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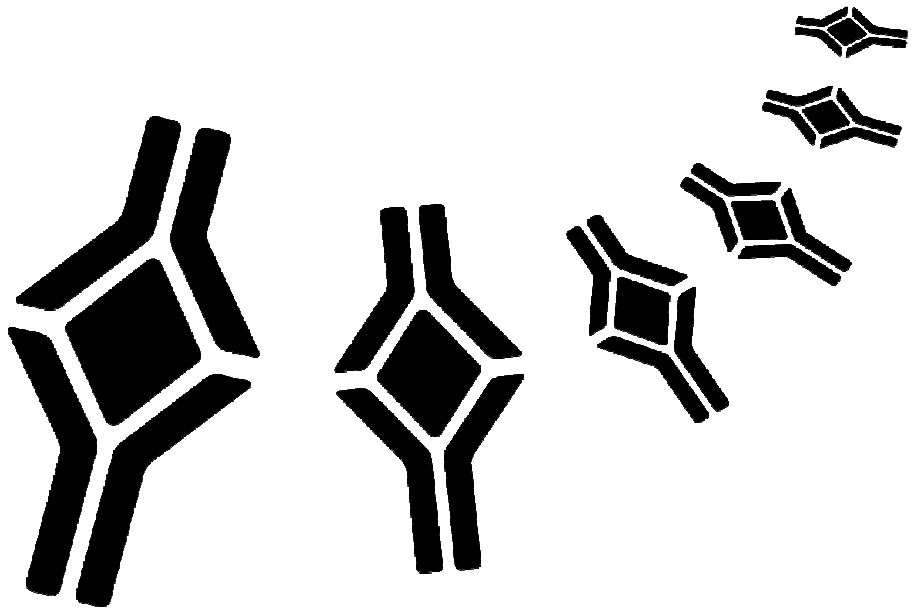
Research
and Diagnostic Products



HUMAN CTRP9 ELISA

Product Data Sheet

Cat. No.: RD191180200R

For Research Use Only

CONTENTS

» **This kit is manufactured by:**
BioVendor – Laboratorní medicína a.s.

» **Use only the current version of Product Data Sheet enclosed with the kit!**

1. INTENDED USE

The RD191180200R Human CTRP9 ELISA is a sandwich enzyme immunoassay for the quantitative measurement of human complement C1q tumor necrosis factor-related protein 9.

Features

- **It is intended for research use only**
- The total assay time is less than 4 hours
- The kit measures CTRP9 protein in human serum and plasma (EDTA, citrate, heparin)
- The kit measures CTRP9 protein in serum of several mammalian species such as dog, horse, monkey and pig
- Assay format is 96 wells
- Quality Control is human serum based. No animal sera are used
- Standard is recombinant protein based
- Components of the kit are provided ready to use, concentrated or lyophilized

2. STORAGE, EXPIRATION

Store the complete kit at 2 - 8° C. Under these conditions, the kit is stable until the expiration date (see label on the box).

For stability of opened reagents see Chapter 9.

3. INTRODUCTION

Complement C1q tumor necrosis factor-related protein 9 (C1q/TNF-related protein 9; CTRP9) is a highly conserved paralog of adiponectin [1]. Of all the CTRP paralogs, CTRP9 shows the highest degree of amino acid identity to adiponectin in its globular C1q domain [1].
CTRP9 protein exists in two isoforms, CTRP9A and CTRP9B. Although human CTRP9A and CTRP9B share 98% amino acid identity, they are encoded by distinct genes and are biochemically distinct [2]. Human CTRP9A but not CTRP9B is expressed by adipose tissue. CTRP9B is expressed at very low levels in tissues. While CTRP9A is robustly secreted as a multimeric protein, CTRP9B requires physical association with CTRP9A or adiponectin for its secretion [2].
CTRP9 is expressed predominantly in adipose tissue and females express higher levels of the transcript than males [1]. Moreover, its expression levels in *ob/ob* mice changed in an age-dependent manner, with significant up-regulation in younger mice. Adenovirus-mediated overexpression of CTRP9 in obese (*ob/ob*) mice significantly lowered serum glucose levels [1].
CTRP9 is a secreted glycoprotein with multiple post-translational modifications in its collagen domain that include hydroxylated prolines and hydroxylated and glycosylated lysines [1]. It is secreted as multimers (predominantly trimers) from transfected cells and circulates in the mouse serum with levels varying according to sex and metabolic state of mice. Furthermore, CTRP9 and adiponectin can be secreted as heterooligomers when cotransfected into mammalian cells, and *in vivo*, adiponectin/CTRP9 complexes can be reciprocally coimmunoprecipitated from the serum of adiponectin and CTRP9 transgenic mice [1].
The functional role of the plasma CTRP9 in ischemic heart disease is unknown [3]. Systemic delivery of CTRP9 reduces myocardial infarct size and apoptosis following ischemia-reperfusion in mice. CTRP9 protects cardiomyocyte from apoptosis through activation of AMP-activated protein kinase (AMPK). CTRP9 prevents acute cardiac ischemic injury via an AMPK-dependent mechanism [3].
The data indicate that CTRP9 functions to attenuate neointimal formation following vascular injury through its ability to inhibit vascular smooth muscle cell (VSMC) growth via cAMP-dependent mechanism, suggesting that the therapeutic approaches to enhance CTRP9 production could be valuable for prevention of vascular restenosis after angioplasty [4].
CTRP9 is a novel vasorelaxive adipocytokine which may exert vasculoprotective effects via the AdipoR1/AMPK/eNOS dependent/NO mediated signaling pathway [5]. The vasoactive potency of CTRP9 exceeded that of adiponectin by 3-fold [5].
Cardiac expression of CTRP9, exceeds adiponectin by >100-fold, and is significantly reduced in high-fat diet-induced diabetic mice [6]. In H9c2 cells, TNF-α strongly inhibited CTRP9 expression (>60 %), and significantly reduced peroxisome proliferator activated receptor-gamma (PPARγ), a known transcription factor promoting adiponectin expression [6].

