The Adiponectin Turbidimetric Immunoassay Reagent Kit

Catalogue number: 51010

For the quantitative determination of Adiponectin
in human serum and plasma

This package insert must be read in its entirety before using this product
Use only the current version of product data sheet enclosed with the kit

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FOR RESEARCH USE ONLY
NOT FOR USE IN DIAGNOSTIC PROCEDURES

Version:3.8

TABLE OF CONTENT

Content	Page
INTRODUCTION	1
PRINCIPLE OF THE ASSAY	1
REAGENTS SUPPLIED	2
OTHER MATERIALS REQUIRED, BUT NOT PROVIDED	2
STORAGE	2
SAMPLE HANDLING	2
ASSAY PROCEDURE	3
TYPICAL STANDARD CURVE	3
CALCULATION	4
ASSAY CHARACTERISTICS	4

INTRODUCTION

Adiponectin (ADPN), also known as apM1, Acrp30, GBP28 and adipoQ, is a circulating hormone predominantly produced from adipose tissue. Many pharmacological studies demonstrated that this protein possesses potent anti-diabetic, anti-atherogenic and anti-inflammatory functions. Supplement of adiponectin protein can decrease blood glucose, improve insulin sensitivity, alleviate fatty liver and prevent atherosclerosis. Decreased circulating levels of plasma adiponectin (hypoadiponectinaemia) are associated with increased body mass index (BMI), and decreased insulin sensitivity.

Recently, many clinical studies demonstrated that circulating adiponectin levels are decreased significantly in type 2 diabetes, coronary heart diseases and stroke. Decreased circulating adiponectin levels indicate the increased risk for the development of aforementioned diseases.

PRINCIPLE OF THE ASSAY

This assay is a turbidimetric immunoassay for the quantitative measurement of ADPN in human serum and urine. A standard or sample is added into a cuvette and mixed with the reaction buffer R1. After a short incubation, the test reagent R2, which is a suspension of microparticles coated with ADPN antibodies, is added into the cuvette and mixed. The presence of ADPN in the standard or sample causes the immune-particles to aggregate. The extent to which the microparticles aggregate is quantified by the amount of light scattering measured as absorbance by a chemistry analyzer. The concentration of ADPN in unknown samples can be interpolated from a reference curve using the standards provided.

REAGENTS SUPPLIED

R1 – Reaction buffer, 30 ml, a ready-to-use buffer solution containing salt, polyether compound and preservative

R2 – Test reagent, 10 ml, a ready-to-use suspension of polymer microparticles coated with rabbit anti-ADPN polyclonal antibodies in storage buffer

ADPN Control –High and Low ADPN control, 1ml per tube

OTHER MATERIALS REQUIRED, BUT NOT PROVIDED

1. Clinical chemistry analyzer
2. Deionized water
3. Analyzer-specific reagent containers for R1 and R2

STORAGE

The kit should be stored at 2-8°C upon receipt. Once opened, the reagents may be stored at 2-8°C for up to 4 weeks.

SAMLE HANDLING

This kit can be used to determine ADPN in human serum and plasma samples. Blood specimens should be collected aseptically into appropriate tubes. Plasma should be prepared by standard techniques for laboratory testing. The prepared specimens should be stored in closed vessels. If the assay cannot be performed within 24 hours or specimens are to be shipped, the specimens should be frozen at −20°C or below. For long-term storage of specimens, −70°C or below is recommended. To avoid freeze-thaw cycles, specimens should be aliquoted. Do not use hemolyzed, hyperlipemic, heat-treated or contaminated specimens. No dilution of the sample is required in this assay.

ASSAY PROCEDURE

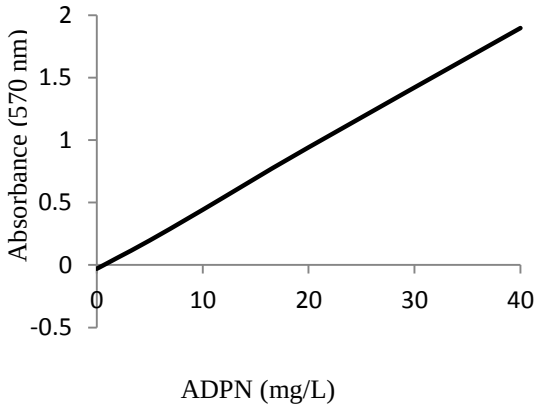
Assay procedures may vary depending on the automated chemistry analyzer to be used. A general example of assay procedures is stated as follow:

1. Dispense 150µl of R1 into a clean cuvette
2. Add 1.5µl of sample and incubate at 37˚C for 5 minutes
3. Further add 50µl of R2
4. Read change of absorbance at 570 nm for 8 minutes after the addition of R2
5. Calculate the concentration of ADPN in unknown sample by interpolation from a reference curve using the standards provided

TYPICAL STANDARD CURVE

The following standard curve is provided for demonstration only. A standard curve should be generated for each assay.

ADPN (mg/L)	Absorbance (570 nm)
0	-0.030
2	0.060
5	0.197
10	0.442
20	0.942
40	1.897



CALCULATION

1. Subtract the absorbance of the blank from that of standards and samples.
2. Generate a standard curve by plotting the absorbance obtained (y-axis) against ADPN concentrations (x-axis). The best fit line can be generated with any curve-fitting software by regression analysis. 4-parameter curve fitting can be used for calculation.
3. Determine ADPN concentration of samples from standard curve.

ASSAY CHARACTERISTICS

A. Sensitivity

The sensitivity is defined as the lower limit of detection and is estimated as the mean of the blank sample plus three times the SD obtained from the blank sample. The sensitivity of ADPN assay is 0.3mg/L.

B. Precision

The precision of the ADPN assay is < 10% CV. Four samples consisting of two ADPN controls and two serum based panels were assayed 20 times separately.

Sample	Mean ADPN (ng/ml)	SD (ng/ml)	CV
Low control	2.7	0.1	3.96%
High control	10.3	0.2	1.70%
Panel 1	19.7	0.2	1.10%
Panel 2	27.0	0.3	1.16%

C. Linearity

The ADPN assay is linear between 1 mg/L to 40 mg/L.

D. Interference

No interference was detected with hemoglobin up to 5 g/L, conjugated bilirubin up to 300 mg/L, free bilirubin up to 300 mg/L, and up to 5g/L lipid emulsion.