pbl assay science
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Certificate of Analysis

Anti-Human Interferon Beta, Clone MMHB-2 (MAb)

Catalog No: 21410-1
Lot No: 5982
Expiration: January 31, 2015
Size: 500 µg/vial

Description: Mouse Monoclonal Antibody against Human Interferon Beta
Clone: MMHB-2
Concentration: 0.5 mg/ml; after reconstitution with 1 ml sterile PBS
Reconstitution: Dissolve contents of the vial by injection of 1 ml sterile PBS
Buffer: Phosphate-buffered saline (PBS) containing 5% trehalose prior to lyophilization
Endotoxin: <1 EU/µg
Antigen: Human Interferon Beta
Isotype: Mouse IgG_1
Purification Method: Protein A or G affinity chromatography
Specificity: Neutralizes human interferon beta; does not neutralize human interferon alpha or gamma

Assay Used to Measure Bioactivity: The exact concentration of antibody required to neutralize human interferon beta activity is dependent on the cytokine concentration, cell type, growth conditions and type of activity studied. The Neutralization $Dose_{50}$ (ND_{50}) for this antibody is defined as that concentration of antibody required to yield one-half maximal inhibition of the cytokine activity on a responsive cell line, when that cytokine is present at a concentration just high enough to elicit a maximum response. The ND_{50} for this antibody on human (HeLa/EMCV) cells is ~7-21 µg/ml in the presence of 10 ng/ml of human interferon beta, based on the anti-viral assay. The specific conditions are described in the figure legends.

Due to the variation in ND_{50} values based on cell type and assay system, we recommend each user determine the neutralizing concentration of this antibody lot in their assay system. Using an A549/EMCV (cell/virus) system, we have not verified with reasonable consistency the neutralizing concentration of this antibody (the concentration required to inhibit the antiviral effect of human interferon beta by one half).

Tested Applications: Neutralization; Direct ELISA (0.5-1.0 µg/ml); Western Blot (1-2 µg/ml).
Optimal dilutions should be determined by each laboratory for each application.

Shipping Conditions: Wet Ice
Physical State of Product During Shipping: Lyophilized
Storage Conditions/Comments: Use a manual defrost freezer. Upon receipt, this product (as supplied) should be kept at -20 to -70°C until the expiration date listed above. After reconstitution, the contents of the tube should be apportioned in separate tubes so that freezing and thawing is kept to a minimum. After reconstitution, this product may be stored at 2 to 8°C for one month and -20 to -70°C for up to 6 months for retention of full activity. For more information on protein handling, visit our Resource Library at www.pblassaysci.com.

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Product Information:

Figure 1

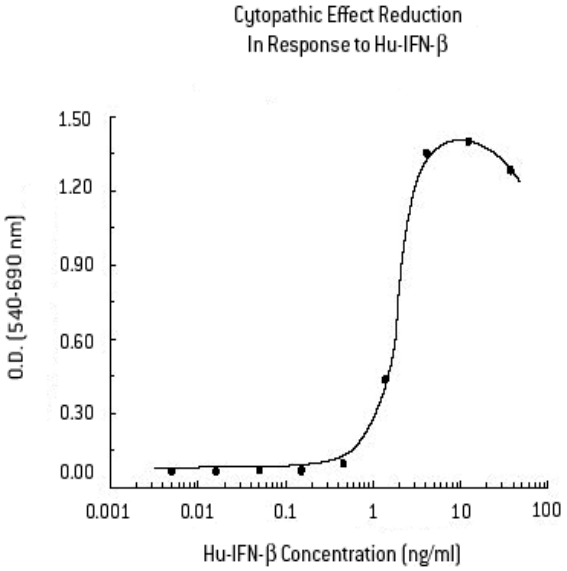

Figure 2

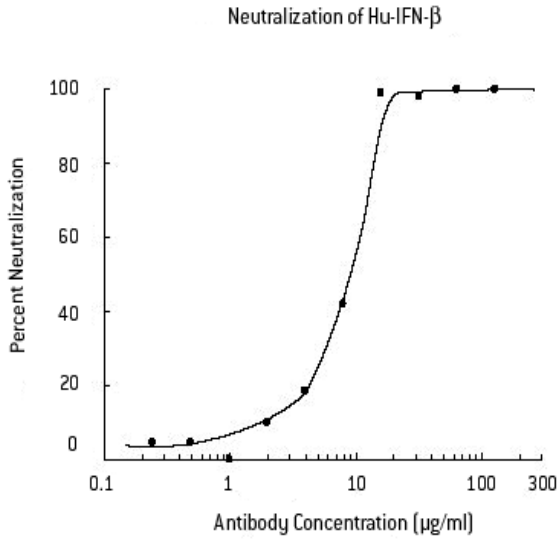

Figure 1. Human IFN-ɓ reduces the cytopathic effect of the lytic virus EMC in a dose-dependent manner, on the human cell line, HeLa. (Meager, A. 1987, *Lymphokines and Interferons, a practical approach,* Clemens, M.J. Morris, A.G. and A.J.H. Gearing, eds. IRL Press, p. 129). The ED_{50} for this effect is typically 2-5 ng/ml.

Figure 2. To measure the ability of the antibody to neutralize the bioactivity of the human interferon beta on HeLa cells, Hu-IFN-ɓ was added to various concentrations of the antibody. The antigen-antibody mixture was added to confluent cultures of the HeLa cells in a 96 well plate. The assay mixture in a total volume of 100 µl, containing antibody at the concentrations as indicated, Hu-IFN-ɓ at 10 ng/ml, was incubated at 37°C for 20-24 hours in a humidified CO_2 incubator. At the end of this incubation period, medium was aspirated from all wells and an appropriate titrated amount of the EMCV in pre-warmed culture medium was added to each test well. After another 20-24 hour incubation, the cells were fixed, stained and scored for cytopathic effect by measurement of optical densities in a microplate reader at 540 nm. The ND_{50} of the antibody is approximately 7-21 µg/ml.

Authorization

Released by: _____ Date: February 4, 2014