<u>Areas of investigation:</u>
Cardiovascular disease; blood pressure regulation and NO metabolism; apoptosis
Diabetology; adipocytokines

4. TEST PRINCIPLE

In the BioVendor Human CTRP9 ELISA, Standards, Quality Control and samples are incubated in microtitration wells pre-coated with polyclonal anti-human CTRP9 antibody. After 60 minutes incubation followed by washing, biotin-labelled polyclonal anti-human CTRP9 antibody is added and incubated with the captured CTRP9 for 60 minutes. After another washing, streptavidin-HRP conjugate is added. After 30 minutes incubation and the last washing step, the remaining conjugate is allowed to react with the substrate solution (TMB). The reaction is stopped by addition of acidic solution and absorbance of the resulting yellow product is measured. The absorbance is proportional to the concentration of CTRP9. A standard curve is constructed by plotting absorbance values against CTRP9 concentrations of Standards and concentrations of unknown samples are determined using this standard curve.

5. PRECAUTIONS

- **For professional use only**
- Wear gloves and laboratory coats when handling immunodiagnostic materials
- Do not drink, eat or smoke in the areas where immunodiagnostic materials are being handled
- This kit contains components of human serum origin. These materials were found non-reactive for HBsAg, HCV antibody and for HIV 1/2 antigen and antibody. However, these materials should be handled as potentially infectious, as no test can guarantee the complete absence of infectious agents
- Avoid contact with the acidic Stop Solution and Substrate Solution, which contains hydrogen peroxide and tetramethylbenzidine (TMB). Wear gloves and eye and clothing protection when handling these reagents. Stop and/or Substrate Solutions may cause skin/eyes irritation. In case of contact with the Stop Solution and the Substrate Solution wash skin/eyes thoroughly with water and seek medical attention, when necessary
- The materials must not be pipetted by mouth

6. TECHNICAL HINTS

- Reagents with different lot numbers should not be mixed
- Use thoroughly clean glassware
- Use deionized (distilled) water, stored in clean containers
- Avoid any contamination among samples and reagents. For this purpose, disposable tips should be used for each sample and reagent
- Substrate Solution should remain colourless until added to the plate. Keep Substrate Solution protected from light
- Stop Solution should remain colourless until added to the plate. The colour developed in the wells will turn from blue to yellow immediately after the addition of the Stop Solution. Wells that are green in colour indicate that the Stop Solution has not mixed thoroughly with the Substrate Solution
- Dispose of consumable materials and unused contents in accordance with applicable national regulatory requirements

7. REAGENT SUPPLIED

Kit Components	*State*	*Quantity*
Antibody Coated Microtiter Strips	ready to use	96 wells
Biotin Labelled Antibody	ready to use	13 ml
Streptavidin-HRP Conjugate	ready to use	13 ml
Master Standard	lyophilized	2 vials
Quality Control	lyophilized	2 vials
Dilution Buffer	ready to use	50 ml
Wash Solution Conc. (10x)	concentrated	100 ml
Substrate Solution	ready to use	13 ml
Stop Solution	ready to use	13 ml
Product Data Sheet + Certificate of Analysis	-	1 pc

8. MATERIAL REQUIRED BUT NOT SUPPLIED

- Deionized (distilled) water
- Test tubes for diluting samples
- Glassware (graduated cylinder and bottle) for Wash Solution (Dilution Buffer)
- Precision pipettes to deliver 10 – 1 000 µl with disposable tips
- Multichannel pipette to deliver 100 µl with disposable tips
- Absorbent material (e.g. paper towels) for blotting the microtitrate plate after washing
- Vortex mixer
- Thermostatic box adjustable to 37° C
- Orbital microplate shaker capable of approximately 300 rpm
- Microplate washer (optional). [Manual washing is possible but not preferable.]
- Microplate reader with 450 ± 10 nm filter, preferably with reference wavelength 630 nm (alternatively another one from the interval 550 – 650 nm)
- Software package facilitating data generation and analysis (optional)

9. PREPARATION OF REAGENTS

» **All reagents need to be brought to room temperature prior to use**

» **Always prepare only the appropriate quantity of reagents for your test**

» **Do not use components after the expiration date marked on their label**

- **Assay reagents supplied ready to use:**

Antibody Coated Microtiter Strips
Stability and storage:
Return the unused strips to the provided aluminium zip-sealed bag with desicant and seal carefully. Remaining Microtiter Strips are stable 3 months stored at 2 - 8° C and protected from the moisture.

Biotin Labelled Antibody
Streptavidin-HRP Conjugate
Dilution Buffer
Substrate Solution
Stop Solution
Stability and storage:
Opened reagents are stable 3 months when stored at 2 - 8° C.

- **Assay reagents supplied concentrated or lyophilized:**

Human CTRP9 Master Standard
Refer to the Certificate of Analysis for current volume of Dilution Buffer needed for reconstitution of standard!!!
Reconstitute the lyophilized Master Standard with Dilution Buffer just prior to the assay. Let it dissolve at least 15 minutes with occasional gentle shaking (not to foam). The resulting concentration of the human CTRP9 in the stock solution is **20 ng/ml**.

Prepare set of standards using Dilution Buffer as follows:

Volume of Standard	*Dilution Buffer*	*Concentration*
Stock	-	20 ng/ml
25 µl of stock	600 µl	800 pg/ml
250 µl of 800 pg/ml	250 µl	400 pg/ml
250 µl of 400 pg/ml	250 µl	200 pg/ml
250 µl of 200 pg/ml	250 µl	100 pg/ml
250 µl of 100 pg/ml	250 µl	50 pg/ml
250 µl of 50 pg/ml	250 µl	25 pg/ml

Prepared Standards are ready to use, do not dilute them.
Stability and storage:
Do not store the reconstituted and/or diluted Standard solutions.

