

Datasheet

Anti-GDF9 Clone 53/1

Product Name	Anti Human GDF9 53/1
Catalogue Number	53/1
Clone, Isotype	53/1, IgG1
Format	IgG
Tested Applications	ELISA, IHC, WB

Description:

GDF9 is plays a vital role in ovarian folliculogenesis, follicle development and fertility. Clone 53/1 can be used in assays to detect oocyte expression and has been shown to neutralize GDF9 biological activity (Gilchrist, R.B. et al.)

Product Details:

Form in stock: IgG, purified – 1.0 mg/mL. Also available as unpurified supernatant.

Host: Mouse

Specificity: Tuberculin coupled peptide with sequence VPAKYSPLSVLTIEPDGSIAYKEYEDMIATKC that recognizes an epitope with the EPDG sequence near the C-terminal region of human GDF9.

Human Histology positive control: Ovary

Fusion partner: Spleen cells from immunised Balb/c mice were fused with cells of the SP2/0 myeloma cell line.

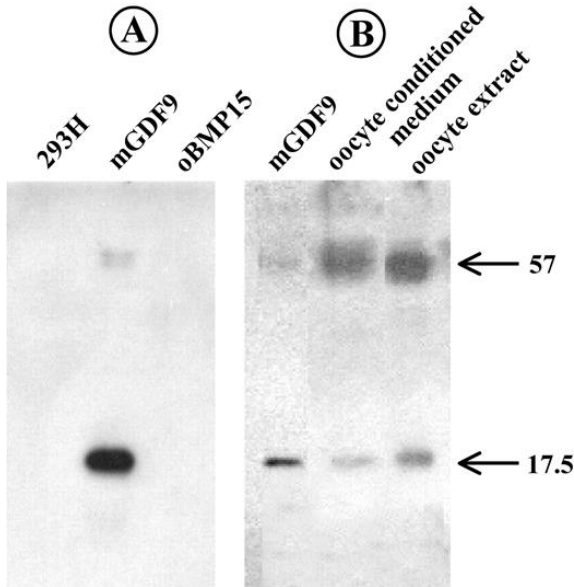
Storage: Store at +4°C or -20°C. Avoid repeated freezing and thawing.

Shelf life: 18 months from date of dispatch.

Regulatory/ Restrictions: For research and commercial purposes.

Applications	Suggested Dilution
Western Blot	1:100-1:1000
Immunohistochemistry	1:25-1:100
ELISA	Assay dependent

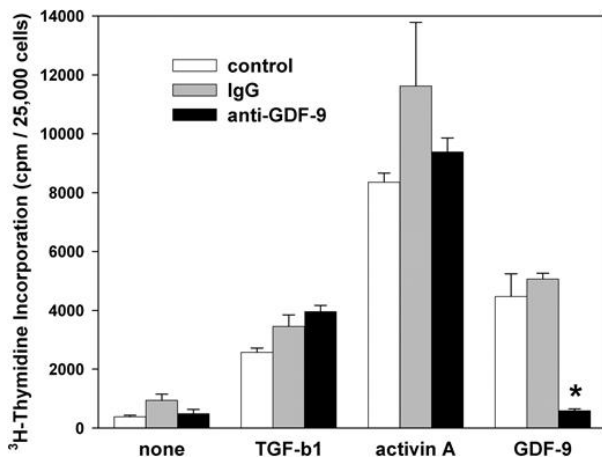
Applications:



Clone 53/1 used to detect GDF9 expression in oocyte-medium via **western blot**

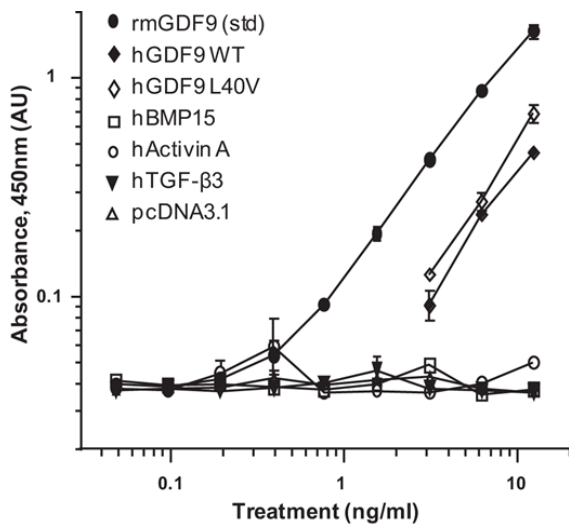
Image caption: GDF9 immunoblot analysis. **A)** Antibody specificity for GDF9 and **(B)** detection of oocyte-secreted GDF9. The recombinant proteins indicated (mGDF9 = mouse GDF9, oBMP15 = ovine BMP15) and oocyte extract or oocyte-conditioned medium were subjected to SDS-PAGE immunoblotting with the anti-GDF9 monoclonal antibody and detected using either ECL (**A**; recombinant proteins) or ECL Advance (**B**; oocyte products). The 57-kDa band is the GDF9 proprotein while the 17.5-kDa band represents the mature GDF9 monomer. 293H = control; conditioned medium from the untransfected human embryonic kidney 293H parent cell line (Gilchrist, R.B. et al.)

Image caption: GDF9 immunoblot analysis. **A)** Antibody specificity for GDF9 and **(B)** detection of oocyte-secreted GDF9. The recombinant proteins indicated (mGDF9 = mouse GDF9, oBMP15 = ovine BMP15) and oocyte extract or oocyte-conditioned medium were subjected to SDS-PAGE immunoblotting with the anti-GDF9 monoclonal antibody and detected using either ECL (**A**; recombinant proteins) or ECL Advance (**B**; oocyte products). The 57-kDa band is the GDF9 proprotein while the 17.5-kDa band represents the mature GDF9 monomer. 293H = control; conditioned medium from the untransfected human embryonic kidney 293H parent cell line (Gilchrist, R.B. et al.)



