Lep Chemi-Luminescent ELISA Kit (Pig) (OKCD03716)

Instructions for use

For the quantitative measurement of Lep in serum, plasma, tissue homogenates and other biological fluids.

This product is intended for research use only.

Lot to lot kit variations can occur. Refer to the manual which has been provided with the kit.

Contents

1. Background

Principle

Aviva Systems Biology Lep Chemi-Luminescent ELISA Kit (Pig) (OKCD03716) is based on standard sandwich enzyme-linked immuno-sorbent assay technology. An antibody specific for Lep has been pre-coated onto a 96-wellplate (12 x 8 Well Strips). Standards or test samples are added to the wells, incubated and removed. A biotinylated detector antibody specific for Lep is added, incubated and followed by washing. Avidin-Peroxidase Conjugate is then added, incubated and unbound conjugate is washed away. An enzymatic reaction is produced through the addition of a luminol substrate which is catalyzed by the HRP to produce light emission. The light emission is read by a luminometer (or photo-multiplier equipped instrument) and the intensity of the emitted light is proportional to the amount of sample Lep captured in well.

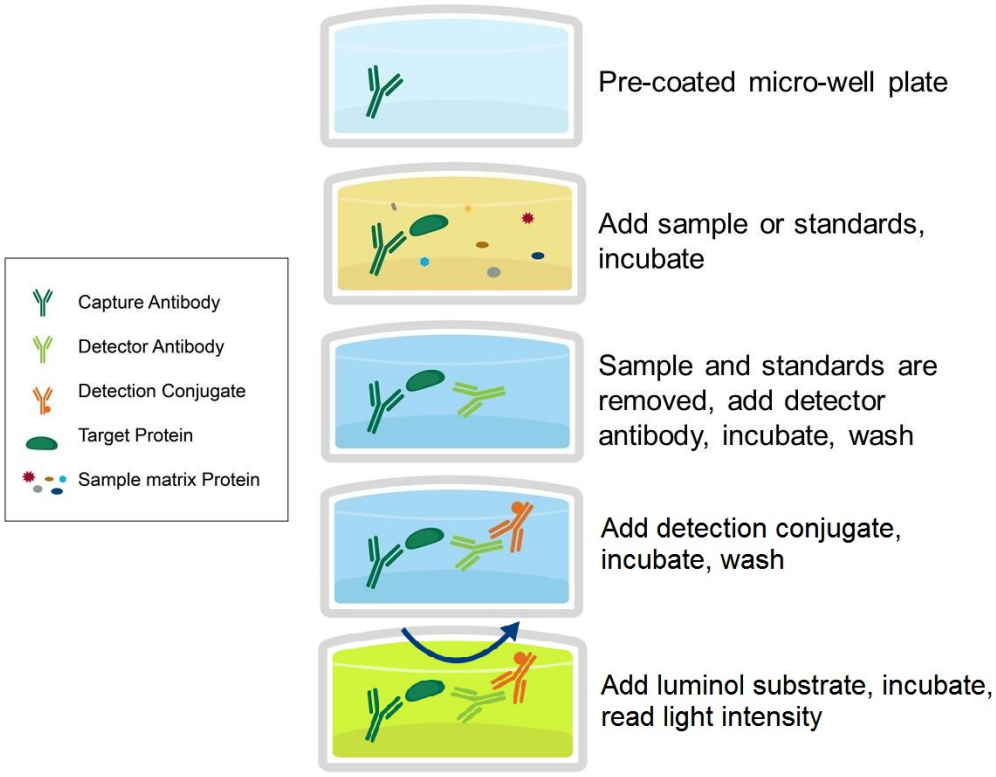
Background

Key player in the regulation of energy balance and body weight control. Once released into the circulation, has central and peripheral effects by binding LEPR, found in many tissues, which results in the activation of several major signaling pathways . In the hypothalamus, acts as an appetite-regulating factor that induces a decrease in food intake and an increase in energy consumption by inducing anorexinogenic factors and suppressing orexigenic neuropeptides, also regulates bone mass and secretion of hypothalamo-pituitary-adrenal hormones. In the periphery, increases basal metabolism, influences reproductive function, regulates pancreatic beta-cell function and insulin secretion, is pro-angiogenic for endothelial cell and affects innate and adaptive immunity . In the arcuate nucleus of the hypothalamus, activates by depolarization POMC neurons inducing FOS and SOCS3 expression to release anorexigenic peptides and inhibits by hyperpolarization NPY neurons inducing SOCS3 with a consequent reduction on release of orexigenic peptides . In addition to its known satiety inducing effect, has a modulatory role in nutrient absorption. In the intestine, reduces glucose absorption by enterocytes by activating PKC and leading to a sequential activation of p38, PI3K and ERK signaling pathways which exerts an inhibitory effect on glucose absorption . Acts as a growth factor on certain tissues, through the activation of different signaling pathways increases expression of genes involved in cell cycle regulation such as CCND1, via JAK2-STAT3 pathway, or VEGFA, via MAPK1/3 and PI3K-AKT1 pathways . May also play an apoptotic role via JAK2-STAT3 pathway and up-regulation of BIRC5 expression. Pro-angiogenic, has mitogenic activity on vascular endothelial cells and plays a role in matrix remodeling by regulating the expression of matrix metalloproteinases (MMPs) and tissue inhibitors of metalloproteinases (TIMPs). In innate immunity, modulates the activity and function of neutrophils by increasing chemotaxis and the secretion of oxygen radicals. Increases phagocytosis by macrophages and enhances secretion of pro-inflammatory mediators. Increases cytotoxic ability of NK cells. Plays a pro-inflammatory role, in synergy with IL1B, by inducing NOS2 wich promotes the production of IL6, IL8 and Prostaglandin E2, through a signaling pathway that involves JAK2, PI3K, MAP2K1/MEK1 and MAPK14/p38 . In adaptive immunity, promotes the switch of memory T-cells towards T helper-1 cell immune responses . Increases CD4+CD25- T-cell proliferation and reduces autophagy during TCR (T-cell receptor) stimulation, through MTOR signaling pathway activation and BCL2 up-regulation .

General Specifications

General Specifications	
Range	0.0137 - 10 ng/m
LOD	< 6.1 pg/mL (Derived by linear regression of OD_{450} of the Mean Blank + 2xSD)
Specificity	Pig Leptin UniProt ID: Q29406 GeneID: 396832 Target Alias: OB, OBS
Cross-Reactivity	No detectable cross-reactivity with other relevant proteins

2. Assay Summary



3. Storage and Stability

• Upon receipt store kit at 4°C for 1 month or -20°C for 6 months, noted exceptions below. Do not use past expiration date.

4. Kit Components

•The following reagents are the provided contents of the kit.

Description	Quantity	Storage Conditions
Anti-Lep Microplate	96 Wells (12 x 8 Well strips)	4°C for 1 Month or -20°C for 6 Months
Lep Lyophilized Standard	2 x 10 ng	
100X Biotinylated Lep Detector Antibody	1 x 120 µL	
100X Avidin-HRP Conjugate	1 x 120 µL	
Standard Diluent	1 x 20 mL	4°C for 6 Month
Detector Antibody Diluent	1 x 12 mL	
Conjugate Diluent	1 x 12 mL	
30X Wash Buffer	1 x 20 mL	
100X Luminol Substrate	1 x 2 mL	
Substrate Diluent	1 x 20 mL	

5. Precautions

• Read instructions fully prior to beginning use of the assay kit.