Quality Control
Refer to the Certificate of Analysis for current volume of Dilution Buffer needed for reconstitution and for current Quality Control concentration!!!
Reconstitute Quality Control with Dilution Buffer just prior to the assay. Let it dissolve at least 15 minutes with occasional gentle shaking (not to foam). Vortex is recommended.

Dilute the reconstituted Quality Control 20x with Dilution Buffer, e.g. 10 µl of Quality Control + 190 µl of Dilution Buffer for singlets, or preferably 15 µl of Quality Control + 285 µl of Dilution Buffer for duplicates. **Mix well** (not to foam). Vortex is recommended.

Stability and storage:
Do not store the reconstituted and/or diluted Quality Control.

Note:
Concentration of analyte in Quality Control need not be anyhow associated with normal and/or pathological concentrations in serum or another body fluid. Quality Control serves just for control that the kit works in accordance with PDS and CoA and that ELISA test was carried out properly.

Wash Solution Conc. (10x)
Dilute Wash Solution Concentrate (10x) ten-fold in distilled water to prepare a 1x working solution. Example: 100 ml of Wash Solution Concentrate (10x) + 900 ml of distilled water for use of all 96-wells.
Stability and storage:
The diluted Wash Solution is stable 1 month when stored at 2 - 8° C. Opened Wash Solution Concentrate (10x) is stable 3 months when stored at 2 - 8° C.

10. PREPARATION OF SAMPLES

The kit measures human CTRP9 in serum and plasma (EDTA, citrate, heparin).

Samples can be assayed immediately after collection, or after long-term storage at -20° C (-70° C). Thoroughly mix thawed samples just prior to the assay and avoid repeated freeze/thaw cycles, which may cause erroneous results. Avoid using hemolyzed or lipemic samples.

An appropriate dilution should be assessed by the researcher (due to the large variability of serum CTRP9 levels different individuals) in advance to batch measurement. Recommended starting dilution is 20x.

Dilute samples 20x with Dilution Buffer just prior to the assay, e.g. 10 µl of sample + 190 µl of Dilution Buffer for singlets, or preferably 15 µl of sample + 285 µl of Dilution Buffer for duplicates. **Mix well** (not to foam). Vortex is recommended.

Stability and storage:
Samples should be stored at -20° C, or preferably at -70° C or lower for long-term storage. Avoid repeated freeze/thaw cycles.
Do not store the diluted samples.

See Chapter 13 for stability of serum and plasma samples when stored at 2 - 8° C, effect of freezing/thawing and effect of sample matrix (serum/plasma) on the concentration of human CTRP9.

Note: It is recommended to use a precision pipette and a careful technique to perform the dilution in order to get precise results.

11. ASSAY PROCEDURE

1. Pipet **100 µl** of diluted Standards, Quality Control, Dilution Buffer (= Blank) and diluted samples, preferably in duplicates, into the appropriate wells. See *Figure 1* for example of work sheet.
2. Incubate the plate at 37° C for **1 hour**, no shaking.
3. Wash the wells 3-times with Wash Solution (0.35 ml per well). After final wash, invert and tap the plate strongly against paper towel.
4. Add **100 µl** of Biotin Labelled Antibody solution into each well.
5. Incubate the plate at room temperature (ca. 25° C) for **1 hour**, shaking at ca. 300 rpm on an orbital microplate shaker.
6. Wash the wells 3-times with Wash Solution (0.35 ml per well). After final wash, invert and tap the plate strongly against paper towel.
7. Add **100 µl** of Streptavidin-HRP Conjugate into each well.
8. Incubate the plate at room temperature (ca. 25° C) for **30 min**, shaking at ca. 300 rpm on an orbital microplate shaker.
9. Wash the wells **3-times** with Wash Solution (0.35 ml per well). After final wash, invert and tap the plate strongly against paper towel.
10. Add **100 µl** of Substrate Solution into each well. Avoid exposing the microtiter plate to direct sunlight. Covering the plate with e.g. aluminium foil is recommended.
11. Incubate the plate for **10 minutes** at room temperature. The incubation time may be extended [up to 20 minutes] if the reaction temperature is below than 20° C. Do not shake the plate during the incubation.
12. Stop the colour development by adding **100 µl** of Stop Solution.
13. Determine the absorbance of each well using a microplate reader set to 450 nm, preferably with the reference wavelength set to 630 nm (acceptable range: 550 - 650 nm). Subtract readings at 630 nm (550 - 650 nm) from the readings at 450 nm. **The absorbance should be read within 5 minutes following step 12.**

Note: If some samples and standard/s have absorbances above the upper limit of your microplate reader, perform a second reading at 405 nm. A new standard curve, constructed using the values measured at 405 nm, is used to determine CTRP9 concentration of off-scale standards and samples. The readings at 405 nm should not replace the readings for samples that were "in range" at 450 nm.

Note 2: Manual washing: Aspirate wells and pipet 0.35 ml Wash Solution into each well. Aspirate wells and repeat twice. After final wash, invert and tap the plate strongly against paper towel. Make certain that Wash Solution has been removed entirely.

	strip 1+2	strip 3+4	strip 5+6	strip 7+8	strip 9+10	strip 11+12
A	**Standard 800**	Sample 1	Sample 9	Sample 17	Sample 25	Sample 33
B	**Standard 400**	Sample 2	Sample 10	Sample 18	Sample 26	Sample 34
C	**Standard 200**	Sample 3	Sample 11	Sample 19	Sample 27	Sample 35
D	**Standard 100**	Sample 4	Sample 12	Sample 20	Sample 28	Sample 36
E	**Standard 50**	Sample 5	Sample 13	Sample 21	Sample 29	Sample 37
F	**Standard 25**	Sample 6	Sample 14	Sample 22	Sample 30	Sample 38
G	**Blank**	Sample 7	Sample 15	Sample 23	Sample 31	Sample 39
H	**QC**	Sample 8	Sample 16	Sample 24	Sample 32	Sample 40

Figure 1: Example of a work sheet.

