Product Data Sheet

Anti-human NKX3.1 IgG, 1.0 mg

Catalog Number: 0314

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Product Description

Anti-human NKX3.1 is the IgG fraction from rabbit anti-sera generated by immunizing the animals with purified recombinant human NKX3.1 protein. The immunogen corresponds to the amino-terminal 123 amino acids of the human protein.[1] The IgG fraction was prepared by Protein A affinity chromatography.

NKX3.1 is located on human chromosome 8p21.1. It is an androgen-regulated homeodomain gene with the expression of the NKX3.1 protein predominantly localized to prostate epithelial cells.[2,3]

Instructions for Use

Reconstitute the antibody by resuspending in 1.0 ml of sterile deionized water. Allow to completely dissolve by incubating on ice for 20 min. This will yield a 1 mg/ml solution of anti-NKX3.1 Ig in 1X PBS. Store at 4˚C. Alternatively, dilute into the desired immunoassay buffer at the working concentration and use immediately. Do not store the antibody at the working concentration.

For use in immunohistochemistry, dilute to 1 µg/ml in PBS or 1% BSA.[1] The concentration of antibody needed for optimal results may vary and should be verified. For formalin-fixed paraffin embedded tissues, perform high-temperature antigen retrieval in 1mM EDTA, pH 8.0.

For use in Western blot, dilute to 0.5-1.0 µg/ml in Tris or phosphate buffered saline pH 7.3 with 2% carrier protein.[4] The concentration of antibody needed for optimal results may vary and should be verified.

For other types of immunoassays, it is recommended that the antibody be titered to determine the optimal concentration.

Storage Conditions

Store product desiccated at -20°C.

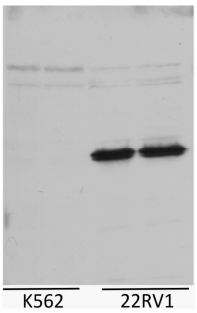
Store reconstituted product at 4°C.

Material Safety Data

FOR RESEARCH USE ONLY. NOT INTENDED OR APPROVED FOR HUMAN, DIAGNOSTICS OR VETERINARY USE. Do not ingest, swallow or inhale. Do not get in eyes, on skin, or on clothing. Wash thoroughly after handling. For complete safety information see full Material Safety Data Sheet.

Example Usage: Immunoblot

To demonstrate the specificity of the anti-human NKX3.1 antibody, 20 µg of total cell extract from human myelogenous leukemia K562 cells and prostate 22RV1 cancer cells were separated by SDS-PAGE and the proteins transferred to PVDF membrane. The membrane was blocked with 2% BSA/TBST for 1 hour and then reacted with anti-human NKX3.1 antibody. The antibody was diluted to a concentration of 1 µg/ml in 0.5% BSA/TBST and the reaction incubated for 1 hour at room temperature. The membrane was washed in TBST three times for 5 minutes each. To detect bound primary antibody, HRP conjugated goat anti-rabbit IgG antibody was diluted 1:10,000 in 0.5% BSA/TBST and incubated with the membrane at room temperature for 1 hour. The membrane was washed 3 times in TBST and developed using chemo-luminescence substrate. The light signals were captured on X-ray film. NKX3.1 protein was detected in prostate cells lysates (22RV1) but not in the leukemia cell lysates (K562).



References

[1] Bethel, C. R. et al. 2006. Decreased NKX3.1 protein expression in focal prostatic atrophy, prostatic intraepithelial neoplasia and adenocarcinoma: association with Gleason score and chromosome 8p deletion. Cancer Res. 66:10683-10690.

[2] Abate-Shen, C., Shen, M. M. and Gelmann, E. 2008. Integrating differentiation and cancer: the Nkx3.1 homeobox gene in prostate organogenesis and carcinogenesis. Differentation; research in biological diversity. 76:717-727.

[3] Gurel, B. et al. 2010. NKX3.1 as a marker of prostatic origin in metastatic tumors. Am. J. Surg. Pathol. 34(8):1097-1105.

4 Guan, B., Pungaliya, P. et al. 2008. Ubiquitination by TOPORS regulates the prostate tumor suppressor NKX3.1. J. Biol. Chem. 283(8):4834-4840.