Instructions for Use

Prostaglandin E_2 ELISA

Enzyme Immunoassay for the quantitative determination of Prostaglandin E_2 (PGE_2) in serum, plasma, urine, saliva and tissue culture media samples.

EIA- 5811

96 wells

DRG Instruments GmbH, Germany
Frauenbergstraße. 18, D-35039 Marburg
Phone: +49 (0)6421-1700 0, Fax: +49 (0)6421-1700 50
Website: www.drg-diagnostics.de
E-mail: drg@drg-diagnostics.de

Distributed by:

DRG International, Inc., USA
841 Mountain Ave., Springfield, NJ 07081
Phone: (973) 564-7555, Fax: (973) 564-7556
Website: www.drg-international.com
E-mail: corp@drg-international.com

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Please use only the valid version of the package insert provided with the kit.
Verwenden Sie nur die jeweils gültige, im Testkit enthaltene, Gebrauchsanweisung.
Si prega di usare la versione valida dell'inserto del pacco a disposizione con il kit.
Por favor, se usa solo la version valida de la metodico técnico incluido aqui en el kit.

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Species Independent

Sample Types Validated:
Saliva, Urine, Serum, EDTA and Heparin Plasma and Tissue Culture Media

FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES.

1 BACKGROUND

Eicosanoid signal transduction pathways are highly conserved and are involved in a number of physiological processes. Prostaglandins are synthesized from arachidonic acid by cyclooxygenase (COX)-1 or -2, which convert the acid into PGH_2. This is further processed by cytosolic or microsomal prostaglandin synthases to become PGE_2 or one of several other prostanoids[1-3]. Prostacyclin is the major cyclooxygenase product in blood vessel walls and it is present in inflammatory fluids in similar concentrations to PGE_2. Prostacyclin is a potent vasodilator and is more potent than PGE_2 in producing hyperalgesia[4]. PGE_2 is produced by a wide variety of tissues[5-14] and in several pathological conditions, including inflammation, arthritis, fever, tissue injury, endometriosis, and a variety of cancers[5,6].

Other biological actions of PGE_2 include vasodilation, modulation of sleep/wake cycles, and facilitation of human immunodeficiency virus replication. It elevates cAMP levels, stimulates bone resorption, and has thermoregulatory effects. It has been shown to be a regulator of sodium excretion and renal hemodynamics[7-12].

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11. L.G. Raisz, et al., “Effect of prostaglandin endoperoxides and metabolites on bone resorption in vitro.” Nature, (1977) 267: 532-534.
12. C.R. Long, Kinoshita Y, Knox FG., “Prostaglandin E_2 induced changes in renal blood flow, renal interstitial hydrostatic pressure and sodium excretion in the rat.” Prostaglandins, (1990) 40: 591-601.

2 ASSAY PRINCIPLE

The Prostaglandin E_2 (PGE_2) ELISA kit is designed to quantitatively measure PGE_2 present in serum, plasma, urine, saliva, tissue and tissue culture media samples.

This ELISA kit allows for the widest variations in sample size, sensitivity and assay timing of any PGE_2 kit. The protocol variations are outlined below.

Please read the complete kit insert before performing this assay. A PGE_2 standard is provided to generate a standard curve for the assay and all samples should be read off the standard curve. Standards or diluted samples are pipetted into a clear microtiter plate coated with an antibody to capture mouse IgG. A PGE_2-peroxidase conjugate is added to the standards and samples in the wells. The binding reaction is initiated by the addition of a monoclonal antibody to PGE_2 to each well. After incubation, the plate is washed and substrate is added. The substrate reacts with the bound PGE_2-peroxidase conjugate. After a short incubation, the reaction is stopped and the intensity of the generated color is detected in a microtiter plate reader capable of measuring 450 nm wavelength.

The concentration of the PGE_2 in the sample is calculated, after making suitable correction for the dilution of the sample, using software available with most plate readers.

3 ASSAY FORMAT OPTIONS

Multi-Format Assay

This Prostaglandin E_2 (PGE_2) Immunoassay kit uses a mouse monoclonal antibody that allows for an exceptional wide range of PGE_2 concentrations to be measured. By varying the volume of sample used in the assay PGE_2 concentrations from 1,000 pg/mL to below 2 pg/mL can be determined. This allows the most sensitive detection of PGE_2 to be measured in any sample.

