# abcam

# Product datasheet

# Anti-LRP5 antibody [EPR22477-218] ab223203





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#### Overview

**Product name** Anti-LRP5 antibody [EPR22477-218]

**Description** Rabbit monoclonal [EPR22477-218] to LRP5

**Host species** Rabbit

**Tested applications** Suitable for: WB, IP

Unsuitable for: Flow Cyt,ICC/IF or IHC-P

Species reactivity Reacts with: Mouse, Rat, Human

Recombinant fragment. This information is proprietary to Abcam and/or its suppliers. **Immunogen** 

Positive control WB: MCF7, HCT 116, MEF, 3T3-L1 and HeLa whole cell lysates; rat liver tissue lysate; His-

tagged mouse LRP5 recombinant protein (aa 1407-1614); 293T transfected with LRP5

overexpression vector whole cell lysate. IP: 3T3-L1 whole cell lysate.

**General notes** This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

# **Properties**

**Form** 

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: 7.2

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol, 0.05% BSA

**Purity** Protein A purified

Clonality Monoclonal Clone number EPR22477-218

**Isotype** IgG

## **Applications**

# The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab223203 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		1/500 - 1/10000. Detects a band of approximately 180-200 kDa (predicted molecular weight: 179 kDa).
IP		1/30.

### **Application notes**

Is unsuitable for Flow Cyt,ICC/IF or IHC-P.

## **Target**

#### **Function**

Component of the Wnt-Fzd-LRP5-LRP6 complex that triggers beta-catenin signaling through inducing aggregation of receptor-ligand complexes into ribosome-sized signalsomes. Cell-surface coreceptor of Wnt/beta-catenin signaling, which plays a pivotal role in bone formation. The Wnt-induced Fzd/LRP6 coreceptor complex recruits DVL1 polymers to the plasma membrane which, in turn, recruits the AXIN1/GSK3B-complex to the cell surface promoting the formation of signalsomes and inhibiting AXIN1/GSK3-mediated phosphorylation and destruction of beta-catenin. Appears be required for postnatal control of vascular regression in the eye. Required for posterior patterning of the epiblast during gastrulation.

# Tissue specificity Involvement in disease

Widely expressed, with the highest level of expression in the liver.

Defects in LRP5 are the cause of vitreoretinopathy exudative type 4 (EVR4) [MIM:601813]. EVR4 is a disorder of the retinal vasculature characterized by an abrupt cessation of growth of peripheral capillaries, leading to an avascular peripheral retina. This may lead to compensatory retinal neovascularization, which is thought to be induced by hypoxia from the initial avascular insult. New vessels are prone to leakage and rupture causing exudates and bleeding, followed by scarring, retinal detachment and blindness. Clinical features can be highly variable, even within the same family. Patients with mild forms of the disease are asymptomatic, and their only disease related abnormality is an arc of avascular retina in the extreme temporal periphery. EVR4 inheritance can be autosomal dominant or recessive.

Genetic variations in LRP5 are a cause of susceptibility to osteoporosis (OSTEOP) [MIM:166710]; also known as senile osteoporosis or postmenopausal osteoporosis. Osteoporosis is characterized by reduced bone mass, disruption of bone microarchitecture without alteration in the composition of bone. Osteoporotic bones are more at risk of fracture. Defects in LRP5 are the cause of osteoporosis-pseudoglioma syndrome (OPPG) [MIM:259770]; also known as osteogenesis imperfecta ocular form. OPPG is a recessive disorder characterized by very low bone mass and blindness. Individualy with OPPG are prone to develop bone fractures and deformations and have various eye abnormalities, including phthisis bulbi, retinal detachments, falciform folds or persistent vitreal vasculature.

Defects in LRP5 are a cause of high bone mass trait (HBM) [MIM:601884]. HBM is a rare phenotype characterized by exceptionally dense bones. HBM individuals show otherwise a completely normal skeletal structure and no other unusual clinical findings.

