

Datasheet for ABIN624937

**ADIPOQ ELISA Kit**

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## Overview

Quantity:	96 tests
Target:	ADIPOQ
Reactivity:	Human
Method Type:	Sandwich ELISA
Detection Range:	25-18.000 pg/mL
Minimum Detection Limit:	25 pg/mL
Application:	ELISA

## Product Details

Purpose:	Human Adiponectin (ACRP30) ELISA Kit for cell culture supernatants, plasma, and serum samples.
Sample Type:	Plasma, Cell Culture Supernatant, Serum
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Specificity:	Cross Reactivity: This ELISA kit shows no cross-reactivity with any of the cytokines tested: Human Angiogenin, BDNF, BLC, ENA-78, FGF-4, IL-1 alpha, IL-1 beta, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-11, IL-12 p70, IL-12 p40, IL-13, IL-15, I-309, IP-10, G-CSF, GM-CSF, IFN-gamma, Leptin (OB), MCP-1, MCP-2, MCP-3, MDC, MIP-1 alpha, MIP-1 beta, MIP-1 delta, PARC, PDGF, RANTES, SCF, TARC, TGF-beta, TIMP-1, TIMP-2, TNF-alpha, TNF-beta, TPO, VEGF.
Sensitivity:	< 25 pg/mL

## Product Details

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- Characteristics:
- Strip plates and additional reagents allow for use in multiple experiments
  - Quantitative protein detection
  - Establishes normal range
  - The best products for confirmation of antibody array data

- Components:
- Pre-Coated 96-well Strip Microplate
  - Wash Buffer
  - Stop Solution
  - Assay Diluent(s)
  - Lyophilized Standard
  - Biotinylated Detection Antibody
  - Streptavidin-Conjugated HRP
  - TMB One-Step Substrate

- Material not included:
- Distilled or deionized water
  - Precision pipettes to deliver 2  $\mu$ L to 1  $\mu$ L volumes
  - Adjustable 1-25  $\mu$ L pipettes for reagent preparation
  - 100  $\mu$ L and 1 liter graduated cylinders
  - Tubes to prepare standard and sample dilutions
  - Absorbent paper
  - Microplate reader capable of measuring absorbance at 450nm
  - Log-log graph paper or computer and software for ELISA data analysis

## Target Details

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Target: ADIPOQ

Alternative Name: Adiponectin ([ADIPOQ Products](#))

Background: Human Adiponectin /Acrp30 is expressed and secreted exclusively by adipose tissue. Adiponectin is a hormone that plays a role in the regulation of glucose and lipid homeostasis. Adiponectin as a potent insulin enhancer can link adipose tissue and whole-body glucose metabolism. Levels of Adiponectin are decreased in obesity and it has been observed that Adiponectin levels are also reduced in patients with coronary artery disease. The Human Adiponectin ELISA (Enzyme-Linked Immunosorbent Assay) kit is an in vitro enzyme-linked immunosorbent assay for the quantitative measurement of human Adiponectin in serum, plasma, cell culture supernatants and urine. This assay employs an antibody specific for human Adiponectin coated on a 96-well plate. Standards and samples are pipetted into the wells and Adiponectin present in a sample is bound to the wells by the immobilized antibody. The wells are washed and biotinylated anti-human Adiponectin antibody is added. After washing away unbound biotinylated antibody, HRP-conjugated streptavidin is pipetted to the wells. The wells are again

## Target Details

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washed, a TMB substrate solution is added to the wells and color develops in proportion to the amount of Adiponectin bound. The Stop Solution changes the color from blue to yellow, and the intensity of the color is measured at 450 nm. Reproducibility: Intra-Assay: CV<10% Inter-Assay: CV<12%.

Gene ID: 9370

UniProt: [Q15848](#)

## Application Details

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Application Notes: Recommended Dilution for serum and plasma samples 30,000 fold

Sample Volume: 100 µL

Plate: Pre-coated

Protocol:

1. Prepare all reagents, samples and standards as instructed in the manual.
2. Add 100 µL of standard or sample to each well.
3. Incubate 2.5 h at RT or O/N at 4 °C.
4. Add 100 µL of prepared biotin antibody to each well.
5. Incubate 1 h at RT.
6. Add 100 µL of prepared Streptavidin solution to each well.
7. Incubate 45 min at RT.
8. Add 100 µL of TMB One-Step Substrate Reagent to each well.
9. Incubate 30 min at RT.
10. Add 50 µL of Stop Solution to each well.
11. Read at 450 nm immediately.