12. CALCULATIONS

Most microtiter plate readers perform automatic calculations of analyte concentration. The Standards curve is constructed by plotting the mean absorbance of Standards (Y) against the known concentration of Standards (X) in logarithmic scale, using the four-parameter algorithm. Results are reported as concentration of CTRP9 (pg/ml) in samples.

Alternatively, the *logit log* function can be used to linearize the standard curve, i.e. *logit* of the mean absorbance (Y) is plotted against *log* of the known concentration of Standards (X).

The measured concentration of samples and Quality Control calculated from the standard curve must be multiplied by their respective dilution factor, because samples and Quality Control have been diluted prior to the assay, e.g. 60 pg/ml (from standard curve) x 20 (dilution factor) = 1.2 ng/ml.

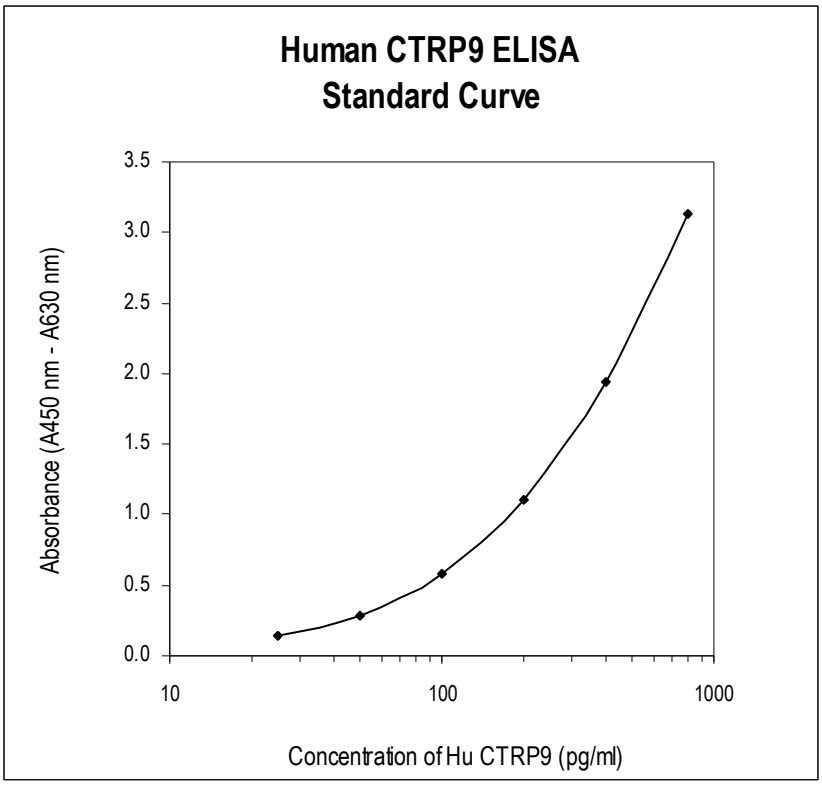


Figure 2: Typical Standard Curve for Human CTRP9 ELISA.

13. PERFORMANCE CHARACTERISTICS

» Typical analytical data of BioVendor Human CTRP9 ELISA are presented in this chapter

- **Sensitivity**

Limit of detection (LOD) (defined as concentration of analyte giving absorbance higher than mean absorbance of blank* plus three standard deviations of the absorbance of blank: A_{blank} + $3xSD_{blank}$) is calculated from the real human CTRP9 values in wells and is: 9.0 pg/ml.
* Dilution Buffer is pipetted into blank wells.

- **Limit of Assay**

Samples with absorbances exceeding the absorbance of the highest standard should be measured again with higher dilution. The final concentration of samples calculated from the standard curve must be multiplied by the respective dilution factor.

- **Specificity**

The antibodies used in this ELISA are specific for human CTRP9 with no detectable crossreactivities to the following proteins: CTRP1, CTRP3, CTRP6, CTRP7, CTRP8 at 10 ng/ml and ADIPONECTIN at 500 ng/ml.

Sera of several mammalian species were measured in the assay. See results below.
For details please contact us at info@biovendor.com

Mammalian serum Sample	*Observed crossreactivity*
Bovine	no
Cat	no
Dog	yes
Goat	no
Hamster	no
Horse	yes
Monkey	yes
Mouse	no
Pig	yes
Rabbit	no
Rat	no
Sheep	no

» **Presented results are multiplied by respective dilution factor**

- **Precision**

Intra-assay (Within-Run) (n=8)

Sample	*Mean (ng/ml)*	*SD (ng/ml)*	*CV (%)*
1	2.27	0.09	4.1
2	1.34	0.09	6.8

Inter-assay (Run-to-Run) (n=8)

Sample	*Mean (ng/ml)*	*SD (ng/ml)*	*CV (%)*
1	7.08	0.65	9.1
2	2.17	0.15	6.7

- **Spiking Recovery**

Serum samples were spiked with different amounts of human CTRP9 and assayed.

Sample	***O**bserved (ng/ml)*	***E**xpected (ng/ml)*	*Recovery **O/E** (%)*
1	1.72	-	-
	3.60	3.72	96.8
	5.40	5.72	94.4
	10.20	9.72	104.9
2	11.76	-	-
	15.36	15.76	97.5
	20.16	19.76	102.0
	29.76	27.76	107.2

- **Linearity**

Serum samples were serially diluted with Dilution Buffer and assayed.

