北京天成新脉生物技术有限公司
AbMax Biotechnology Co., LTD
北京经济技术开发区科创十四街 99 号 18 号楼
2 单元 201
99 Kechuang 14th St, Bldg 18, 2-201
BDA, Beijing, China 101111
Tel: 86-10-59755729, Fax: 86-10-59755718
E-mail:info@antibodychina.com
www.abmaxcn.com

Mouse anti-Rabies Virus, Biotin-conjugated (mouse monoclonal IgG)

Catalog # 12-0061

Immunogen: Attenuated rabies virus

Specificity: Recognizes the N protein of rabies virus from SRV9, aG, and all wild strains tested.

Formulation: 0.1mg of biotinlyated mouse monoclonal IgG in **0.1ml** of 0.01M PBS, pH 7.2, 0.15M NaCl, 0.01% sodium azide, 20% glycerol .

Protein was determined by OD_{280} absorbance.

Physical State: Frozen liquid

Storage: Store the product at -20℃. Product is stable for about 6 weeks at 2-8℃ as an undiluted liquid. Prepare working dilution prior to use. Avoid repeated freezing and thawing.

FOR IN VITRO RESEARCH USE ONLY

Reference

1. Muyang Wang, et al. “Development of Diagnostic Sandwich ELISA Kit for Rabies Virus”. (submitted)

Disclaimer: The product was co-developed with Diagnostic Laboratory for Rabies and Wildlife-associated Viral Zoonoses, Ministry of Agriculture, China.

Application

Biotinylated anti-nucleocapsid monoclonal antibody is used for detection of rabies virus in impression smears of animal brain tissues.

Composition

Biotinylated anti-nucleocapsid monoclonal antibody:0.1mL.

Preservative

20%Glycerol PBS containing0.01%sodiumazide .

Direct Rapid Immunohistochemical Test (DRIT)

Methods

1.Label the microscope slides:Place a small piece of selected brain issue on a wooden spatula and make an impression smear directly onto the slide. The routine use of positive and negative controls is essential.

2.Air-dry slides at room temperature.

3.Immerse slides in 10% buffered formalin at room temperature in biosafety cabinet for 10 minutes. After this step slides can be removed from the cabinet and rest of the procedure can be carried out on bench.

4.Remove the slides from formalin and dip-rinse slides several times to wash off any fixative in wash buffer PBSwith1%Tween80(TPBS).

5.Immerse slides in 3% hydrogen peroxide for10minutes.

6.Remove hydrogen peroxide by dip-rinsing in TPBS. Transfer slides to the next rinse and repeat one more wash.

7.Air-dry the smear thoroughly.

8.Place the slides in a humidity chamber. Add 50μL biotinylated monoclonal antibodies (working dilution of 1:200 is recommended) to the slide by drop to cover the impression, then incubate at 37℃ for 10 minutes.

9.Shake off the conjugate. Dip-rinse slides with TPBS. Shake off TPBS and blot buffer from slide edges and surrounding area of the impression.

10.Place the slides back in the humidity chamber and cover the impressions with 20-50μL streptavidin-peroxidase complex according to Manufacture's instruction.

11.Incubate at room temperature for10minutes.

12.Shake off streptavidin-peroxidase and dip-rinse slides with TPBS. Shake off buffer and blot buffer from slide edges and surrounding area of the impression.

13.Incubate slides with peroxidase substrate, amino-ethylcarbizole (AEC) in a Humidity chamber. Add enough substrate by drop to cover the impression.

14.Incubate at room temperature for 10 minutes. After incubation, shake off substrate.

15.Dip-rinse slides in distilled water. Remove excess water by blotting slide edges and around the impression.

16. Counter stain with freshly prepared Haematoxylin for 5 minutes in humidity chamber.

17.Rinse with tap water to remove the stain.

18.Put the slides into the jar, fill the jar with tap water and leave for 10 minutes.

19. View slides by light microscopy, using a 20×objective to scan the field, and a 40×objective for higher power inspection (rabies virus antigen appears as red in clusions against the blue neuronal background, see the pictures below).

Appendix

Preparation of peroxidase substrate amino-ethylcarbizole(AEC)

Reagents：

1.Amino-ethylcarbizole(AEC)substrate,SIGMAno.A6926

2.N,N,Dimethyl formarnide GR, EM Science

3.Acetatebuffer,0.1M,pH5.2

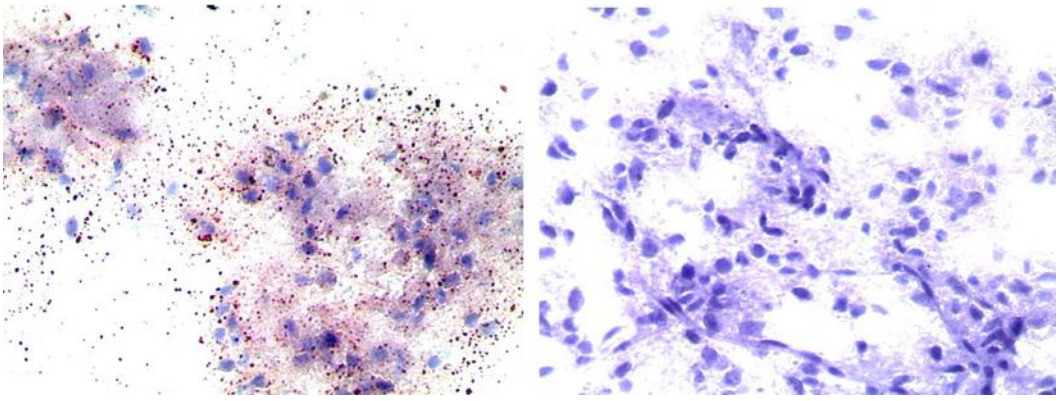
4.3%Hydrogenperoxide

To prepare AEC stock stock solution :

Dissolve 20mg 3-amino 9-ethylcarbazole (AEC) in 5 ml of N, N, dimethylformamide in a glass Wheaton jar.TheAEC stock should be stored at 4℃ for 1 to 2 months.

To prepare AEC working dilution:

Add 7 ml of acetate buffer, 0.5 ml of AEC stock solution and 0.075 ml 3% hydrogen peroxide to a 15 ml centrifuge tube. Filter mixture using a 10 ml syringe with syringe filter (0.45μm) into separate 15 ml centrifuge tube. Once made the mixture is only stable for 2-3 hours.



A. Rabies virus positive

B. Rabies virus negative