The monoclonal antibody, clone 3H10, displays extremely fast kinetics for binding PGE_2 and incubation for 2 hours or overnight yields identical binding curves (OD variation will be seen between 2 hour and overnight incubation but %B/B0 curves will be very similar). This lack of sensitivity to the time of incubation allows any format of the PGE_2 assay to be run to fit your workflow.

Regular Format:

For samples with PGE_2 concentrations from 500 to 3.9 pg/mL

The Regular Format uses 50 µL of sample or standard to give results in 2.5 hours.

Low Sample Volume Format:

For samples with PGE_2 concentrations from 1,000 to 15.6 pg/mL

The Low Sample Volume Format uses 25 µL of sample or standard for results in 2.5 hours, but uses lower sample volumes.

High Sensitivity Format:

For samples with PGE_2 concentrations from 500 to 1.95 pg/mL

The High Sensitivity Format uses 100 µL of sample or standard and gives results in 2.5 hours, but is the highest sensitivity kit of any type available.

4 SUPPLIED COMPONENTS

Coated Clear 96 Well Plate
A **c**lear plastic microtiter plate coated with goat anti-mouse IgG.
1 each

Prostaglandin E_2 Standard
Prostaglandin E_2 at 20,000 pg/mL in a special stabilizing solution.
Must be stored at -20 °C.
70 µL

Prostaglandin E_2 Antibody
A mouse monoclonal antibody specific for Prostaglandin E_2.
3 mL

Prostaglandin E_2 Conjugate
A Prostaglandin E_2-peroxidase conjugate in a special stabilizing solution.
Must be stored at -20 °C.
3 mL

Assay Buffer Concentrate
A 5X concentrate that must be diluted with deionized or distilled water.
28 mL

Wash Buffer Concentrate
A 20X concentrate that should be diluted with deionized or distilled water.
30 mL

TMB Substrate
11 mL

Stop Solution
A 1 M solution of hydrochloric acid. CAUSTIC.
5 mL

Plate Sealer
1 each

5 STORAGE INSTRUCTIONS

The unopened kit must be stored at -20 °C.

Once opened the kit can be stored at 4 °C up to the expiration date on the kit label, **except for the PGE$_2$ Standard and PGE$_2$ Conjugate. These must be stored at -20 °C.**

The PGE2 Conjugate will lose about 40% of its signal when stored at -20 °C. No change in %B/B0 will be seen for standards or samples. It can be stored at -80 °C without loss of signal up to the expiration date on the kit label.

The frozen PGE$_2$ Conjugate can be freeze-thawed multiple times.

6 OTHER MATERIALS REQUIRED

- Distilled or deionized water.
- Repeater pipet, such as an Eppendorf repeater, with disposable tips to accurately dispense 25, 50 and 100 µL.
- A microplate shaker.
- Colorimetric 96 well microplate reader capable of reading optical density at 450 nm.
- Software for converting raw relative optical density readings from the plate reader and carrying out four parameter logistic curve (4PLC) fitting. Contact your plate reader manufacturer for details.

7 PRECAUTIONS

As with all such products, this kit should only be used by qualified personnel who have had laboratory safety instruction. The complete insert should be read and understood before attempting to use the product.

The antibody coated plate needs to be stored desiccated. The silica gel pack included in the foil ziploc bag will keep the plate dry. The silica gel pack will turn from blue to pink if the ziploc has not been closed properly.

This kit utilizes a peroxidase-based readout system. Buffers, including other manufacturers Wash Buffers, containing sodium azide will inhibit color production from the enzyme. Make sure **all** buffers used for samples are **azide free**. Ensure that any plate washing system is rinsed well with deionized water prior to using the supplied Wash Buffer as prepared in chapter 10.

The Stop Solution is acid. The solution should not come in contact with skin or eyes. Take appropriate precautions when handling this reagent.

8 SAMPLE TYPES

Prostaglandin E_2 (PGE_2) is identical across all species and we expect this kit may measure PGE_2 from sources other than human. The end user should evaluate recoveries of PGE_2 in other samples being tested.

This assay has been validated for saliva, urine, serum, EDTA and heparin plasma samples and for tissue culture samples.

A general cyclooxygenase inhibitor, such as meclofenamic acid or indomethacin at 15 µM should be added immediately after collection of any biological samples, such as serum and plasma.

All samples should be frozen rapidly in dry ice/ethanol and **stored at -80 °C**.

Samples containing visible particulates should be centrifuged prior to use.

Severely hemolyzed samples should not be used in this kit.

All samples with high lipid content may interfere with the measurement of PGE_2 and may be extracted as described below. An online resource for the extraction of bioactive lipids can be found at: http://lipidlibrary.aocs.org/topics/spe alm/index.htm#ext.

