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Background: Although originally identified based on their roles in calcium and bone homeostasis, the vitamin D receptor (VDR/NR1I1) and its ligand 1α, 25-dihydroxycholecalciferol (1α, 25(OH)2D3) are now recognized to exert biological effects in almost every tissue of the human body. Targets for vitamin D signaling include the central nervous system, skin, immune system, endocrine glands, kidney, and colon. At the cellular level, vitamin D signaling affects proliferation, differentiation, and apoptosis of both normal and transformed cells. Within the steroid receptor gene family, VDR belongs to the NR1 subfamily that also includes NR1I2/PXR and NR1I3/CAR. The human VDR gene is composed of 11 exons that encode six domains (A-F) of the full length VDR protein, which includes an N-terminal dual zinc finger DNA binding domain, a C-terminal ligand-binding activity domain, and an extensive unstructured region that links the two functional domains together (1). Upon 1α, 25(OH)2D3 binding to the hormone ligand-binding domain, VDR is stabilized by the hormone ligand-binding domain, VDR associates with the retinoic acid receptor (RXR) through dimerization domains. The 1α, 25(OH)2D3-VDR-RXR complex binds to the vitamin D response elements (VDREs) in the promoters of target genes through the DNA-binding domain. Ligand-induced conformation changes in VDR result in the dissociation of the co-repressor, silencing-mediator for retinoid and thyroid hormone receptors (SMRT), and allows interaction of the VDR activation function (AF2) transactivation domain with transcriptional coactivators (1).

Studies have shown that variable VDR expression is associated with different forms or stages of cancer and likely results from tissue-type variation in 1α, 25(OH)2D3 signaling. In the case of colon cancer, research indicates that VDR expression is relatively higher in hyperplastic colon polyps and during early tumorigenesis but diminishes in later stage, poorly differentiated tumors. Multiple studies suggest that 1α, 25(OH)2D3 may be an attractive target for development as a therapeutic anticancer agent. (4)

Specificity/Sensitivity: Vitamin D3 Receptor (D2K6W) Rabbit mAb recognizes endogenous levels of total vitamin D3 receptor protein. This antibody does not cross-react with vitamin D3 receptor-like proteins. Based upon sequence alignment, this antibody is predicted to react with both VDRB1 and VDRB2 isoforms.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues near the amino terminus of human vitamin D receptor isoform A protein.

Background References:


Recommended Antibody Dilutions:

Western blotting 1:1000
Immunoprecipitation 1:100
Immunohistochemistry (Paraflin) 1:200†
Unmasking buffer: Citrate
Antibody diluent: SignalStain® Antibody Diluent #8112
Chromatin IP 1:50
Optimal ChIP conditions: 10 µl of antibody & 10 µg of chromatin (4 x 106 cells) per IP. Antibody validated using SimpleChIP® Enzymatic ChIP Kits.

For product specific protocols please see the web page for this product at www.cellsignal.com.

Western blot analysis of extracts from various cell lines using Vitamin D3 Receptor (D2K6W) Rabbit mAb.

IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v nonfat dry milk, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.

For more information, visit www.cellsignal.com.
Western blot analysis of kidney tissue extracts from Vdr (+/+) and Vdr (-/-) mice using Vitamin D3 Receptor (D2K6W) Rabbit mAb (upper) and GAPDH (D16H11) XP® Rabbit mAb #5174 (lower). (Vdr (+/+) and Vdr (-/-) kidneys were kindly provided by Dr. Marie Demay, Massachusetts General Hospital).

Immunohistochemical analysis of paraffin-embedded human breast carcinoma using Vitamin D3 Receptor (D2K6W) Rabbit mAb.

Immunoprecipitation of vitamin D3 receptor from T-47D cell extracts, using Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (lane 2) or Vitamin D3 Receptor (D2K6W) Rabbit mAb (lane 3). Lane 1 is 10% input. Western blot analysis was performed using Vitamin D3 Receptor (D2K6W) Rabbit mAb.

Chromatin immunoprecipitations were performed with cross-linked chromatin from LS180 cells treated with calcitriol (10nM, 3 hours) and either Vitamin D3 Receptor (D2K6W) Rabbit mAb or Normal Rabbit IgG #2729 using SimpleChIP® Enzymatic Chromatin IP Kit (Magnetic Beads) #9003. The enriched DNA was quantified by real-time PCR using SimpleChIP® Human c-Fos Upstream Primers #25661, human UCA1 promoter primers, and SimpleChIP® Human α Satellite Repeat Primers #4486. The amount of immunoprecipitated DNA in each sample is represented as signal relative to the total amount of input chromatin, which is equivalent to one.

Immunohistochemical analysis of paraffin-embedded mouse kidney, Vdr (+/+; left) or Vdr (-/-; right), using Vitamin D3 Receptor (D2K6W) Rabbit mAb (Tissues courtesy of Dr. Marie Demay, Massachusetts General Hospital).