



#### As of 19 March 2007 (Vers. 1.0)



#### 1 INTENDED USE

This Adiponectin Human High Molecular Weight (HMW) ELISA kit is used for the nonradioactive quantification of Human HMW Adiponectin in serum, plasma, and adipocyte extracts or culture media samples with a simple sample pretreatment.

Sample treatment specifically removes Hexameric and Trimeric Adiponectin in samples and allows for specific measurement of HMW Adiponectin only. The antibody pair does not recognize other human adipokines tested. This Human HMW Adiponectin kit is sufficient to measure 37 unknown samples in duplicate.

This kit is for research purpose only.

#### **2** PRINCIPLES OF PROCEDURE

This assay is a Sandwich ELISA based, sequentially, on: 1) capture of Adiponectin molecules from samples to the wells of a microtiter plate coated with a monoclonal antiadiponectin antibodies, 2) washing of unbound materials from samples, 3) binding of a second biotinylated polyclonal anti-adiponectin antibody to the captured molecules , 4) washing of unbound materials from samples, 5) binding of streptavidin-horseradish peroxidase conjugate to the immobilized biotinylated antibodies, 6) washing of excess of free enzyme conjugates, and 7) quantification of immobilized antibody-enzyme conjugates by monitoring horseradish peroxidase activities in the presence of the substrate 3,3',5,5'-tetramethylbenzidine. The enzyme activity is measured spectrophotometrically by the increased absorbance at 450 nm – 590 nm after acidification of formed products.

Since the increase in absorbance is directly proportional to the amount of captured Human HMW Adiponectin in the unknown sample, the latter can be derived by interpolation from a reference curve generated in the same assay with reference standards of known concentrations of Human HMW Adiponectin.

#### **3 REAGENTS SUPPLIED**

Each kit is sufficient to run one 96-well plate and contains the following reagents:

#### A. Human HMW Adiponectin ELISA Plate

Coated with Monoclonal Anti-Adiponectin Antibodies Quantity: 1 Strip plate Preparation: Ready to Use Note: Unused strips should be resealed in the foil pouch with the desiccant provided and stored at 2-8°C.

### **B.** Adhesive Plate Sealer

Quantity: 2 sheets Preparation: Ready to Use

## C. 10X HRP Wash Buffer Concentrate

10X concentrate of 50 mM Tris Buffered Saline containing Tween-20 Quantity: 2 bottles containing 50 ml each Preparation: Dilute 1:10 with distilled or deionized water





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### D. Human HMW Adiponectin Standard Adiponectin Calibrator, lyophilized. Quantity: 200 ng/ml upon hydration. Preparation: Reconstitute with 0.5 ml distilled or deionized water to obtain 200 ng/ml. E. Human HMW Adiponectin Quality Controls 1 and 2 One vial each, lyophilized, containing diluted serum at two different levels of Adiponectin. Quantity: 0.5 ml/bottle upon hydration Preparation: Reconstitute each vial with 0.5 ml distilled or deionized water.

- **F.** Sample Digestion Solution Quantity: 600 μl Preparation: Ready to use
- **G.** Sample Digestion Buffer For the Digesting of Samples Only) Quantity: 8 ml Preparation: ready to use
- H. Sample Dilution Buffer (10X) For Sample Dilution Only) Quantity: 1 ml Preparation: Dilute 1:10 with 1X Assay Buffer
- I. 10X Assay Buffer For Diluting Sample Dilution Buffer Only) Quantity: 50ml (10X)
   Preparation: Dilute 1:10 with distilled or deionized water
- J. Assay Running Buffer For Use in Running Assay and Diluting Standards Only) 0.05M Phosphosaline containing 0.025M EDTA, 0.08% Sodium Azide, 1% BSA Quantity: 13 ml Preparation: Ready to Use
- **K.** Sample Preparation Plates ( Quantity: Two 96-well solid plates with 2 plate sealers Preparation: Ready to Use
- L. Human HMW Adiponectin Detection Antibody Pre-titered Biotinylated Polyclonal anti-Adiponectin Antibody Quantity: 12 ml Preparation: Ready to Use

### M. Enzyme Solution

Pre-titered Streptavidin-Horseradish Peroxidase Conjugate in Buffer Quantity: 12 ml Preparation: Ready to Use

N. Substrate (Light sensitive, avoid unnecessary exposure to light)

 3, 3', 5, 5'-tetramethylbenzidine in buffer
 Quantity: 12 ml
 Preparation: Ready to Use.