Sample	*Dilution*	***O**bserved (ng/ml)*	***E**xpected (ng/ml)*	*Recovery **O/E** (%)*
1	-	9.16	-	-
	2x	4.52	4.58	98.7
	4x	2.16	2.29	94.3
	8x	1.20	1.15	104.8
2	-	24.88	-	-
	2x	12.48	12.44	100.3
	4x	6.00	6.22	96.5
	8x	3.28	3.11	105.5

- **Effect of sample matrix**

EDTA, citrate and heparin plasma samples were compared to respective serum samples from the same 10 individuals. Results are shown below:

Volunteer No.	*Serum (ng/ml)*	*Plasma (ng/ml)*		
		EDTA	*Citrate*	*Heparin*
1	3.62	4.50	3.52	4.98
2	1.04	0.90	0.72	0.96
3	15.76	15.68	10.24	12.64
4	11.56	9.48	7.44	9.88
5	4.50	3.46	3.48	4.06
6	0.58	0.50	0.40	0.44
7	2.38	2.50	2.06	2.90
8	13.98	12.50	12.26	15.68
9	5.24	4.92	4.20	5.50
10	4.56	4.08	3.64	4.52
Mean (ng/ml)	**6.32**	**5.85**	**4.80**	**6.16**
Mean Plasma/Serum (%)		**92.6**	**75.9**	**97.4**
Coefficient of determination R^2		**0.98**	**0.95**	**0.94**

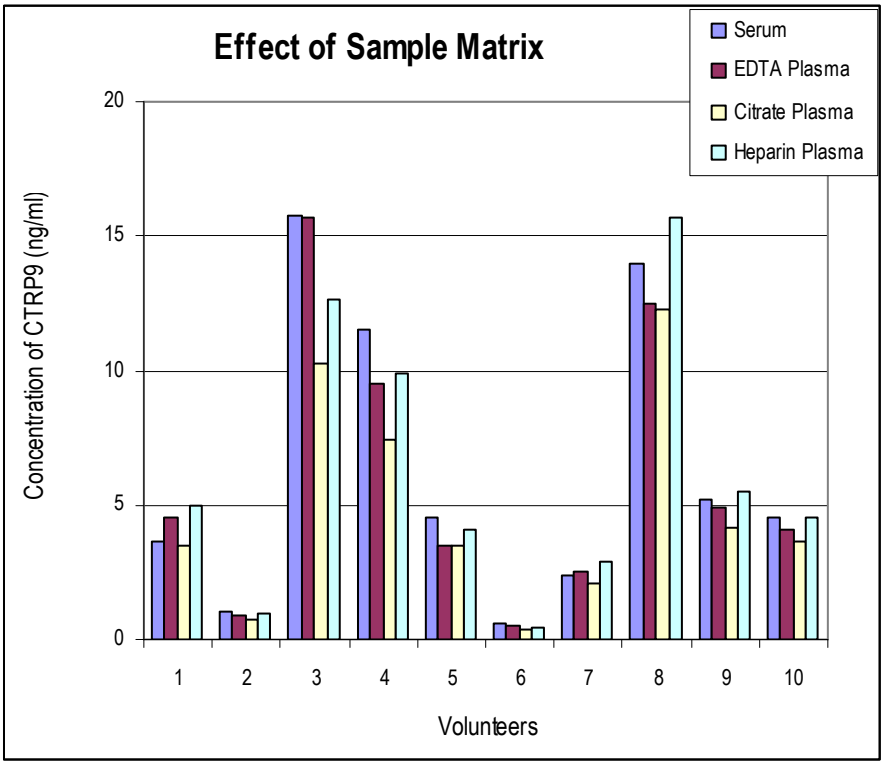


Figure 3: CTRP9 levels measured using Human CTRP9 ELISA from 10 individuals using serum, EDTA, citrate and heparin plasma, respectively.

- **Stability of samples stored at 2-8°C**

Samples should be stored at -20° C, or preferably at -70° C. However, no significant decline in concentration of human CTRP9 was observed in serum and plasma samples after 7 days when stored at 2 - 8° C. To avoid microbial contamination, samples were treated with ε-aminocaproic acid and sodium azide, resulting in the final concentration of 0.03% and 0.1%, respectively.

Sample	*Storage Conditions*	*Serum (ng/ml)*	*Plasma (ng/ml)*		
			EDTA	*Citrate*	*Heparin*
1	-20° C	11.76	10.88	9.12	6.08
	2 - 8° C, 1 day	12.08	11.04	9.12	5.52
	2 - 8° C, 7 days	10.96	10.56	8.88	5.36
2	-20° C	4.50	3.46	3.48	4.06
	2 - 8° C, 1 day	3.94	3.68	3.16	3.84
	2 - 8° C, 7 days	4.36	3.58	3.12	4.00
3	-20° C	0.58	0.50	0.40	0.44
	2 - 8° C, 1 day	0.50	0.38	0.44	0.44
	2 - 8° C, 7 days	0.40	0.42	0.34	0.50

- **Effect of Freezing/Thawing**

No decline was observed in concentration of human CTRP9 in serum and plasma samples after repeated (5x) freeze/thaw cycles. However it is recommended to avoid unnecessary repeated freezing/thawing of the samples.

Sample	*Number of f/t cycles*	*Serum (ng/ml)*	*Plasma (ng/ml)*		
			EDTA	*Citrate*	*Heparin*
1	1x	101.12	92.16	72.32	97.28
	3x	97.28	99.84	96.64	90.88
	5x	115.20	102.40	90.88	103.04
2	1x	2.50	2.54	2.16	2.98
	3x	2.72	2.96	2.24	2.92
	5x	2.58	2.74	2.14	2.78
3	1x	112.96	108.16	88.64	114.88
	3x	104.64	129.60	94.40	119.68
	5x	112.64	111.04	87.36	109.12

14. DEFINITION OF THE STANDARD

The recombinant human CTRP9A protein is used as the standard. The recombinant CTRP9A produced in *E. coli* is a 33.7 kDa protein consisting of 324 amino acid residues of human CTRP9A and 10 extra AA.

