

Product Datasheet

Glutamate Antibody (S1) NBP2-29884

Unit Size: 0.1 ml

Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.

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NBP2-29884**Glutamate Antibody (S1)**

Product Information	
Unit Size	0.1 ml
Concentration	Please see the vial label for concentration. If unlisted please contact technical services.
Storage	Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	S1
Preservative	0.07% Sodium Azide
Isotype	IgM
Purity	Ion exchange chromatography
Buffer	PBS
Product Description	
Host	Mouse
Species	Rat
Specificity/Sensitivity	Glutamate The cross-reactivities were determined using an ELISA test by competition experiments with the following compounds: Compound Cross-reactivity Glutamate-G-BSA 1 Aspartate-G-BSA 1/100,000 GABA-G-BSA 1/100,000 Glutamate 1/100,000 Abbreviations: (G) Glutaraldehyde (BSA) Bovine Serum Albumin NOTE: Antibody reactivity REQUIRES glutaraldehyde fixation thus some glutaraldehyde (0.5%-2.0%) needs to be included in the tissue fixation procedure inorder for the proper reactivity.
Immunogen	Glutamate-Glutaraldehyde-BSA.
Product Application Details	
Applications	Immunohistochemistry
Recommended Dilutions	Immunohistochemistry 1:500-1:2500



Application Notes

Immunohistochemistry: using free floating sections by the PAP technique on rat hippocampus. **PROTOCOL for Glutamate Detection by Immunohisto/cytochemistry.** Example for a rat brain. 1. **SOLUTIONS TO BE PREPARED** - Solution must be prepared as needed. Solution A: 0.1M cacodylate, 10g/L sodium metabisulfite, pH 6.2. Solution B: 0.1M cacodylate, 10g/L sodium metabisulfite, 3-5% glutaraldehyde, pH 7.5. 2. **RAT PERFUSION** - The rat is anaesthetized with sodium pentobarbital or Nembutal and perfused intracardially through the aorta using a pump with Solution A (30 mL): 150-300 mL/min, Solution B (500 mL): 150-300 mL/min. 3. **POST FIXATION:** 15 to 30 minutes in Solution B, then 4 soft washes in 0.05M Tris with 8.5 g/L sodium metabisulfite, pH 7.5 (Solution C). 4. **TISSUE SECTIONING:** Vibratome or cryostat sections can be used. 5. **REDUCTION STEP:** Sections are reduced with Solution C containing 0.1M sodium borohydride for 10 minutes. The sections are washed 4 times in solution C without sodium borohydride. 6. **APPLICATION OF GLUTAMATE ANTIBODY:** Use a final dilution of 1:2,500-1:10,000 in Solution C containing 0.1% Triton X100 and 2% non-specific serum. Incubate 12 sections per 2 mL diluted antibody overnight, +2-8C. Then wash the sections three times for 10 minutes each in Solution C. (Note that the antibody may be usable at a higher dilution. This should be explored to minimize the possibility of high background. Additionally, note that a change in the buffering system as indicated in the protocol may change the background and antibody recognition). The specific reaction is then revealed by PAP procedure. 7. **SECOND ANTIBODY:** Incubate the sections with a 1:50 to 1:200 dilution of goat anti-mouse in Solution B containing 1% non-specific serum for either 3 hrs at 20C or 1-2 hr at 37C. Then wash the sections, 3 times, for 10 minutes each with Solution C. 8. **PAP:** Incubate the sections with the appropriate dilution of peroxidase anti-peroxidase (for free floating method) in Solution C for 1-2 hours at 37C. Then wash sections 3 times for 10 min each in solution C. 9. **VISUALIZATION:** The antigen-antibody complexes are visualized using DAB-4-HCl (25 mg/100 mL) (or other chromogen) in 0.05M Tris and filtrated; 0.05% hydrogen peroxide is added. Incubate the sections for 10 minutes at room temp. Stop the reaction by transferring the sections to 5 mL 0.05M Tris. Mount sections on chrome-alum coated slides. Dry overnight at 37C. Rehydrate sections using conventional histological procedures. Coverslip using rapid mounting media. For research use only; not for use as a diagnostic.



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Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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