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#### **O.** Stop Solution (Caution: Corrosive Solution)

0.3 M HCl Quantity: 12 ml Preparation: Ready to Use

#### 4 STORAGE AND STABILITY

Prior to use, all components in the kit can be stored up to 2 weeks at 2-8°C except for the Sample Digestion Solution, Sample Dilution Buffer, and Sample Digestion Buffer which must be stored at  $\leq$  -20°C.

For longer storage (> 2 weeks), freeze diluted Wash Buffer, Assay Buffer, and reconstituted Standards and Controls at  $\leq -20^{\circ}$ C.

Minimize repeated freeze and thaw of the HMW Adiponectin Standards and Quality Controls.

Unused microtiter strips should be resealed in the foil pouch with the desiccant provided and stored at 2-8°C. Refer to expiration dates on all reagents prior to use.

Do not mix reagents from different kits unless they have the same lot numbers.

#### **5 REAGENT PRECAUTIONS**

#### A. Sodium Azide

Sodium Azide has been added to certain reagents as a preservative. Although the concentrations are low, Sodium Azide may react with lead and copper plumbing to form highly explosive metal azides. On disposal, flush with a large volume of water to prevent azide build up.

#### **B.** Hydrochloric Acid

Hydrochloric Acid is corrosive and can cause eye and skin burns. It is harmful if swallowed and can cause respiratory and digestive tract burns. Avoid contact with skin and eyes. Do not swallow or ingest.

#### 6 MATERIALS REQUIRED BUT NOT PROVIDED

- 1. Pipettes and Pipette Tips: 10 µl 20 µl or 20 µl 100 µl
- 2. Multi-Channel Pipettes and Pipette Tips:  $5 \sim 50 \ \mu l$  and  $50 \sim 300 \ \mu l$
- 3. Buffer and Reagent Reservoirs
- 4. Vortex Mixer
- 5. Deionized Water
- 6. Microtiter Plate Reader capable of reading absorbency at 450 nm
- 7. Orbital Microtiter Plate Shaker
- 8. Absorbent Paper or Cloth
- 9. 37°C Incubator





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#### 7 SAMPLE COLLECTION AND STORAGE

- 1. To prepare serum samples, whole blood is directly drawn into a centrifuge tube that contains no anti-coagulant. Let blood clot at room temperature for 30 min.
- 2. Promptly centrifuge the clotted blood at 2,000 to 3,000 x g for 15 minutes at  $4 \pm 2$  °C.
- 3. Transfer and store serum samples in separate tubes. Date and identify each sample.
- 4. Use freshly prepared serum or aliquot and store samples at  $\leq -20^{\circ}$ C for later use. For long-term storage, keep at -70°C. Avoid freeze/thaw cycles.
- 5. To prepare plasma samples, whole blood should be collected into centrifuge tubes containing enough K<sub>3</sub>EDTA to achieve a final concentration of 1.735 mg/ml and centrifuged immediately after collection. Observe the same precautions in the preparation of serum samples.
- 6. If heparin is to be used as an anticoagulant, the effect on the assay outcome at the dose of heparin used should be predetermined.
- 7. Avoid using samples with gross hemolysis or lipemia.

#### 8 SAMPLE PREPARATION

#### **IMPORTANT NOTE:**

SAMPLE PREPARATION SHOULD BE PERFORMED JUST PRIOR TO SET UP OF THE ASSAY.