15. PRELIMINARY POPULATION DATA

The following results were obtained when serum samples from 155 unselected donors (89 men + 66 women) 20 - 65 years old were assayed with the BioVendor Human CTRP9 ELISA in our laboratory.

Sex	Age (years)	n	Mean	SD	Min	Max	Median
			CTRP9 (ng/ml)				
Men	20-39	42	7.08	12.69	0.00	62.08	1.38
	40-65	47	7.79	21.27	0.00	124.80	0.21
Women	20-39	38	9.82	36.30	0.00	227.84	1.27
	40-61	28	3.97	11.54	0.02	58.88	0.15

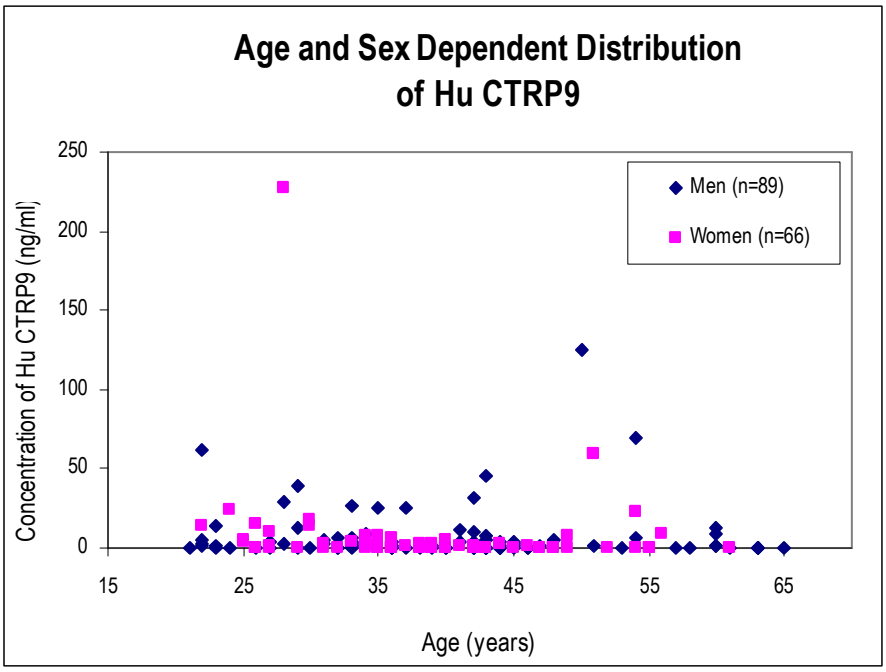


Figure 4a: Human CTRP9 concentration plotted against donor age and sex.

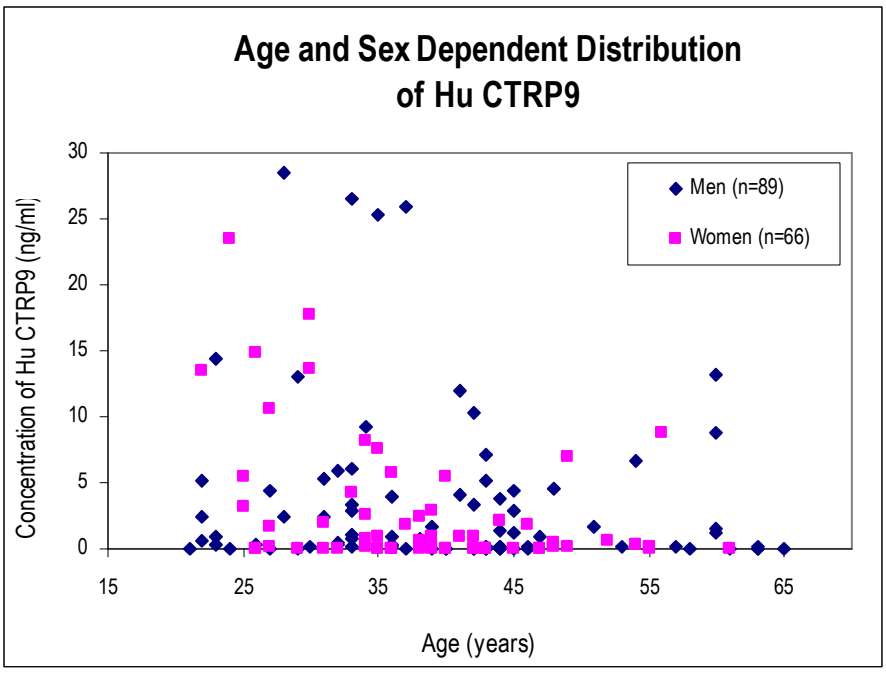


Figure 4b: Human CTRP9 concentration plotted against donor age and sex.

- **Reference range**

It is recommended that each laboratory include its own panel of control sample in the assay. Each laboratory should establish its own normal and pathological reference ranges for CTRP9 levels with the assay.

16. METHOD COMPARISON

The BioVendor Human CTRP9 ELISA has not been compared to any commercial immunoassay.