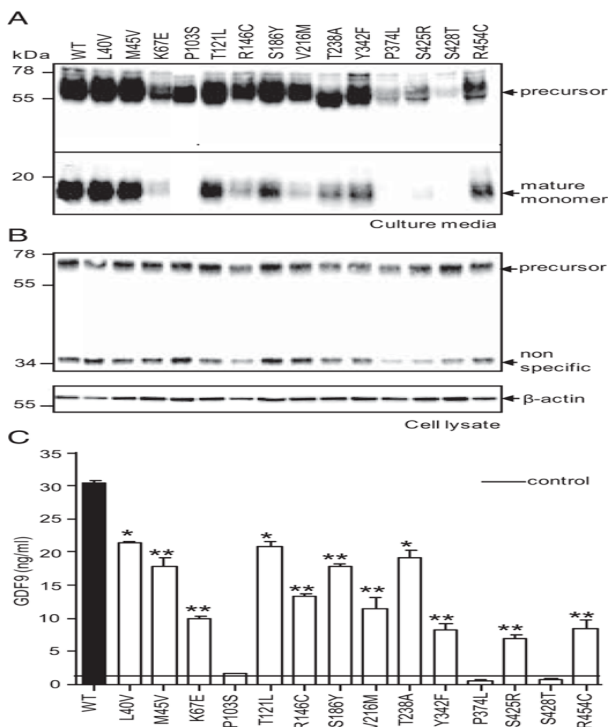
Biological neutralizing capacity of 53/1 by **ELISA**

Image caption: Biological neutralizing specificity of anti-GDF9-53 with some members of the TGFβ superfamily. Mouse MGC were cultured with human TGFβ1 (0.5 ng/ml), human activin A (50 ng/ml), or mouse GDF9 (40 ng/ml), either in the absence or presence of a high neutralizing dose of 40 μg/ml of mAb-GDF9-53 or 40 μg/ml human IgG. (Gilchrist, R.B. et al.)



Clone 53/1 used to detect GDF9 expression by ELISA

Image caption: GDF9 ELISA. A GDF9 ELISA was developed to measure the amount of GDF9 in HEK-293T conditioned medium. Recombinant mouse GDF9 (●) was used as a standard, and the specificity of the assay was assessed using a range of TGF-β family members; wild-type human GDF9 (◆), human GDF9 L40V (◇), human BMP15 (□), human activin A (○), and human TGF-β3 (▼). Dilutions of concentrated media from cells transfected with empty vector, pcDNA3.1 (r), were included as controls. The ELISA has a specificity of less than 0.1%, with a sensitivity of 0.2 ng/mL. Values represent mean ± SEM in duplicate, from a representative experiment. (Courtney, M et al.)



Clone 53/1 used to detect GDF9 expression in oocyte-medium by western blot

Image caption: ... Samples were detected under reducing conditions using GDF9 mAb53 specific for the mature domain. The 60-kDa GDF9 precursor and 20-kDa mature monomer are shown... (Courtney, M et al.)

Dilution used: 1:5000

References:

1. Gilchrist, R.B., Ritter, L.J., Cranfield, M., Jeffery, L.A., Amato, F., Scott, S.J., Myllymaa, S., Kaivo-Oja, N., Lankinen, H., Mottershead, D.G., Groome, N.P., Ritvos, O. (2004) Immunoneutralization of Growth Differentiation Factor 9 Reveals It Partially Accounts for Mouse Oocyte Mitogenic Activity. *Biol Reprod*; 71 (3): 732-739.
2. Simpson, C.M., Robertson, D.M., Al-Musawi, S.L., Heath, D.A., McNatty, K.P., Ritter, L.J., Mottershead, D.G., Gilchrist, R.B., Harrison, C.A., Stanton, P.G. (2014) Aberrant GDF9 Expression and Activation Are Associated With Common Human Ovarian Disorders. *Journal of Clinical Endocrinology & Metabolism*; 99 (4): E615-E624.
3. Simpson, C.M., Stanton, P.G., Walton, K.L., Chan, K.L., Ritter, L.J., Gilchrist, R.B., Harrison, C.A. (2012) Activation of Latent Human GDF9 by a Single Residue Change (Gly³⁹¹Arg) in the Mature Domain. *Endocrinology*; 153 (3): 1301-1310. **WB, Dilution used 1:5000**
4. Li, J.-J., Sugimura, S., Mueller, T.D., White, M.A., Martin, G.A., Ritter, L.J., Mottershead, D.G. (2015) Modifications of Human Growth Differentiation Factor 9 to Improve the Generation of Embryos From Low Competence Oocytes. *Molecular Endocrinology*, 29(1), 40-52. **WB, Dilution used 1:5000**
5. Mottershead, D.G., Pulkki, M.M., Muggalla, P., Pasternack, A., Tolonen, M., Myllymaa, S., Korchynskyi, O., Nishi, Y., Yanase, T., Lun, S., Juengel, J.L., Laitinen, M., Ritvos, O. (2008) Characterization of recombinant human growth differentiation factor-9 signaling in ovarian granulosa cells. *Molecular and Cellular Endocrinology*, Volume 283, Issues 1-2, Pages 58-67, ISSN 0303-7207. **WB, Dilution used 1:10000**