Human Nanog Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF1997

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Nanog in direct ELISAs and Western blots. In direct ELISAs, less than 1% cross-reactivity with recombinant mouse Nanog is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	*E. coli*-derived recombinant human Nanog Trp153-Val305 Accession # ABZ92376
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

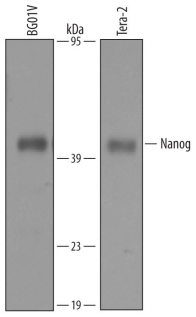
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	1 µg/mL	See Below
Chromatin Immunoprecipitation (ChIP)	5 µg/5 x 10^6 cells	See Below
Immunocytochemistry	5-15 µg/mL	See Below

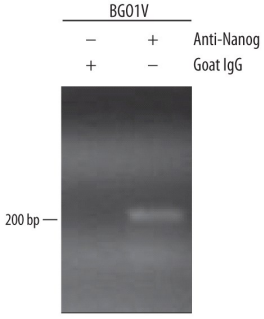
DATA

Western Blot



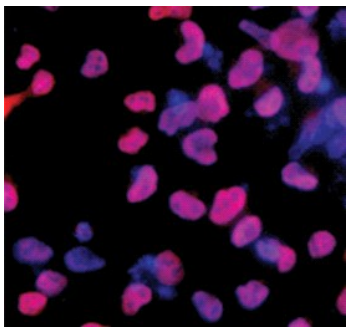
Detection of Human Nanog by Western Blot. Western blot shows lysates of BG01V human embryonic stem cells and Tera-2 human embryonic lung carcinoma cell line. PVDF membrane was probed with 1 µg/mL of Goat Anti-Human Nanog Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1997) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). A specific band was detected for Nanog at approximately 40 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 9.

Chromatin Immunoprecipitation (ChIP)



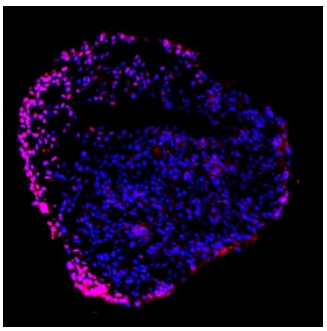
Detection of Nanog-regulated Genes by Chromatin Immunoprecipitation. BG01V human embryonic stem cells were fixed using formaldehyde, resuspended in lysis buffer, and sonicated to shear chromatin. Nanog/DNA complexes were immunoprecipitated using 5 µg Goat Anti-Human Nanog Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1997) or control antibody (Catalog # AB-108-C) for 15 minutes in an ultrasonic bath, followed by Biotinylated Anti-Goat IgG Secondary Antibody (Catalog # BAF109). Immunocomplexes were captured using 50 µL of MagCellect Streptavidin Ferrofluid (Catalog # MAG999) and DNA was purified using chelating resin solution. The *nanog* promoter was detected by standard PCR.

Immunocytochemistry



Nanog in BG01V Human Stem Cells. Nanog was detected in immersion fixed BG01V human embryonic stem cells using 10 µg/mL Goat Anti-Human Nanog Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1997) for 3 hours at room temperature. Cells were stained with the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (red; Catalog # NL001) and counterstained with DAPI (blue). View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

Immunocytochemistry



Nanog in BG01V Human Stem Cells-derived Embryoid Body. Nanog was detected in immersion fixed BG01V human embryonic stem cells-derived embryoid body using 10 µg/mL Goat Anti-Human Nanog Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1997) for 3 hours at room temperature. Cells were stained with the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (red; Catalog # NL001) and counterstained with DAPI (blue). View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

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BACKGROUND

Nanog is a member of the homeobox family of DNA binding transcription factors that has been shown to maintain pluripotency of embryonic stem cells. Its expression is high in undifferentiated embryonic stem cells is down-regulated during embryonic stem cell differentiation, concomitant with loss of pluripotency (1-3).

References:

1. Mitsui, K. *et al.* (2003) Cell **11**:631.
2. Chambers, I. *et al.* (2003) Cell **113**:643.
3. Hart, A.H. *et al.* (2004) Dev. Dyn. **230**:187.