Reagent Preparation:

1. Bring all reagents and samples to room temperature (18 - 25 °C) before use.
2. Sample dilution: If your samples need to be diluted, Assay Diluent A (Item D) should be used for dilution of serum/plasma samples. Assay Diluent C should be used for dilution of cell culture supernates/urine. Suggested dilution for normal serum/plasma: 30,000 fold\*. For example, add 2 µL of serum/plasma into a tube with 398.0 µL Assay Diluent A to prepare a 200-fold diluted sample. Mix through and then pipette 2 µL of prepared 200-fold diluted sample into a tube with 298 µL 1x Assay Diluent A to prepare a final 30,000 fold diluted sample. \*Please note that levels of the target protein may vary between different specimens. Optimal dilution factors for each sample must be determined by the investigator.
3. Assay Diluent B should be diluted 5-fold with deionized or distilled water before use.
4. Preparation of standard: Briefly spin the vial of Item C and then add 800 µL Assay Diluent A (for serum/plasma samples) or Assay Diluent C (for cell culture supernates/urine) into Item C vial to prepare a 50 ng/mL standard. Dissolve the powder thoroughly by a gentle mix. Add 180

$\mu\text{L}$  Adiponectin standard (50 ng/mL) from the vial of Item C, into a tube with 320  $\mu\text{L}$  Assay Diluent A or Assay Diluent C to prepare a 18,000 pg/mL stock standard solution. Pipette 400  $\mu\text{L}$  Assay Diluent A or Assay Diluent C into each tube. Use the stock standard solution to produce a dilution series. Mix each tube thoroughly before the next transfer. Assay Diluent A or Assay Diluent C serves as the zero standard (0 pg/mL). The 18,000 pg/mL standard in Assay Diluent A may be saturated, we recommend to start from 6,000 pg/mL for Assay Diluent A Standard curve. 200  $\mu\text{L}$  180  $\mu\text{L}$  standard + 320  $\mu\text{L}$  200myl 200  $\mu\text{L}$  200  $\mu\text{L}$  200  $\mu\text{L}$  18,000 6,000 2,000 666.7 222.2 74.07 24.69 0 pg/mL pg/mL pg/mL pg/mL pg/mL pg/mL pg/mL pg/mL

5. If the Wash Concentrate (20x) (Item B) contains visible crystals, warm to room temperature and mix gently until dissolved. Dilute 20 ml of Wash Buffer Concentrate into deionized or distilled water to yield 400 ml of 1x Wash Buffer.

6. Briefly spin the Detection Antibody vial (Item F) before use. Add 100  $\mu\text{L}$  of 1x Assay Diluent B into the vial to prepare a detection antibody concentrate. Pipette up and down to mix gently (the concentrate can be stored at 4 °C for 5 days). The detection antibody concentrate should be diluted 80-fold with 1x Assay Diluent B and used in step 4 of Part VI Assay Procedure.

7. Briefly spin the HRP-Streptavidin concentrate vial (Item G) ) and pipette up and down to mix gently before use. HRP-Streptavidin concentrate should be diluted 80-fold with 1x Assay Diluent B. For example: Briefly spin the vial (Item G) and pipette up and down to mix gently. Add 100  $\mu\text{L}$  of HRP-Streptavidin concentrate into a tube with 8 ml 1x Assay Diluent B to prepare a 80 fold diluted HRP-Streptavidin solution (don't store the diluted solution for next day use). Mix well.

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### Assay Procedure:

1. Bring all reagents and samples to room temperature (18 - 25 °C) before use. It is recommended that all standards and samples be run at least in duplicate.
2. Add 100  $\mu\text{L}$  of each standard (see Reagent Preparation step 2) and sample into appropriate wells. Cover well and incubate for 2.5 hours at room temperature or over night at 4 °C with gentle shaking.
3. Discard the solution and wash 4 times with 1x Wash Solution. Wash by filling each well with Wash Buffer (300 myl) using a multi-channel Pipette or autowasher. Complete removal of liquid at each step is essential to good performance. After the last wash, remove any remaining Wash Buffer by aspirating or decanting. Invert the plate and blot it against clean paper towels.
4. Add 100  $\mu\text{L}$  of 1x prepared biotinylated antibody (Reagent Preparation step 6) to each well. Incubate for 1 hour at room temperature with gentle shaking.
5. Discard the solution. Repeat the wash as in step
6. Add 100  $\mu\text{L}$  of prepared Streptavidin solution (see Reagent Preparation step 7) to each well. Incubate for 45 minutes at room temperature with gentle shaking.
7. Discard the solution. Repeat the wash as in step

## Application Details

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8. Add 100  $\mu$ L of TMB One-Step Substrate Reagent (Item H) to each well. Incubate for 30 minutes at room temperature in the dark with gentle shaking.
9. Add 50  $\mu$ L of Stop Solution (Item I) to each well. Read at 450 nm immediately.