17. TROUBLESHOOTING AND FAQS

›› Weak signal in all wells

Possible explanations:

- Omission of a reagent or a step
- Improper preparation or storage of a reagent
- Assay performed before reagents were allowed to come to room temperature
- Improper wavelength when reading absorbance

›› High signal and background in all wells

Possible explanations:

- Improper or inadequate washing
- Overdeveloping; incubation time with Substrate Solution should be decreased before addition of Stop Solution

›› High coefficient of variation (CV)

Possible explanation:

- Improper or inadequate washing
- Improper mixing Standards or samples

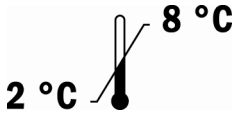
18. REFERENCES

References to CTRP9:

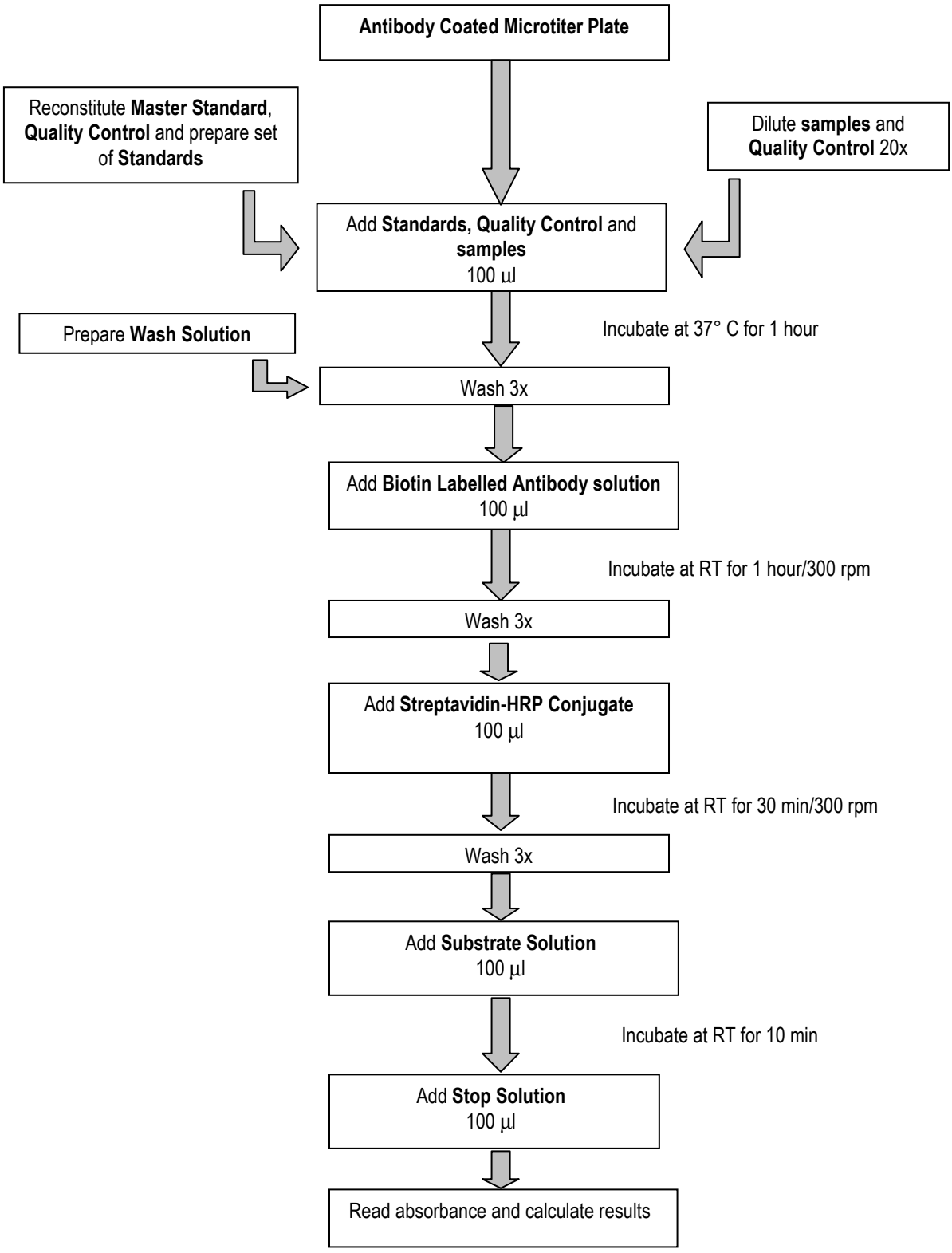
1. Wong GW, Krawczyk SA, Kitidis-Mitrokostas C, Ge G, Spooner E, Hug C, Gimeno R, Lodish HF. Identification and characterization of CTRP9, a novel secreted glycoprotein, from adipose tissue that reduces serum glucose in mice and forms heterotrimers with adiponectin. *FASEB J.* **23** (1): 241-258. (2009)
2. Peterson JM, Wei Z, Wong GW. CTRP8 and CTRP9B are novel proteins that hetero-oligomerize with C1q/TNF family members. *Biochem Biophys Res Commun.* **388**(2): 360-5. (2009)
3. Kambara T, Ohashi K, Shibata R, Ogura Y, Maruyama S, Enomoto T, Uemura Y, Shimizu Y, Yuasa D, Matsuo K, Miyabe M, Kataoka Y, Murohara T, Ouchi N. CTRP9 protein protects against myocardial injury following ischemia-reperfusion through AMP-activated protein kinase (AMPK)-dependent mechanism. *J Biol Chem.* **287**(23): 18965-73. (2012)
4. Uemura Y, Shibata R, Ohashi K, Enomoto T, Kambara T, Yamamoto T, Ogura Y, Yuasa D, Joki Y, Matsuo K, Miyabe M, Kataoka Y, Murohara T, Ouchi N. Adipose-derived factor CTRP9 attenuates vascular smooth muscle cell proliferation and neointimal formation. *FASEB J.* **27**(1): 25-33. (2013)
5. Zheng Q, Yuan Y, Yi W, Lau WB, Wang Y, Wang X, Sun Y, Lopez BL, Christopher TA, Peterson JM, Wong GW, Yu S, Yi D, Ma XL. C1q/TNF-related proteins, a family of novel adipokines, induce vascular relaxation through the adiponectin receptor-1/AMPK/eNOS/nitric oxide signaling pathway. *Arterioscler Thromb Vasc Biol.* **31**(11): 2616-23. (2011)
6. Su H, Yuan Y, Wang XM, Lau WB, Wang Y, Wang X, Gao E, Koch WJ, Ma XL. Inhibition of CTRP9, a novel and cardiac-abundantly expressed cell survival molecule, by TNFα-initiated oxidative signaling contributes to exacerbated cardiac injury in diabetic mice. *Basic Res Cardiol.* **108**(1): 315. (2013)

