20	33-3700
Rb x Velis-1	ZMAL1	51-5000
Rb x Velis-2	ZMAL2	51-5400
Rb x Velis-3	ZMAL3	51-5600
Gt x PSD-93	B111	51-6800
Rb x PSD-93	ZMD.98	34-4700
Rb x PSD-95 (C-term)	PDZ95	51-6900
Sh x PSD-95 (N-term)	B122	51-6700
Rb x SAP97	PDZ97	51-7000
Rb x SAP102	PDZ102	51-7100
Protein A	Sepharose® 4B	10-1041
Rec-Protein G	Sepharose® 4B	10-1241

<u>Conjugate</u>	<u>ZyMAX™ Goat x Rabbit IgG (H+L)</u>	<u>ZyMAX™ Goat x Mouse IgG (H+L)</u>
Purified	81-6100	81-6500
FITC	81-6111	81-6511
TRITC	81-6114	81-6514
Cy™3	81-6115	81-6515
Cy™5	81-6116	81-6516
HRP	81-6120	81-6520
AP	81-6122	81-6522
Biotin	81-6140	81-6540

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