### Calculation of Results:

Calculate the mean absorbance for each set of duplicate standards, controls and samples, and subtract the average zero standard optical density. Plot the standard curve on log-log graph paper or using Sigma plot software, with standard concentration on the x-axis and absorbance on the y-axis. Draw the best-fit straight line through the standard points.

Typical Data: These standard curves are for demonstration only. A standard curve must be run with each assay. Assay Diluent A Adiponectin concentration (pg/mL) 1 10 100 1000 10000 O D =4 50 n m 0.01 0.1 1 10 Assay Diluent C Adiponectin concentration (pg/mL) O D =4 50 n m 0.01 0.1 1 10 10 100 1,000 10,000 100,000

Sensitivity: The minimum detectable dose of Adiponectin is typically less than 25 pg/mL.

Recovery: Recovery was determined by spiking various levels of human Adiponectin into human serum, plasma and cell culture media. Mean recoveries are as follows: Sample Type Average % Recovery Range (%) Serum 93.57 83-104 Plasma 90.29 80-103 Cell culture media 96.59 83-107

Linearity: Sample Type Serum Plasma Cell Culture Media 1:2 Average % of Expected 95 94 92 Range (%) 83-104 84-103 82-103 1:4 Average % of Expected 93 92 93 Range (%) 84-104 83-102 85-104

Reproducibility: Intra-Assay: CV<10 % Inter-Assay: CV<12 %

### Assay Precision:

Intra-Assay: CV< 10 % Inter-Assay: CV< 12 %

### Restrictions:

For Research Use only

## Handling

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### Handling Advice:

Avoid repeated freeze-thaw cycles.

### Storage:

-20 °C

### Storage Comment:

The entire kit may be stored at -20°C for up to 1 year from the date of shipment. Avoid repeated freeze-thaw cycles. The kit may be stored at 4°C for up to 6 months. For extended storage, it is recommended to store at -80°C.

### Expiry Date:

6 months

## Publications

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### Product cited in:

Kapil, Duseja, Sharma, Singla, Chakraborti, Das, Ray, Dhiman, Chawla: "Small intestinal bacterial

overgrowth and toll-like receptor signaling in patients with non-alcoholic fatty liver disease." in:

**Journal of gastroenterology and hepatology**, Vol. 31, Issue 1, pp. 213-21, (2016) ([PubMed](#)).

Wu, Ding, Han, Arriens, Wei, Han, Pedroza, Jiang, Anolik, Petri, Sanz, Saxena, Mohan: "Antibody-Array-Based Proteomic Screening of Serum Markers in Systemic Lupus Erythematosus: A Discovery Study." in: **Journal of proteome research**, Vol. 15, Issue 7, pp. 2102-14, (2016) ([PubMed](#)).

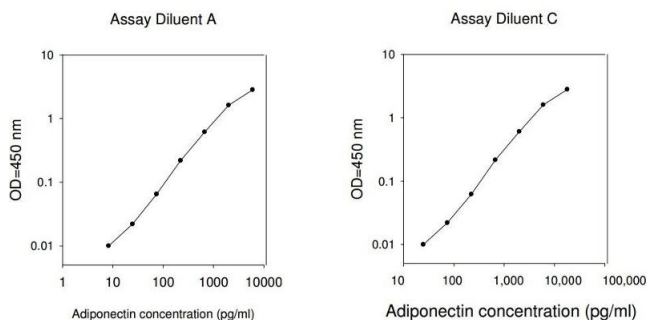
Nobili, Alisi, Cutrera, Carpino, De Stefanis, D'Orta, De Vito, Cucchiara, Gaudio, Musso: "Altered gut-liver axis and hepatic adiponectin expression in OSAS: novel mediators of liver injury in paediatric non-alcoholic fatty liver." in: **Thorax**, Vol. 70, Issue 8, pp. 769-81, (2015) ([PubMed](#)).

Oral, Gulec, Kurt, Yilmaz, Aydin, Kirpinar: "The effects of atypical antipsychotic usage duration on serum adiponectin levels and other metabolic parameters." in: **The Eurasian journal of medicine**, Vol. 43, Issue 1, pp. 39-44, (2015) ([PubMed](#)).

Cho, Choi, Park, Yi, Hossen, Kim, Ro, Cha, Yoo, Kim, Lee: "Alcohol-induced Hyperlipidemia Is Ameliorated by Orally Administered DWP208, a Sodium Succinate Form of ZYM201." in: **The Korean journal of physiology & pharmacology : official journal of the Korean Physiological Society and the Korean Society of Pharmacology**, Vol. 18, Issue 6, pp. 469-74, (2015) ([PubMed](#)).

There are more publications referencing this product on: [Product page](#)

Images



**ELISA**  
**Image 1.**