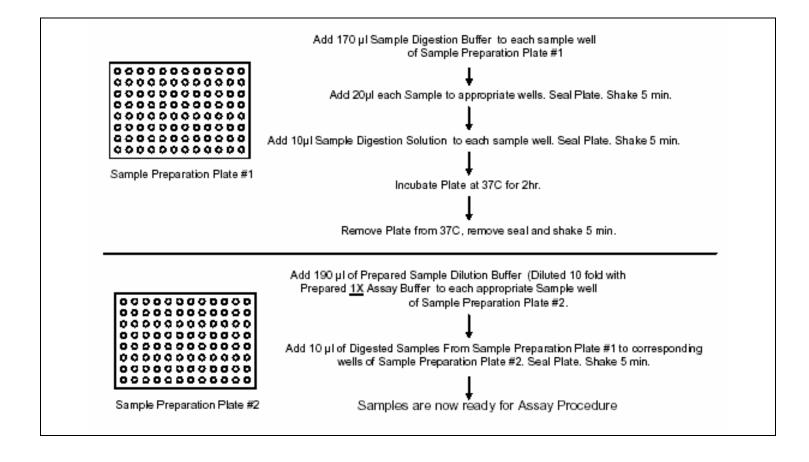
- Add 170 μl of Sample Digestion Buffer (Not to be confused with Sample Digestion Solution) to the appropriate sample well of one of the solid Sample Preparation Plates for each sample to be digested.
   NOTE: Human HMW Assay Procedure only allows space for 37 samples in duplicate.
- 2. Add 20 µl of each sample to the appropriate wells.
- 3. Seal plate and shake for 5 minutes on an orbital microtiter plate shaker set to rotate at moderate speed, approximately 500 rpm.
- 4. Remove plate from shaker and add 10 μl of Sample Digestion (Mix Sample Digestion Solution well before adding to wells) to each sample well (Samples are now 1:10).
- 5. Seal plate and shake for 5 minutes on an orbital microtiter plate shaker set to rotate at moderate speed, approximately 500 rpm.
- 6. Place plate carefully in 37°C incubator for 2 hours (Prepare Sample Dilution Buffer while incubating Step 7).
- Prepare 1X Sample Dilution Buffer: Dilute the 10X Assay Buffer concentrate 10 fold by mixing the entire content of the bottle of 10X Assay Buffer with 450 ml deionized water. Use this 1X Assay Buffer to dilute the 10X Sample Dilution Buffer 10 fold by adding 900 µl of Sample Dilution to 8.1 ml of 1X Assay Buffer. Mix well.





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- After the Step 6 2 hour incubation, remove plate from 37°C incubator. Carefully remove plate sealer and shake plate for 5 minutes on an orbital microtiter plate shaker set to rotate at moderate speed, approximately 500 rpm.
- 9. Add 190 μl of Prepared 1X Sample Dilution Buffer (prepared in Step 7) to the appropriate wells of the 2<sup>nd</sup> unused- solid Sample Preparation Plate.
- Add 10 μl of each digested sample to corresponding wells of the 2<sup>nd</sup> Sample preparation plate containing the 1X Sample Dilution Buffer (Samples are now 1:200).
   Seal and shake plate for 5 minutes on an orbital microtiter plate shaker set to rotate at moderate speed, approximately 500 rpm.
- 11. Proceed with Assay. Samples should be used immediately.
  - NOTE: 1:10 Digested samples may be sealed and kept at -20°C for future use. 1:200 Diluted Samples should be discarded after immediate use.



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#### 9 STANDARD AND QUALITY CONTROLS PREPARATION

#### **A. Standard Preparation**

- 1. Use care in opening the lyophilized Standard vial. Using an Eppendorf pipette, reconstitute the Human HMW Adiponectin Standard with 0.5 ml distilled or deionized water into the glass vial to give a concentration prescribed in the analysis sheet. Invert and mix gently, let sit for 5 minutes then vortex gently.
- Label seven tubes 1, 2, 3, 4, 5, 6 and 7. Add 0.25 ml Assay Running Buffer to each of the seven tubes. Prepare serial dilutions by adding 0.25 ml of the reconstituted standard to tube 1, mix well and transfer 0.25 ml of tube 1 to tube 2, mix well and transfer 0.25 ml of tube 2 to tube 3, mix well and transfer 0.25 ml of tube 3 to tube 4, mix well and transfer 0.25 ml of tube 4 to tube 5, mix well and transfer 0.25 ml of tube 5 to tube 6, mix well and transfer 0.25 ml of tube 6 to tube 7 and mix well.
   Note: Do not use a Repeater pipette. Change tip for every dilution. Wet tip with standard before dispensing. Unused portions of standard should be stored at ≤ -20°C. Avoid multiple freeze/thaw cycles.