Defects in LRP5 are a cause of endosteal hyperostosis Worth type (WENHY) [MIM:144750]; also

known as autosomal dominant osteosclerosis. WENHY is an autosomal dominant sclerosing bone dysplasia clinically characterized by elongation of the mandible, increased gonial angle, flattened forehead, and the presence of a slowly enlarging osseous prominence of the hard palate (torus palatinus). Serum calcium, phosphorus and alkaline phosphatase levels are normal. Radiologically, it is characterized by early thickening of the endosteum of long bones, the skull and of the mandible. With advancing age, the trabeculae of the metaphysis become thickened. WENHY becomes clinically and radiologically evident by adolescence, does not cause deformity except in the skull and mandible, and is not associated with bone pain or fracture. Affected patients have normal height, proportion, intelligence and longevity.

Defects in LRP5 are the cause of osteopetrosis autosomal dominant type 1 (OPTA1) [MIM:607634]. Osteopetrosis is a rare genetic disease characterized by abnormally dense bone, due to defective resorption of immature bone. The disorder occurs in two forms: a severe autosomal recessive form occurring in utero, infancy, or childhood, and a benign autosomal dominant form occurring in adolescence or adulthood. OPTA1 is characterized by generalized osteosclerosis most pronounced in the cranial vault. Patients are often asymptomatic, but some suffer from pain and hearing loss. It appears to be the only type of osteopetrosis not associated with an increased fracture rate.

Defects in LRP5 are the cause of van Buchem disease type 2 (VBCH2)[MIM:607636]. VBCH2 is an autosomal dominant sclerosing bone dysplasia characterized by cranial osteosclerosis, thickened calvaria and cortices of long bones, enlarged mandible and normal serum alkaline phosphatase levels.

Sequence similarities

Belongs to the LDLR family.

Contains 4 EGF-like domains.

Contains 3 LDL-receptor class A domains. Contains 20 LDL-receptor class B repeats.

Post-translational modifications

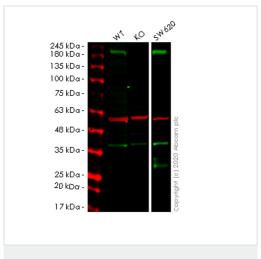
Phosphorylation of cytoplasmic PPPSP motifs regulates the signal transduction of the Wnt

signaling pathway through acting as a docking site for AXIN1.

**Cellular localization** 

Membrane. Endoplasmic reticulum. Chaperoned to the plasma membrane by MESD.

# **Images**



Western blot - Anti-LRP5 antibody [EPR22477-218] (ab223203)

**All lanes :** Anti-LRP5 antibody [EPR22477-218] (ab223203) at 1/500 dilution

Lane 1: Wild-type HEK293T cell lysate

Lane 2: LRP5 knockout HEK293T cell lysate

Lane 3: SW620 cell lysate

Lysates/proteins at 20 µg per lane.

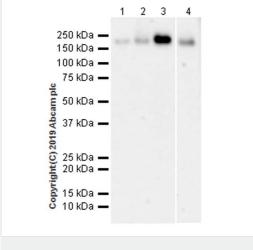
#### Secondary

All lanes: Goat anti-Rabbit lgG H&L (IRDye® 800CW)

preadsorbed (ab216773) at 1/10000 dilution

**Predicted band size:** 179 kDa **Observed band size:** 180-200 kDa **Lanes 1-3:** Merged signal (red and green). Green - ab223203 observed at 180-200 kDa. Red - loading control <u>ab7291</u> observed at 50 kDa.

ab223203 Anti-LRP5 antibody [EPR22477-218] was shown to specifically react with LRP5 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line <a href="mailto:ab266618">ab266618</a> (knockout cell lysate <a href="mailto:ab257202">ab257202</a>) was used. Wild-type and LRP5 knockout samples were subjected to SDS-PAGE. ab223203 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (<a href="mailto:ab7291">ab7291</a>) were incubated overnight at 4°C at 1 in 500 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<a href="mailto:ab216773">ab216773</a>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (<a href="mailto:ab216776">ab216776</a>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-LRP5 antibody [EPR22477-218] (ab223203)

**All lanes :** Anti-LRP5 antibody [EPR22477-218] (ab223203) at 1/1000 dilution

**Lane 1 :** MCF7 (human breast adenocarcinoma epithelial cell), whole cell lysate

**Lane 2 :** HCT116 (human colorectal carcinoma epithelial cell), whole cell lysate

**Lane 3**: HeLa (human cervix adenocarcinoma epithelial cell), whole cell lysate

Lane 4: Rat liver tissue lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 179 kDa

Exposure time: 3 minutes

The expression profile observed is consistent with what has been

described in the literature (PMID: 25808845).

