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Anti-Ephrin type-B receptor 4 EphB4 Antibody

Catalog Number: A00690

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Overview

Product Name	Anti-Ephrin type-B receptor 4 EphB4 Antibody
Reactive Species	Human, Mouse
Description	Boster Bio Anti-Ephrin type-B receptor 4 EphB4 Antibody catalog # A00690. Tested in WB, IHC, IF, ELISA applications. This antibody reacts with Human, Mouse.
Application	ELISA, IF, IHC, WB
Clonality	Polyclonal
Formulation	Liquid in PBS containing 50% glycerol, 0.5% stabilizing protein and 0.02% sodium azide. *This antibody is supplied in a stabilized formulation. Compatibility with conjugation reactions depends on the chemistry of the conjugation method used. For conjugation methods that are not compatible with the stabilizing components present in this formulation, a carrier-free antibody format is required.
Storage Instructions	Store at -20°C for one year. For short term storage and frequent use, store at 4°C for up to one month. Avoid repeated freeze-thaw cycles.
Host	Rabbit
Uniprot ID	P54760

Technical Details

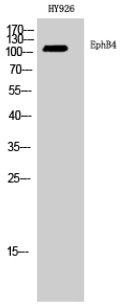
Immunogen	The antiserum was produced against synthesized peptide derived from human EPHB4. AA range:571-620
Cross Reactivity	No cross reactivity with other proteins.
Isotype	IgG
Form	Liquid
Concentration	1 mg/ml
Purification	Immunogen affinity purified

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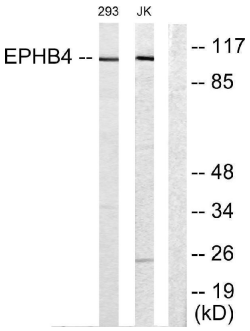
Suggested Dilutions	WB 1:500-1:2000 IHC 1:100-1:300 IF 1:200-1:1000 ELISA 1:20000

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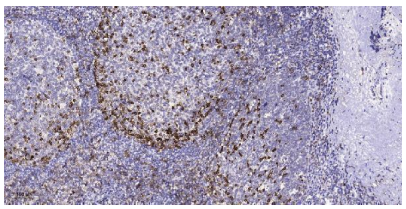
Anti-Ephrin type-B receptor 4 EphB4 Antibody (A00690) Images



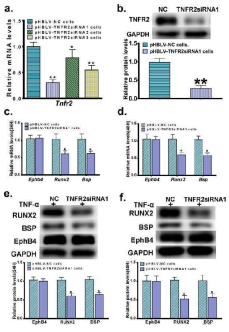
Western Blot analysis of HY926 cells using EphB4 Polyclonal Antibody diluted at 1:2000



Western blot analysis of lysates from Jurkat and 293 cells, using EPHB4 Antibody. The lane on the right is blocked with the synthesized peptide.



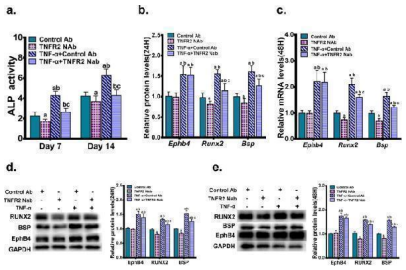
Immunohistochemical analysis of paraffin-embedded human tonsil. 1, Antibody was diluted at 1:200 (4° overnight). 2, Tris-EDTA, pH9.0 was used for antigen retrieval. 3, Secondary antibody was diluted at 1:200 (room temperature, 45min).



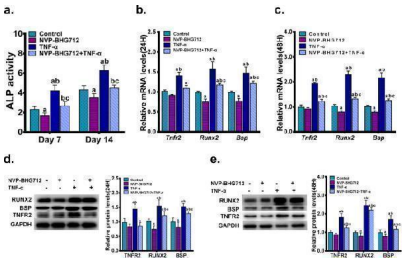
The effect of the lentivirus-mediated shRNA interference of TNFR2 on TNF-alpha-stimulated EphB4 expression and osteogenic differentiation. a MC3T3-E1 cells stably transduced with lentiviral particles were selected with puromycin and named as pHBLV-TNFR2siRNA1 cells, pHBLV-TNFR2siRNA2 cells, pHBLV-TNFR2siRNA3 cells and pHBLV-NC cells, respectively. The mRNA levels of Tnfr2 were determined in these cells, among which the pHBLV-TNFR2siRNA1 cells displayed the highest TNFR2 gene silencing efficiency and were selected to continue the following studies. b TNFR2 protein levels in pHBLV-TNFR2siRNA1 cells and pHBLV-NC cells. c , d mRNA levels of Ephb4 , Runx2 and Bsp in pHBLV-TNFR2siRNA1 cells and pHBLV-NC cells cultured in the osteogenic induction medium supplemented with 0.5 ng/ml TNF-alpha for 24 h (c) or 48 h (d). e , f Protein levels of EphB4, RUNX2 and BSP in pHBLV-TNFR2siRNA1 cells and pHBLV-NC cells cultured in the osteogenic induction medium supplemented with 0.5 ng/ml TNF-alpha for 24 h (e) or 48 h (f). *, $p < 0.05$ vs. the pHBLV-NC group; **, $p < 0.01$ vs. the pHBLV-NC group Index in PubMed under a CC BY license. PMID: 32299362