	Volume of Deionized Water to add	Volume of Standard to add	Standard Concentration (ng/ml)
	0.5 mL	0	X (refer to analysis sheet for exact concentration)
Tube #	Volume of Assay Running Buffer to add	Volume of Standard to add	Standard Concentration (ng/ml)
1	0.25 mL	0.25 mL of reconstituted standard	X/2
2	0.25 mL	0.25 mL of tube 1	X/4
3	0.25 mL	0.25 mL of tube 2	X/8
4	0.25 mL	0.25 mL of tube 3	X/16
5	0.25 mL	0.25 mL of tube 4	X/32
6	0.25 mL	0.25 mL of tube 5	X/64
7	0.25 mL	0.25 mL of tube 6	X/128

#### **B.** Quality Control 1 and 2 Preparation

Use care in opening the lyophilized Quality Control vials.

Using an Eppendorf pipette, reconstitute each of the Human HMW Adiponectin Quality Control 1 and Quality Control 2 with 0.5 ml distilled or deionized water into the glass vials.

Invert and mix gently, let sit for 5 minutes then mix well.





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#### **10 ASSAY PROCEDURE**

#### Pre-warm all reagents to room temperature prior to setting up the assay.

- 1. Dilute the 10X Wash Buffer concentrate 10 fold by mixing the entire content of each bottle of Wash Buffer with 450 ml deionized water (Dilute both bottles with 900 ml deionized water).
- 2. Remove the required number of strips from the Microtiter Assay Plate. Unused strips should be resealed in the foil pouch and stored at 2-8°C. Assemble the strips in an empty plate holder and wash each well 3 times with 300 µl of diluted Wash Buffer per wash. Decant wash buffer and remove the residual amount from all wells by inverting the plate and tapping it smartly onto absorbent towels several times.

Do not let wells dry before proceeding to the next step. If an automated machine is used for the assay, follow the manufacturer's instructions for all washing steps described in this protocol.

- 3. Add 90 µl Assay Running Buffer to all wells.
- 4. Add in duplicate 10 µl Assay Running Buffer to blank wells.
- 5. Add in duplicate 10 µl Human HMW Adiponectin Standards in the order of ascending concentration to the appropriate wells. Add in duplicate 10 µl QC1 and 10 µl QC2 to the appropriate wells. Add sequentially 10 µl of the pretreated unknown samples in duplicate to the remaining wells. For best result all additions should be completed within 30 minutes.
- 6. Cover the plate with plate sealer and incubate at room temperature for 2 hours on an orbital microtiter plate shaker set to rotate at moderate speed, approximately 400 to 500 rpm.
- 7. Remove plate sealer and decant solutions from the plate. Tap as before to remove residual solutions in the wells.
- 8. Wash wells 3 times with 1X Wash Buffer, 300 μl per well per wash. Decant and tap firmly after each wash to remove residual buffer.
- 9. Add 100 µl Detection Antibody to all wells. Cover the plate with plate sealer and incubate at room temperature for 1 hour on an orbital microtiter plate shaker set to rotate at moderate speed, approximately 400 to 500 rpm.
- 10. Remove plate sealer and decant solutions from the plate. Tap as before to remove residual solutions in the wells.
- 11. Wash wells 3 times with 1X Wash Buffer, 300 μl per well per wash. Decant and tap firmly after each wash to remove residual buffer.
- 12. Add 100 μl Enzyme Solution to each well. Cover plate with sealer and incubate with moderate shaking at room temperature for 30 minutes on the microtiter plate shaker.
- 13. Remove sealer, decant solutions from the plate, and tap plate to remove the residual fluid.
- 14. Wash wells 3 times with 1X Wash Buffer, 300 μl per well per wash. Decant and tap firmly after each wash to remove residual buffer.





