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Product datasheet

Anti-Cryptochrome I/CRY1 antibody ab245564

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Overview

Product name	Anti-Cryptochrome I/CRY1 antibody
Description	Rabbit polyclonal to Cryptochrome I/CRY1
Host species	Rabbit
Tested applications	**Suitable for:** WB, IP
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide within Human Cryptochrome I/CRY1 aa 536-586. The exact sequence is proprietary. NP_004066.1 Database link: Q16526
Positive control	WB: HCT116, Caco-2, HeLa and HEK-293T whole cell lysate. IP: Cryptochrome I/CRY1 IP in HeLa whole cell lysate.
General notes	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 6.8 Preservative: 0.09% Sodium azide Constituents: Tris buffered saline, 0.1% BSA
Purity	Immunogen affinity purified
Purification notes	ab245564 was affinity purified using an epitope specific to Cryptochrome I/CRY1 immobilized on solid support.
Clonality	Polyclonal
Isotype	IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab245564 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		1/2000 - 1/10000. Predicted molecular weight: 66 kDa.
IP		Use at 2-5 µg/mg of lysate.

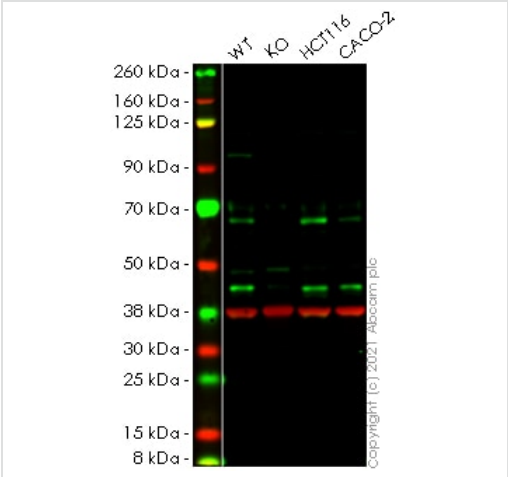
Target

Function

Transcriptional repressor which forms a core component of the circadian clock. The circadian clock, an internal time-keeping system, regulates various physiological processes through the generation of approximately 24 hour circadian rhythms in gene expression, which are translated into rhythms in metabolism and behavior. It is derived from the Latin roots 'circa' (about) and 'diem' (day) and acts as an important regulator of a wide array of physiological functions including metabolism, sleep, body temperature, blood pressure, endocrine, immune, cardiovascular, and renal function. Consists of two major components: the central clock, residing in the suprachiasmatic nucleus (SCN) of the brain, and the peripheral clocks that are present in nearly every tissue and organ system. Both the central and peripheral clocks can be reset by environmental cues, also known as Zeitgebers (German for 'timegivers'). The predominant Zeitgeber for the central clock is light, which is sensed by retina and signals directly to the SCN. The central clock entrains the peripheral clocks through neuronal and hormonal signals, body temperature and feeding-related cues, aligning all clocks with the external light/dark cycle. Circadian rhythms allow an organism to achieve temporal homeostasis with its environment at the molecular level by regulating gene expression to create a peak of protein expression once every 24 hours to control when a particular physiological process is most active with respect to the solar day. Transcription and translation of core clock components (CLOCK, NPAS2, ARNTL/BMAL1, ARNTL2/BMAL2, PER1, PER2, PER3, CRY1 and CRY2) plays a critical role in rhythm generation, whereas delays imposed by post-translational modifications (PTMs) are important for determining the period (tau) of the rhythms (tau refers to the period of a rhythm and is the length, in time, of one complete cycle). A diurnal rhythm is synchronized with the day/night cycle, while the ultradian and infradian rhythms have a period shorter and longer than 24 hours, respectively. Disruptions in the circadian rhythms contribute to the pathology of cardiovascular diseases, cancer, metabolic syndromes and aging. A transcription/translation feedback loop (TTFL) forms the core of the molecular circadian clock mechanism. Transcription factors, CLOCK or NPAS2 and ARNTL/BMAL1 or ARNTL2/BMAL2, form the positive limb of the feedback loop, act in the form of a heterodimer and activate the transcription of core clock genes and clock-controlled genes (involved in key metabolic processes), harboring E-box elements (5'-CACGTG-3') within their promoters. The core clock genes: PER1/2/3 and CRY1/2 which are transcriptional repressors form the negative limb of the feedback loop and interact with the CLOCK NPAS2-ARNTL/BMAL1
ARNTL2/BMAL2 heterodimer inhibiting its activity and thereby negatively regulating their own expression. This heterodimer also activates nuclear receptors NR1D1/2 and RORA/B/G, which form a second feedback loop and which activate and repress ARNTL/BMAL1 transcription, respectively. CRY1 and CRY2 have redundant functions but also differential and selective