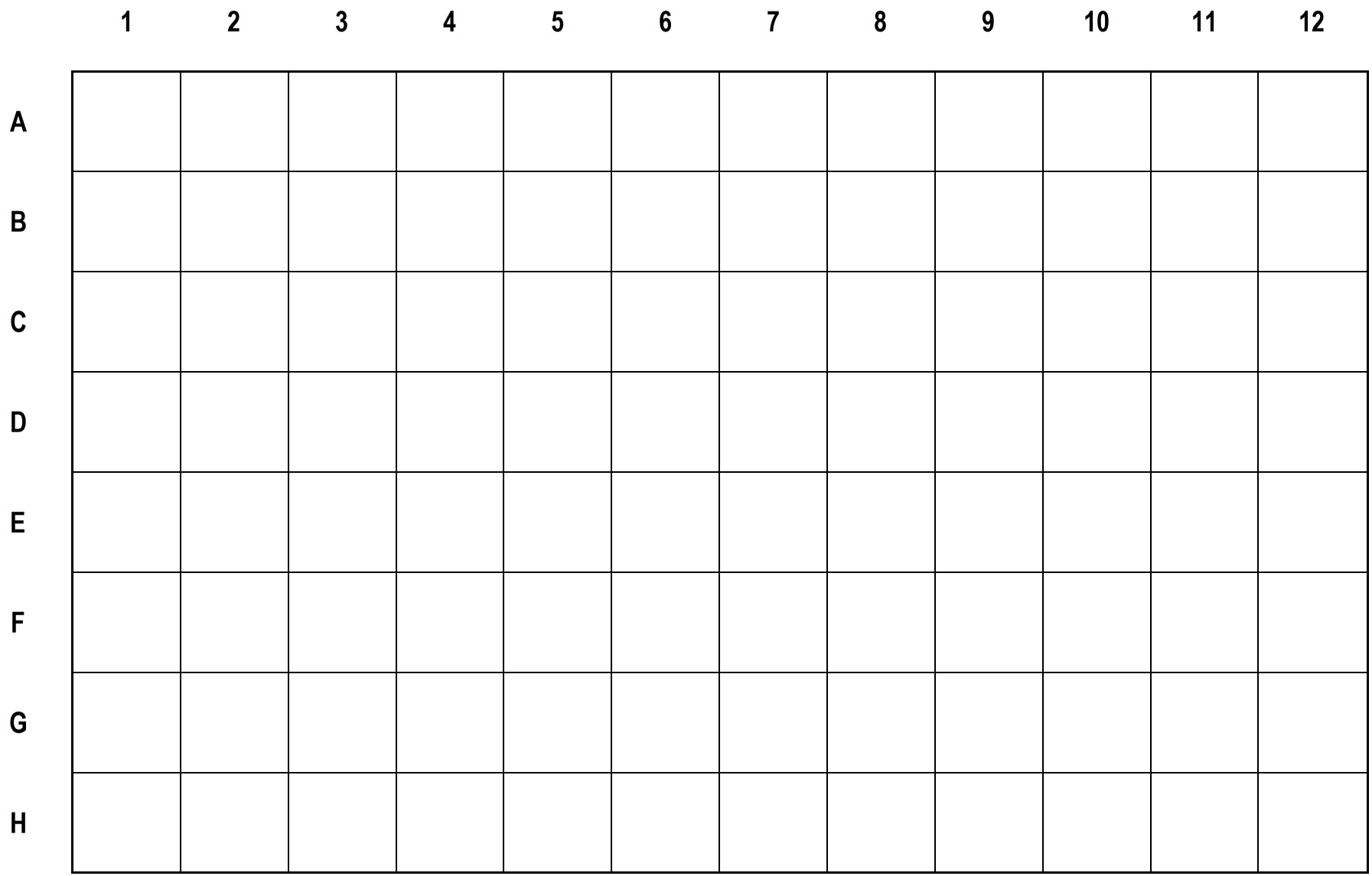
For more references on this product see our WebPages at www.biovendor.com

19. EXPLANATION OF SYMBOLS

REF	Catalogue number
Cont.	Content
LOT	Lot number
	See instructions for use
	Biological hazard
	Expiry date
	Storage conditions
	Identification of packaging materials

Assay Procedure Summary

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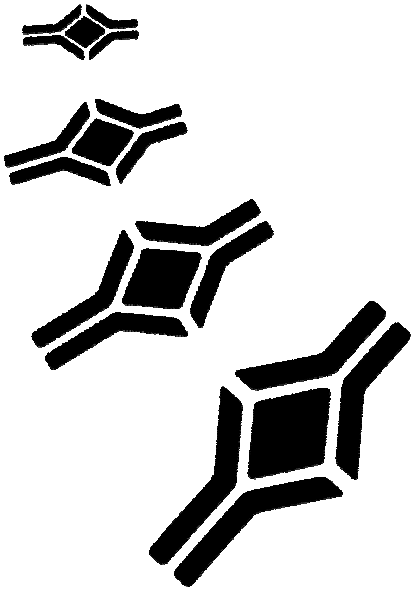

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