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- 15. Add 100 μl of Substrate (TMB) to each well, cover plate with sealer and shake on the plate shaker for approximately 5 to 20 minutes. Blue color should be formed in wells of the Human HMW Adiponectin standards with intensity proportional to increasing concentrations of Human HMW Adiponectin. Note: Please be aware that the color may develop more quickly or more slowly than the recommended incubation time depending on the localized room temperature. Please visually monitor the color development to optimize the incubation time.
- 16. Remove sealer and add 100 μl Stop Solution [CAUTION: CORROSIVE SOLUTION] and shake plate by hand to ensure complete mixing of solution in all wells. The blue color should turn to yellow after acidification. Read absorbance at 450 nm and 590 nm in a plate reader within 5 minutes and ensure that there are no air bubbles in any well. Record the difference of absorbance units. The absorbance of highest Adiponectin standard should be approximately 2.2 2.8, or not to exceed the capability of the plate reader used.



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#### **11 MICROTITER PLATE ARRANGEMENT**

Adiponectin Human HMW ELISA

12								
11								
10								
6								
8								
7								
6								
5								
4	Sample 2	Sample 2	etc.					
3	Reconst. Std.	Reconst. Std.	QC 1	QC 1	QC 2	QC 2	Sample 1	Sample 1

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2	Tube 4	Tube 4	Tube 3	Tube 3	Tube 2	Tube 2	Tube 1	Tube 1
1	Blank	Blank	Tube 7	Tube 7	Tube 6	Tube 6	Tube 5	Tube 5
	¥	В	C	D	E	Ц	Ð	Н





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#### **12 CALCULATIONS**

The dose-response curve of this assay fits best to a sigmoidal 4- or 5-parameter logistic equation. The results of unknown samples can be calculated with any computer program having a 4- or 5-parameter logistic function. Final results should be multiplied by a dilution factor of 200.

Note: When sample volumes assayed differ from 10  $\mu$ l, an appropriate mathematical adjustment must be made to accommodate for the dilution factor

(e.g., if 5 µl of sample is used, then calculated data must be multiplied by 2).

When sample volume assayed is less than 10 µl, compensate the volume deficit with Assay Running Buffer.

#### **13 INTERPRETATION**

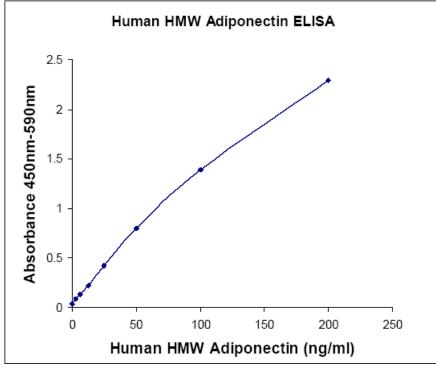
- 1. The assay will be considered accepted when all Quality Control values fall within the calculated Quality Control Range. If any QC's fall outside the control range, review results with a supervisor.
- 2. If the difference between duplicate results of a sample is >15% CV, repeat the sample.
- 3. The limit of sensitivity of this assay is 0.5 ng/ml Human HMW Adiponectin (10 µl sample size).
- 4. The appropriate range of this assay is 1.56 ng/ml to 200 ng/ml Human HMW Adiponectin (10 μl sample size). Any result greater than 200 ng/ml in a 10 μl sample should be diluted using Sample Dilution Buffer, and the assay repeated until the results fall within range.





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### HUMAN HMW ADIPONECTIN ELISA GRAPH STANDARD CURVE



Typical Standard Curve, not to be used to calculate data.

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#### 14 ASSAY CHARACTERISTICS

#### 14.1 Sensitivity

The lowest level of HMW Adiponectin that can be detected by this assay is 0.5 ng/ml when using a 10  $\mu$ l sample size.

#### 14.2 Specificity

With simple sample pretreatment, this assay specifically measures HMW Adiponectin only and does not recognize Hexameric and Trimeric Adiponectin or other human adipokines.

