



Product Specification Sheet

NANOG antibody

- Cat # NANOG11-A
- Cat # NANOG11-C

Rabbit anti-Human NANOG antibody
Recombinant NANOG protein control for Western blotting

SIZE: 100 µg
SIZE: 100 µl

NANOG is a divergent homeobox protein and a core component of the transcriptional circuitry that helps embryonic stem cells (ESCs) maintain pluripotency by suppressing cell determination factors. NANOG, as a key regulator of pluripotency and acting synergistically with other factors, has been described as a crucial transcription factor in various types of cancer. NANOG plays a pivotal role in pluripotency acquisition and lineage specification in higher vertebrates, and its expression is restricted to primordial germ cells (PGCs) during early embryonic development. Mammalian NANOG self-associates via conserved tryptophan-repeat motifs in the C-terminal domain (CTD) to maintain pluripotency.

The human NANOG protein coded by the NANOG1 gene, consists of 305 amino acids and possesses 3 functional domains: the N-terminal domain, the C-terminal domain, and the conserved homeodomain motif. The homeodomain region facilitates DNA binding. The human Nanog 1 gene is located on chromosome 12, and the mRNA contains a 915 bp open reading frame (ORF) with 4 exons and 3 introns. The NANOG homeobox gene plays a pivotal role in self-renewal and maintenance of pluripotency in human, mouse and other vertebrate embryonic stem cells, and in pluripotent cells of the blastocyst inner cell mass. NANOG, a marker of stemness, impacts tumor progression and therapeutic resistance in cancer cells.

Source of Antigen or Antibodies

Uniprot: P20263

Host: Rabbit

Clonality: Polyclonal

Purification: Ammonium sulfate followed by peptide affinity purification

Immunogen: 20 amino acid synthetic peptide derived from Human NANOG conjugated to KLH

Species Reactivity: Human

Cross reactivity: The peptide region used as an immunogen exhibits 94% homology with Rhesus Macaque and Crab-eating macaque. 89% Chimpanzee and 89% Gorilla, 83% Orangutan. Due to low homology, it is not recommended for species other than Human or Monkey.

Subcellular Location: Nucleus

Recommended Secondary Antibody: Goat anti-Rabbit IgG-HRP (ADI cat#20320)

Negative Control: Non-immune Rabbit IgG (ADI cat# 20009-1).

Form & Storage of Antibodies

Affinity purified IgG

Concentration: 0.5 mg/ml Volume: 200 µl
Supplied in PBS pH 7.4 + 0.1% BSA
The antibody can be made available carrier free or conjugated to HRP, Biotin, or FITC on request

Lyophilized powder

Reconstitute powder in 200 µl distilled water to 0.5 mg/ml

Storage:

Short-term: 4°C for 1 month

Long-term: at -20°C or below in suitable aliquots after reconstitution for 1 year. Do not expose to multiple freeze/thaw cycles or store working, diluted solutions.

Recommended Usage

Western Blotting: 0.5-2 µg/ml

Predicted band size: ~35 kDa
Observed band size: ~40 kDa

NANOG11-C: Contains a recombinant full length human NANOG protein at a concentration of 1 ng/µl in Laemmli buffer (62.5 mM Tris-HCL, pH 6.8, 2% SDS, 10% glycerol, 5% BME, and 0.002% bromophenol blue). Heat for 5 minutes at 95°C then load 1-5 µl. Store at -20°C in suitable size aliquots, do not expose to multiple freeze/thaw cycles. **Note:** Due to the addition of tags, the protein appears slightly larger than native NANOG.

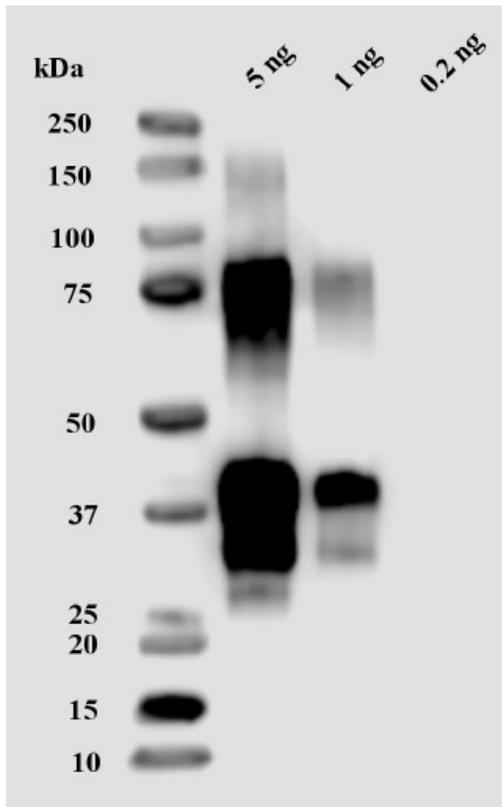
Immunohistochemistry: Not tested for use in-house. We suggest 1-10 µg/ml, permeabilization with Triton X-100 may be necessary.

Above concentrations are a *suggestion*, user's must optimize their assay based on their own conditions. The antibody may work in other applications such as Immunocytochemistry or IP. These methods have not been tested by ADI.

**This product is for In vitro research use only.*

Related materials also available from ADI

Catalog#	Description
SOX2-A antibody	Rabbit anti-Mouse Transcription Factor SOX-2
KLF411-A antibody	Rabbit anti-Mouse Krueppel-like factor 4 (KLF4)
OCT411-A	Rabbit anti-Mouse OCT4 antibody
AB-21110	Mouse Monoclonal Anti-Human c-Myc
NANOG11-A	1917011A



5, 1, and 0.2 ng of full length recombinant Human NANOG (**NANOG11-C**) was heated for 5 minutes at 95°C then separated on a 10% SDS-PAGE gel. The gel was run for ~1 hour and 30 minutes at 100V and transferred to a 0.2 μ m nitrocellulose membrane using the 'Mixed MW' settings on a Transblot Turbo (Biorad). The blot was blocked for 1 hour at room temperature with Fish plasma (Aquablock, EastCoastBio). **NANOG11-A** was diluted with TBST+0.1% BSA to 1 μ g/ml and incubated overnight at 4°C. The blot was washed with TBS-T 3 times for 5 minutes each. Goat anti-rabbit IgG HRP (**ADI cat#20320**) was diluted 1:10,000 (50 ng/ml) in TBST+0.1% BSA and incubated for 1 hour at room temperature. The blot was washed with TBST 3 times for 5 minutes each. The blot was then incubated with ADI Femto ECL substrate (**ADI cat#80210**) for 5 minutes and imaged on a CCD imaging system (LI-COR, C-DiGit).