Blocking/Dilution buffer: 5% NFDM/TBST.

All lanes: Anti-LRP5 antibody [EPR22477-218] (ab223203) at 1/1000 dilution

Lane 1: MEF (mouse embryonic fibroblast (immortalized)), whole cell lysate at 20 µg

Lane 2: Untreated 3T3-L1 (mouse embryonic fibroblast), whole cell lysate at 20 µg

Lane 3: 3T3-L1 differentiated adipocytes whole cell lysate 20ug



1/100000 dilution

All lanes: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at

Predicted band size: 179 kDa Observed band size: 180-200 kDa

The expression profile observed is consistent with what has been described in the literature (PMID: 25808845).

Differentiation procedure:

https://www.abcam.com/protocols/differentiation-of-3t3-I1-cells-intoadipocyte-like-cells-protocol

Exposure times: Lane 1: 3 mins; Lane 2-3: 26 secs.

Blocking/Dilution buffer: 5% NFDM/TBST.

All lanes: Anti-LRP5 antibody [EPR22477-218] (ab223203) at 1/1000 dilution

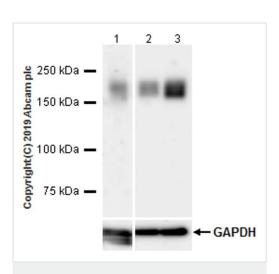
Lane 1: His-tagged mouse LRP5 recombinant protein (aa 1407-

Lane 2: His-tagged mouse LRP6 recombinant protein (aa 1394-1613)

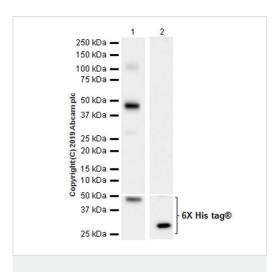
# Secondary

All lanes: Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 179 kDa Observed band size: 25.1.37.5 kDa



Western blot - Anti-LRP5 antibody [EPR22477-218] (ab223203)



Western blot - Anti-LRP5 antibody [EPR22477-218] (ab223203)

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: lane 1: 5.5 secs; lane 2: 59secs

Immunoprecipitation - Anti-LRP5 antibody [EPR22477-218] (ab223203)

LRP5 was immunoprecipitated from 0.35 mg 3T3-L1 (mouse embryonic fibroblast) differentiated adipocytes whole cell lysate 10µg with ab223203 at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab223203. VeriBlot for IP Detection Reagent (HRP) (ab131366) was used at 1/1000 dilution.

Lane 1: 3T3-L1 (mouse embryonic fibroblast) differentiated adipocytes whole cell lysate 10µg.

Lane 2: ab223203 IP in 3T3-L1 differentiated adipocytes whole cell lysate.

Lane 3: Rabbit monoclonal  $\lg G (\underline{ab172730})$  instead of ab223203 in 3T3-L1 differentiated adipocytes whole cell lysate.

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: 3 mins.

Differentiation procedure:

https://www.abcam.com/protocols/differentiation-of-3t3-l1-cells-into-adipocyte-like-cells-protocol.

1 2 3 4 5 6

250 kDa — .
150 kDa — .
100 kDa — .
75 kDa — .
37 kDa — .
25 kDa — .
20 kDa — .
15 kDa — .
16 kDa — .

Western blot - Anti-LRP5 antibody [EPR22477-218] (ab223203)

**Lanes 1-3**: Anti-LRP6 (phospho S1490) + LRP5 (phospho S1503) antibody [EP2360Y] (ab76417) at 1/10000 dilution

**Lanes 4-6**: Anti-LRP5 antibody [EPR22477-218] (ab223203) at 1/10000 dilution

**Lanes 1 & 4 :** 293T transfected with blank vector whole cell lysate **Lanes 2 & 5 :** 293T transfected with LRP6 overexpression vector

whole cell lysate

**Lanes 3 & 6:** 293T transfected with LRP5 overexpression vector whole cell lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 179 kDa

Observed band size: 180 kDa

Exposure time: 180 seconds

Blocking and dilution buffer: 5%NFDM/TBST.



Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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