#### 14.3 Precision

Intra-Assay Variation

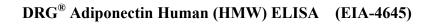
Sample No.	Mean HMW Adiponectin Levels (ng/ml)	Intra-Assay % CV
1	5.95	0.97
2	11.10	1.27
3	13.65	3.41
4	21.33	2.14
5	25.43	3.31
6	27.25	2.62
7	39.45	3.08
8	65.33	2.62

The intra-assay variations of the DRG Adiponectin Human HMW ELISA kits were studied on eight human serum samples with varying concentrations of endogenous HMW Adiponectin. The mean intra-assay variations were calculated from results of 4 duplicate determinations in each assay of the indicated samples.





Inter-Assay Variation



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Sample No.	Mean HMW Adiponectin Levels (ng/ml)	Inter-Assay % CV
1	13.33	4.13
2	21.23	8.14
3	26.60	5.03
4	28.65	3.14
5	34.78	7.79
6	38.35	9.10
7	60.75	3.01
8	61.50	3.8

The inter-assay variations of the DRG Adiponectin Human HMW ELISA kits were studied on eight human serum samples with varying concentrations of endogenous HMW Adiponectin. The mean inter-assay variations of each sample were calculated from results of three separate assays with duplicate samples in each assay.





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#### Recovery

Spike & Recovery of Human	HMW Adiponectin	in Serum

Sample No.	HMW Adiponectin Added ng/ml	Expected ng/ml	Observed ng/ml	% of Recovery
1	0	19.4	19.4	
	3.125	22.5	22.9	101.66
	25.0	44.4	49.1	110.59
	100.0	119.4	141	118.09
2	0	28.9	28.9	
	3.125	32.0	33.4	104.9
	25.0	53.9	58.3	108.16
	100.0	128.9	148.5	115.21
3	0	32.4	32.4	
	3.125	35.5	36.4	102.46
	25.0	57.4	60.4	105.23
	100.0	132.4	149.4	112.84
4	0	36.7	36.7	
	3.125	39.8	40.6	101.95
	25.0	61.7	67.2	108.91
	100.0	136.7	152.7	111.7
5	0	52.8	52.8	
	3.125	55.9	56	100.13
	25.0	77.8	81.5	104.76
	100.0	152.8	167.2	109.42
	r	r		r
6	0	85.7	85.7	
	3.125	88.8	89.6	100.87
	25.0	110.7	116.5	105.24
	100.0	185.7	203	109.32

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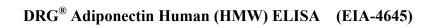


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Varying amounts of Human HMW Adiponectin were added to six Human serum samples and the HMW Adiponectin content was determined in three separate assays. The % of recovery = observed HMW Adiponectin concentrations/expected HMW Adiponectin concentrations x 100%.





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### 14.4 Linearity

Effect of Serum Dilution

Sample No.	Volume Sampled	Expected ng/ml	Observed ng/ml	% Of Expected
1	10 µl	24.5	24.5	
	5 µl	12.25	13	106.12
	2.5µl	6.13	6.3	102.86
	1.25µl	3.06	3	97.96
2	10 µl	28.1	28.1	
	5 µl	14.05	14.6	103.91
	2.5µl	7.03	7	99.64
	1.25µl	3.51	3.5	99.64
3	10 µl	31.5	31.5	
	5 µl	15.75	15.5	98.41
	2.5µl	7.88	8	101.59
	1.25µl	3.94	4.1	104.13
4	10 µl	42.6	42.6	
	5 µl	21.3	22.1	103.76
	2.5µl	10.65	10.5	98.59
	1.25µl	5.33	5.5	103.29
5	10 µl	68.4	68.4	
	5 µl	34.2	32.5	95.03
	2.5µl	17.1	17.3	101.17
	1.25µl	8.55	8.9	104.09
6	10 µl	73.1	73.1	
	5 µl	36.55	36.7	100.41
	2.5µl	18.28	18.8	102.87
	1.25µl	9.14	9.6	105.06

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Six Human serum samples with the indicated sample volumes were assayed in three separate experiments. Required amounts of assay buffer were added to compensate for lost volumes below 10  $\mu$ l. The resulting dilution factors of 1.0, 2.0, 4.0, and 8.0 representing 10  $\mu$ l, 5  $\mu$ l, 2.5  $\mu$ l, and 1.25  $\mu$ l sample volumes assayed, respectively, were applied in the calculation of observed HMW Adiponectin concentrations.

