

Product Specification Sheet

**Phospho OBRb (Tyr985) Antibodies**

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|---|---|---------------------|
| <input type="checkbox"/> <b>Cat #</b> OBR14-P | Mouse OBRb <b>Phospho (Y985)</b> peptide # 1                    | <b>SIZE:</b> 100 ug |
| <input type="checkbox"/> <b>Cat #</b> OBR15-P | Mouse OBRb <b>non-phospho</b> peptide # 2                       | <b>SIZE:</b> 100 ug |
| <input type="checkbox"/> <b>Cat #</b> OBR14-A | Rabbit Anti-Mouse <b>phospho-OBRb (Y985)</b> IgG # 1, aff. pure | <b>SIZE:</b> 100 ug |

Obesity, a common nutritional disorder, is associated with diabetes, hypertension, hyperlipidemia, cancer and many other health related problems. Obese gene, leptin, mediates its effects via the Leptin receptor or obese receptors (**OBR** or **LR**) that is expressed in several tissues including hypothalamus. The OBR has at least 6 alternatively spliced forms (**OBRa-f** or **LRA-f**) that contain a common extracellular domain. OBRb or long isoform contains a unique ~ 300 aa long cytoplasmic tail that is required for leptin signaling. OBR is abnormally spliced (truncation of cytoplasmic domain) in db/db mice resulting into severe obesity.

Leptin is structurally related to cytokine family and it activates cytokine-like signal transduction by stimulating the JAK-STAT pathway via the OBRb, a member of IL-6 receptor family of class I cytokine receptors. Leptin binding to OBRb results in the activation of JAK2 and subsequent phosphorylation of specific tyrosines (**Tyr985** and **Tyr1138**) located at the C-terminal, cytoplasmic region of OBRb. Phosphorylation of OBRb **Tyr1138** recruits the SH2 domain containing STAT3, its autophosphorylation, translocation to the nucleus, and modulation of gene transcription, most notably induction of suppressor of cytokine signaling 3 gene (**SOCS3** or **CIS3**). SOCS3 inhibits leptin signaling. SOCS3 have been reported to be elevated in some obese animals. **Tyr985** phosphorylation of OBRb recruits the SH2 domain containing protein-tyrosine phosphatase **SHP-2**, it's binding to GRB2, and subsequent activation of **ERK**, a group of ser/thr kinases involved in the regulation of gene transcription. Therefore, OBRb Tyr phosphorylation plays in critical role in leptin signaling.

**Source of Antigen and Antibodies**

<b>Antigen</b>	An 11 aa sequence ( <b>designated OBR14-P, phosphorylated peptide</b> ) within the cytoplasmic C-terminus of mouse OBRb (protein accession #P48356, refs 1 980-990 aa; <b>C-QP SVK (Y)AT LVS; Tyr985 phosphorylated</b> ) (1) coupled to KLH. <b>Control non-phosphorylated peptide (cat # OBR15-P)</b> was used to negatively adsorb the phosphopeptide antiserum and making it specific for the OBRb-Tyr985.
<b>Ab Host/type</b>	Rabbit, polyclonal Aff pure IgG ( <b>cat #OBR14-A</b> ) purified over the antigen column
<b>2-ab</b>	Cat # 20320, goat anti-rabbit IgG-HRP (AP, biotin, FITC conjugates also available)
<b>-ve control</b>	# 20009-1, Rabbit (non-immune) IgG, purified, suitable for ELISA, Western, IHC as -ve control

**Form & Storage of Antibodies/Peptide Control**

**Affinity pure IgG**

- 100 ug/100ul     solution     lyophilized powder

Supplied in **Buffer:** PBS+0.1% BSA

**Reconstitute powder** in PBS at 1mg/ml

**Control/blocking peptide**

- 100 ug/100 ul     solution     lyophilized powder

Supplied in Buffer: PBS pH 7.5,

**Reconstitute powder in PBS at 1 mg/ml.**

**Storage**

**Short-term:** unopened, undiluted liquid vials at -20°C and powder at 4°C or -20°C..

**Long-term:** at -20°C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

**Stability:** 6-12 months at -20°C or below.

**Shipping:** 4°C for solutions and room temp for powder

**Recommended Usage**

Working antibody dilution (2-10 ug/ml) should be optimized and adjusted according to the sample composition, and technique employed (**Western blotting, immunoprecipitations, histochemistry**) and sensitivity of detection (colorimetric or Chemiluminescent substrates).

**ELISA** (0.1-2 ug/ml using 10-100 ng of coated peptide/well).

**Specificity and crossreactivity**

Mouse OBR14-P sequence is specific for the long isoform (OBRb). It is 100% conserved in rat, 91% in human, and monkey OBRb. The antibody is specific for OBRb phosphorylated at Tyr985. It does not react with non-phosphorylated OBRb or OBRb phosphorylated at Tyr 1138. Antibody cross-reactivity in various species has not been studied. Phosphorylated OBRb peptide (Cat # OBR14-P) and non-phosphorylated peptide (Cat # OBR15-P) can be used as suitable controls to confirm antibody specificity in various assay. Control peptide, because of its low mol. Wt (<3 kDa), is not suitable for Western. It should be used for ELISA or antibody blocking experiments (use 5-10 ug control peptide per 1 ug of aff pure IgG or 1 ul antiserum) to confirm antibody specificity

**General References:** Bjorbaek C et al (1999) J. Biol. Chem. 274, 30059; Bjorbaek C et al (2000) J. Biol. Chem. 275, 40649; Bank AS et al (2000) J. Biol. Chem. 275, 14563; Bjorbaek C et al (1998) Mol. Cell 1, 619; Tartaglia, LA (1995) Cell 83, 1263; Chen H et al (1996) Cell 84, 491-495; Lee GH et al (1996) Nature 379, 632-635;

This product is for In vitro research use only.

**Related material available from ADI**

Recombinant Leptin, Antibodies to Leptin, Leptin receptor, leptin receptor (Y985) and Y1138, and OB-RGRP antibodies.

OBR14-S-P-P

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