The effect of the impaired binding between TNF-alpha and

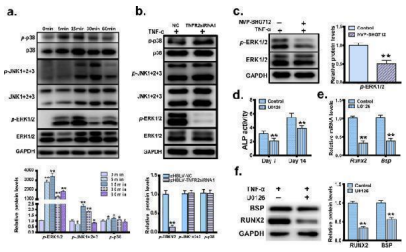
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TNFR2 on TNF-alpha-stimulated EphB4 expression and osteogenic differentiation. MC3T3-E1 cells were treated with an anti-mouse TNFR2/ CD120b/TNFRSF1B neutralizing antibody (TNFR2 NAb) at the concentration of 0.2 ug/ml, and were cultured in the osteogenic induction medium supplemented with or without 0.5 ng/ml TNF-alpha. Cells treated with 0.2 ug/ml of the normal rabbit IgG negative control antibody (control Ab) served as negative controls. (a) ALP activities were determined 7d or 14d after the treatment. (b, c) mRNA levels of Ephb4 , Runx2 and Bsp were determined after 24 h (b) or 48 h (c). (d, e) Protein levels of EphB4, RUNX2 and BSP were determined after 24 h (d) or 48 h (e). a, $p < 0.05$ vs. the control Ab group; b, $p < 0.05$ vs. the TNFR2 NAb group; c, $p < 0.05$ vs. the TNF-alpha + control Ab group Index in PubMed under a CC BY license. PMID: 32299362



The effect of inhibited EphB4 forward signaling on TNF-alpha-stimulated TNFR2 expression and osteogenic differentiation. (a) A potent inhibitor of EphB4 auto-phosphorylation, NVP-BHG712, was used to suppress EphB4 forward signaling. MC3T3-E1 cells were pretreated with 200 nM NVP-BHG712 in the regular culture medium for 1 h. Cells were then incubated in osteogenic induction medium supplemented with 200 nM NVP-BHG712 and/or 0.5 ng/ml TNF-alpha for 7d or 14d. MC3T3-E1 cells cultured in osteogenic induction medium served as controls. The ALP activities were determined. (b, c) MC3T3-E1 cells were pretreated with 200 nM NVP-BHG712 for 1 h in the regular culture medium, and then incubated in osteogenic induction medium supplemented with 200 nM NVP-BHG712 and/or 0.5 ng/ml TNF-alpha. Cells cultured in osteogenic induction medium served as controls. mRNA levels of Tnfr2 , Runx2 and Bsp were determined after 24 h (b) or 48 h (c) of incubation. (d, e) MC3T3-E1 cells were pretreated with 200 nM NVP-BHG712 for 1 h in the regular culture medium, and then incubated in osteogenic induction medium supplemented with 200 nM NVP-BHG712 and/or 0.5 ng/ml TNF-alpha. Cells cultured in osteogenic induction medium served as controls. Protein levels of TNFR2, RUNX2 and BSP were determined after 24 h (d) or 48 h (e) of incubation. a, $p < 0.05$ vs. the control group; b, $p < 0.05$ vs. the NVP-BHG712 group; c, $p < 0.05$ vs. the TNF-alpha group Index in PubMed under a CC BY license. PMID: 32299362



EphB4, TNFR2 and MAPK signaling pathways comprise a signaling axis to mediate the positive effect of TNF-alpha on osteogenic differentiation. a Levels of p38, p -p38, ERK1/2, p -ERK1/2, JNK1 + 2 + 3 and p -JNK1 + 2 + 3 in MC3T3-E1 cells treated with TNF-alpha for 0 min, 5 min, 15 min, 30 min and 60 min. b Levels of p38, p -p38, ERK1/2, p -ERK1/2, JNK1 + 2 + 3 and p -JNK1 + 2 + 3 in the pHBLV-TNFR2siRNA1 cells and the pHBLV-NC cells treated with or without 0.5 ng/ml TNF-alpha in regular culture medium for 15 min. c MC3T3-E1 cells were pretreated with or without 200 nM NVP-BHG712 in the regular culture medium for 1 h, and then 0.5 ng/ml TNF-alpha was added into the medium. The cells were incubated for another 15 min. Levels of ERK1/2 and p -ERK1/2 were

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determined. d-f MC3T3-E1 cells were cultured in the regular culture medium and pretreated with the ERK inhibitor U0126 (10 uM) for 1 h. The culture medium was then switched to the osteogenic induction medium supplemented with 0.5 ng/ml TNF-alpha and U0126 (10 uM). Cells treated without U0126 (10 uM) served as controls. ALP activities were determined 7d or 14d after the treatment (d). mRNA levels (e) and protein levels (f) of BSP and RUNX2 were determined 3 days after the treatment. *, $p < 0.05$ vs. the control group; **, $p < 0.01$ vs. the control group Index in PubMed under a CC BY license. PMID: 32299362

1 Publications Citing This Product

1. PubMed ID: 10.1186/s12860-020-00273-2, EphB4/ TNFR2/ERK/MAPK signaling pathway comprises a signaling axis to mediate the positive effect of TNF-alpha on osteogenic differentiation.

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