The % expected

= observed HMW Adiponectin concentrations/expected HMW Adiponectin concentrations x 100%.

#### **15 QUALITY CONTROLS**

The ranges for Quality Control 1 and 2 are provided on the card insert

#### **16 TROUBLESHOOTING GUIDE**

- 1. To obtain reliable and reproducible results the operator should carefully read this manual and fully understand all aspects of each assay step before attempting to run the assay.
- 2. Throughout the assay the operator should adhere strictly to the procedures with good laboratory practice.
- 3. Have all necessary reagents and equipment ready on hand before starting. Once the assay has been started all steps should be completed with precise timing and without interruption.
- 4. Avoid cross contamination of any reagents or samples to be used in the assay.
- 5. Make sure all reagents and samples are added to the bottom of each well.
- 6. Careful and complete mixing of solutions in the well is critical. Poor assay precision will result from incomplete mixing or cross well contamination due to inappropriate mixing.
- 7. Remove any air bubble formed in the well after acidification of substrate solution because bubbles interfere with spectrophotometric readings.
- 8. Do not let the absorbency reading of the highest standard reach 3.0 units or higher after acidification.
- 9. High absorbance in background or blank wells could be due to 1) cross well contamination by standard solution or sample or 2) inadequate washing of wells with Wash Buffer or 3) overexposure to light after substrate has been added.

#### **17 ORDERING INFORMATION**

#### A. Conditions of Sale

All products are for research or manufacturing use only. They are not intended for use in clinical diagnosis or for administration to human or animals. All products are intended for in vitro use only.

#### B. Material Safety Data Sheets (MSDS)

Material safety data sheets for DRG products may be ordered by fax or phone.









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#### SYMBOLS USED WITH DRG ELISA'S

