

The Adiponectin Turbidimetric Immunoassay 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: 6.1



ImmunoDiagnostics Limited

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PACKING SPECIFICATION

Cat. No.	Size	Approximately tests
51010-05	R1: 15ml, R2: 5ml	100
51010-10	R1: 30ml, R2: 10ml	200
51010-20	R1: 60ml, R2: 20ml	400
51010-50	R1: 150ml, R2: 50ml	1000
51010-100	R1: 300ml, R2: 100ml	2000

INTRODUCTION

Adiponectin, also known as apM1, Acrp30, adipoQ and GBP28, is a circulating hormone predominantly produced from adipose tissue. It is a glycoprotein with molecular weight of approximately 30 kDa and has circulating levels of 2-30 µg/mL in humans. Structurally it resembles complementary factor C1q. During assembly, adiponectin forms complexes of different molecular weight and function. Adiponectin has anti-diabetic, insulin-sensitizing and anti-inflammatory properties. Decreased circulating levels of plasma adiponectin (hypoadiponectinemia) are associated with increased body mass index (BMI), and decreased insulin sensitivity. A large number of longitudinal studies in different ethnic groups uniformly demonstrated that circulating adiponectin levels are decreased significantly in type 2 diabetes (T2D) and related complications, and low circulating adiponectin levels indicate the increased risk for the development of T2D.

PRINCIPLE OF THE ASSAY

This assay is a turbidimetric immunoassay for the quantitative measurement of adiponectin in human serum and plasma. 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 an antibody highly specific to adiponectin, is added into the cuvette and mixed. The presence of adiponectin 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 adiponectin in unknown samples can be interpolated from a reference curve using the standards provided.

REAGENTS SUPPLIED

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

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

OTHER MATERIALS REQUIRED

1. Clinical chemistry analyzer
2. Adiponectin Calibrator (provided separately, Cat. #51010-S1)
3. Adiponectin Control (optional, provided separately, Cat. #51010-C1)
4. Deionized water
5. 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 adiponectin 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

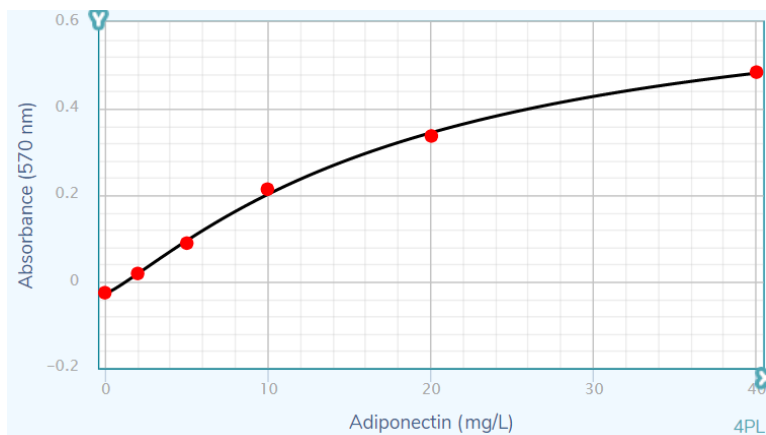
Assay procedures may vary depending on the automated chemistry analyzer to be used. A general example of assay procedures is stated as follow:

1. Dispense 150 μ l of R1 into a clean cuvette
2. Add 1.5 μ l of serum or plasma sample or adiponectin calibrator and incubate at 37°C for 5 minutes
3. Further add 50 μ l of R2
4. Read change of absorbance at 570 nm for 8 minutes after the addition of R2
5. Calculate the concentration of Adiponectin 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.

Adiponectin (mg/L)	Absorbance (570 nm)
0	-0.0253
2	0.0191
5	0.0892
10	0.2139
20	0.3369
40	0.4844



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 adiponectin 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 adiponectin 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 Adiponectin assay is 0.3mg/L.

B. Precision

The precision of the adiponectin assay is < 5% CV. Four samples consisting of two adiponectin controls and two serum samples were assayed 20 times separately.

Sample	Mean Adiponectin (mg/L)	SD (mg/L)	CV
Low control	2.7	0.1	3.96%
High control	10.3	0.2	1.70%
Panel 1	4.7	0.2	1.10%
Panel 2	8.0	0.3	1.16%

C. Linearity

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

D. Interference

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



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