	contributions at least in defining the pace of the SCN circadian clock and its circadian transcriptional outputs. More potent transcriptional repressor in cerebellum and liver than CRY2, though more effective in lengthening the period of the SCN oscillator. On its side, CRY2 seems to play a critical role in tuning SCN circadian period by opposing the action of CRY1. With CRY2, is dispensable for circadian rhythm generation but necessary for the development of intercellular networks for rhythm synchrony. Capable of translocating circadian clock core proteins such as PER proteins to the nucleus. Interacts with CLOCK-ARNTL/BMAL1 independently of PER proteins and is found at CLOCK-ARNTL/BMAL1-bound sites, suggesting that CRY may act as a molecular gatekeeper to maintain CLOCK-ARNTL/BMAL1 in a poised and repressed state until the proper time for transcriptional activation. Represses the CLOCK-ARNTL/BMAL1 induced transcription of BHLHE40/DEC1. Represses the CLOCK-ARNTL/BMAL1 induced transcription of ATF4, MTA1, KLF10 and NAMPT (By similarity). May repress circadian target genes expression in collaboration with HDAC1 and HDAC2 through histone deacetylation. Mediates the clock-control activation of ATR and modulates ATR-mediated DNA damage checkpoint. In liver, mediates circadian regulation of cAMP signaling and gluconeogenesis by binding to membrane-coupled G proteins and blocking glucagon-mediated increases in intracellular cAMP concentrations and CREB1 phosphorylation. Besides its role in the maintenance of the circadian clock, is also involved in the regulation of other processes. Represses glucocorticoid receptor NR3C1/GR-induced transcriptional activity by binding to glucocorticoid response elements (GREs). Plays a key role in glucose and lipid metabolism modulation, in part, through the transcriptional regulation of genes involved in these pathways, such as LEP or ACSL4.
Sequence similarities	Belongs to the DNA photolyase class-1 family. Contains 1 photolyase/cryptochrome alpha/beta domain.
Post-translational modifications	Phosphorylation on Ser-247 by MAPK is important for the inhibition of CLOCK-ARNTL/BMAL1-mediated transcriptional activity. Phosphorylation by CSNK1E requires interaction with PER1 or PER2. Phosphorylation at Ser-71 and Ser-280 by AMPK decreases protein stability. Phosphorylation at Ser-568 exhibits a robust circadian rhythm with a peak at CT8, increases protein stability, prevents SCF(FBXL3)-mediated degradation and is antagonized by interaction with PRKDC. Ubiquitinated by the SCF(FBXL3) and SCF(FBXL21) complexes, regulating the balance between degradation and stabilization. The SCF(FBXL3) complex is mainly nuclear and mediates ubiquitination and subsequent degradation of CRY1. In contrast, cytoplasmic SCF(FBXL21) complex-mediated ubiquitination leads to stabilize CRY1 and counteract the activity of the SCF(FBXL3) complex. The SCF(FBXL3) and SCF(FBXL21) complexes probably mediate ubiquitination at different Lys residues. Ubiquitination at Lys-11 and Lys-107 are specifically ubiquitinated by the SCF(FBXL21) complex but not by the SCF(FBXL3) complex. Ubiquitination may be inhibited by PER2.
Cellular localization	Cytoplasm. Nucleus. Translocated to the nucleus through interaction with other clock proteins such as PER2 or ARNTL/BMAL1.

Images



Western blot - Anti-Cryptochrome I/CRY1 antibody (ab245564)

All lanes : Anti-Cryptochrome I/CRY1 antibody (ab245564) at 1/2000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : CRY1 knockout HeLa cell lysate
Lane 3 : HCT116 cell lysate
Lane 4 : Caco-2 cell lysate

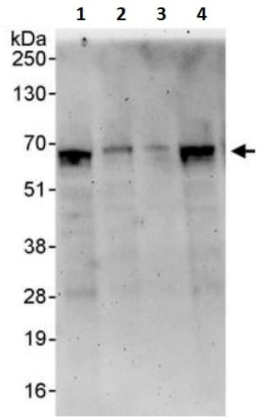
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 66 kDa
Observed band size: 66 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab245564 observed at 66 kDa. Red - loading control ab8245 (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab245564 was shown to react with Cryptochrome I/CRY1 in wild-type HeLa cells in Western blot with loss of signal observed in CRY1 knockout cell line ab265791 (CRY1 knockout cell lysate ab258382). Wild-type HeLa and CRY1 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with ab245564 and ab8245 (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 2000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



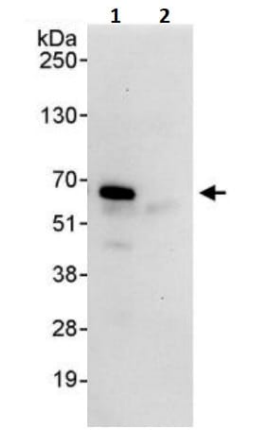
Western blot - Anti-Cryptochrome I/CRY1 antibody (ab245564)

All lanes : Anti-Cryptochrome I/CRY1 antibody (ab245564) at 0.04 µg/ml

Lane 1 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate at 50 µg
Lane 2 : HeLa whole cell lysate at 15 µg
Lane 3 : HeLa whole cell lysate at 5 µg
Lane 4 : HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate at 50 µg

Predicted band size: 66 kDa

Exposure time: 3 minutes



Immunoprecipitation - Anti-Cryptochrome I/CRY1 antibody (ab245564)

Cryptochrome I/CRY1 was immunoprecipitated from HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate (1 mg per IP reaction; 20% of IP loaded).

ab245564 used for IP at 3 µg/mg lysate. For WB 1 µg/ml.

Lane 1: ab245564 IP in HeLa whole cell lysate.
Lane 2: Control IgG in HeLa whole cell lysate.
Chemiluminescence detection: 10 seconds.

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