Symbol	English	Deutsch	Francais	Español	Italiano
- <b>m</b>	Consult instructions for use	Gebrauchsanweisung beachten	Consulter les instructions d'utilisation	Consulte las instrucciones de uso	Consultare le istruzioni per l'uso
CE	European Conformity	CE-Konfirmitäts- kennzeichnung	Conformité aux normes européennes	Conformidad europea	Conformità europea
IVD	In vitro diagnostic device	In-vitro-Diagnostikum	Ussage Diagnostic in vitro	Para uso Diagnóstico in vitro	Per uso Diagnostica in vitro
RUO	For research use only	Nur für Forschungszwecke	Seulement dans le cadre de recherches	Sólo para uso en investigación	Solo a scopo di ricerca
REF	Catalogue number	Katalog-Nr.	Numéro de catalogue	Número de catálogo	Numero di Catalogo
LOT	Lot. No. / Batch code	Chargen-Nr.	Numéro de lot	Número de lote	Numero di lotto
T	Contains sufficient for <n> tests/</n>	Ausreichend für "n" Ansätze	Contenu suffisant pour "n" tests	Contenido suficiente para <n> ensayos</n>	Contenuto sufficiente per "n" saggi
$\mathbf{X}$	Storage Temperature	Lagerungstemperatur	Temperature de conservation	Temperatura de conservación	Temperatura di conservazione
$\Sigma$	Expiration Date	Mindesthaltbarkeits- datum	Date limite d'utilisation	Fecha de caducidad	Data di scadenza
AAA	Legal Manufacturer	Hersteller	Fabricant	Fabricante	Fabbricante
Distributed by	Distributor	Distributeur	Distributeur	Distribuidor	Distributtore
Content	Content	Inhalt	Conditionnement	Contenido	Contenuto
Volume/No.	Volume / No.	Volumen/Anzahl	Volume/Quantité	Volumen/Número	Volume/Quantità
Symbol	Portugues	Dansk	Svenska	Ελληνικά	
- <b></b>	Consulte as instruções de utilização	Se brugsanvisning	Se bruksanvisningen	Εγχειρίδιο χρήστη	
			e		
CE	Conformidade com as normas europeias	Europaeisk overensstemmelse	Europeisk överensstämmelse	Ευρωπαϊκή Συμμόρφωση	
C E			Europeisk	Ευρωπαϊκή Συμμόρφωση in vitro διαγνωστικό	
	normas europeias	overensstemmelse	Europeisk överensstämmelse		
IVD	normas europeias	overensstemmelse	Europeisk överensstämmelse		
IVD RUO	normas europeias Diagnóstico in vitro	overensstemmelse In vitro diagnostik	Europeisk överensstämmelse Diagnostik in vitro	in vitro διαγνωστικό	
IVD RUO REF	normas europeias Diagnóstico in vitro Catálogo n.º	overensstemmelse In vitro diagnostik Katalognummer	Europeisk överensstämmelse Diagnostik in vitro Katalog nummer	in vitro διαγνωστικό Αριθμός καταλόγου	
IVD RUO REF	normas europeias Diagnóstico in vitro Catálogo n.º	overensstemmelse In vitro diagnostik Katalognummer Lot nummer Indeholder tilsttrækkeligt	Europeisk överensstämmelse Diagnostik in vitro Katalog nummer Batch-nummer Innehåller tillräckligt till	in vitro διαγνωστικό Αριθμός καταλόγου Αριθμός Παρτίδος Περιεχόμενο επαρκές για	
IVD RUO REF	normas europeias Diagnóstico in vitro Catálogo n.º No do lote Temperatura de	overensstemmelse In vitro diagnostik Katalognummer Lot nummer Indeholder tilsttrækkeligt til "n" test	Europeisk överensstämmelse Diagnostik in vitro Katalog nummer Batch-nummer Innehåller tillräckligt till "n" tester	in vitro διαγνωστικό Αριθμός καταλόγου Αριθμός Παρτίδος Περιεχόμενο επαρκές για «ιn» εξετάσεις Θερμοκρασία	
IVD RUO REF	normas europeias Diagnóstico in vitro Catálogo n.º No do lote Temperatura de conservação	overensstemmelse In vitro diagnostik Katalognummer Lot nummer Indeholder tilsttrækkeligt til "n" test Opbevarings-temperatur	Europeisk överensstämmelse Diagnostik in vitro Katalog nummer Batch-nummer Innehåller tillräckligt till "n" tester Förvaringstempratur	in vitro διαγνωστικό Αριθμός καταλόγου Αριθμός Παρτίδος Περιεχόμενο επαρκές για «n» εξετάσεις Θερμοκρασία αποθήκευσης	
IVD RUO REF	normas europeias Diagnóstico in vitro Catálogo n.º No do lote Temperatura de conservação Prazo de validade	overensstemmelse In vitro diagnostik Katalognummer Lot nummer Indeholder tilsttrækkeligt til "n" test Opbevarings-temperatur Udløbsdato	Europeisk överensstämmelse Diagnostik in vitro Katalog nummer Batch-nummer Innehåller tillräckligt till "n" tester Förvaringstempratur Bäst före datum	in vitro διαγνωστικό Αριθμός καταλόγου Αριθμός Παρτίδος Περιεχόμενο επαρκές για «n» εξετάσεις Θερμοκρασία αποθήκευσης Ημερομηνία λήξης	
	normas europeias Diagnóstico in vitro Catálogo n.º No do lote Temperatura de conservação Prazo de validade	overensstemmelse In vitro diagnostik Katalognummer Lot nummer Indeholder tilsttrækkeligt til "n" test Opbevarings-temperatur Udløbsdato	Europeisk överensstämmelse Diagnostik in vitro Katalog nummer Batch-nummer Innehåller tillräckligt till "n" tester Förvaringstempratur Bäst före datum	in vitro διαγνωστικό Αριθμός καταλόγου Αριθμός Παρτίδος Περιεχόμενο επαρκές για «n» εξετάσεις Θερμοκρασία αποθήκευσης Ημερομηνία λήξης	