9 SAMPLE VALUES

The normal reference range for serum Prostaglandin E_2 (containing COX inhibitors) is 25 - 1,000 pg/mL.

Typical normal mouse PGE_2 serum levels are 45 - 150 ng/mL.

Normal 24-hour urine PGE_2 levels are between 400 - 620 ng/24 hours.

10 SAMPLE PREPARATION

Serum and Plasma Samples

Serum and plasma samples should be diluted ≥ 1:10 with the supplied diluted Assay Buffer prior running in the assay.

Mouse serum and plasma samples need to be diluted ≥ 1:20 with the supplied diluted Assay Buffer prior running in the assay to minimize any interference of mouse IgG on the assay. Typical normal mouse PGE_2 serum levels are 45 - 150 ng/mL.

Urine Samples

Urine samples should be diluted ≥ 1:8 with the supplied diluted Assay Buffer prior running in the assay.

Saliva Samples

Saliva samples should be diluted ≥ 1:2 with the supplied diluted Assay Buffer prior running in the assay.

For Saliva Sample Handling Instructions please contact drg@drg-diagnostics.de.

Tissue Culture Media

For measuring prostaglandin E_2 in tissue culture media (TCM), samples should be read off a standard curve generated in TCM. Samples may need to be diluted further in TCM. We have validated the assay using RPMI-1640.

Extracted Samples

For a detailed Extraction protocol please contact drg@drg-diagnostics.de.

The ethanol concentration in the final diluted Assay Buffer dilution added to the well should be < 5%.

Use all samples within 2 hours of preparation.

11 ASSAY REAGENT PREPARATION

Allow the kit reagents to thaw and come to room temperature for 30 - 60 minutes. We recommend that all standards and samples be run in duplicate to allow the end user to accurately determine prostaglandin E_2 concentrations. Ensure that all samples have reached room temperature and have been diluted as appropriate prior to running them in the kit.

11.1 Assay Buffer

Dilute Assay Buffer Concentrate 1:5 by adding one part of the concentrate to four parts of deionized water.

Once diluted this is stable at 4 °C for 3 months.

11.2 Wash Buffer

Dilute Wash Buffer Concentrate 1:20 by adding one part of the concentrate to nineteen parts of deionized water.

Once diluted this is stable at room temperature for 3 months.

12 REGULAR FORMAT

12.1 Standard Preparation - Regular Format

Label test tubes as #1 through #8.

Pipet 390 µL of Assay Buffer into tube #1 and 200 µL into tubes #2 to #8.

The Prostaglandin E_2 stock solution contains an organic solvent. Prerinse the pipet tip several times to ensure accurate delivery.
Carefully add 10 µL of the Prostaglandin E_2 stock solution to tube #1 and vortex completely.
Take 200 µL of the Prostaglandin E_2 solution in tube #1 and add it to tube #2 and vortex completely.
Repeat the serial dilutions for tubes #3 through #8.
The concentration of Prostaglandin E_2 in tubes 1 through 8 will be 500, 250, 125, 62.5, 31.25, 15.625, 7.813 and 3.906 pg/mL.

Use all Standards within 2 hours of preparation.

	Std 1	Std 2	Std 3	Std 4	Std 5	Std 6	Std 7	Std 8
Assay Buffer (µL)	**390**	200	200	200	200	200	200	200
Addition	Stock	Std 1	Std 2	Std 3	Std 4	Std 5	Std 6	Std 7
Vol of Addition (µL)	**10**	200	200	200	200	200	200	200
Final Conc (pg/mL)	500	250	125	62.5	31.25	15.625	7.813	3.906

12.2 Assay Protocol - Regular Format

1. Use the plate layout sheet on the back page to aid in proper sample and standard identification. Determine the number of wells to be used and return unused wells to the foil pouch with desiccant. Seal the ziploc plate bag and store at 4 °C.
2. Pipet 50 µL of samples or standards into wells in the plate.
3. Pipet 75 µL of Assay Buffer into the non-specific binding (NSB) wells.
4. Pipet 50 µL of Assay Buffer into wells to act as maximum binding wells (B0 or 0 pg/mL).
5. Add 25 µL of the Prostaglandin E_2 Conjugate to each well using a repeater pipet.
6. Add 25 µL of the Prostaglandin E_2 Antibody to each well, **except the NSB wells**, using a repeater pipet.
7. Gently tap the sides of the plate to ensure adequate mixing of the reagents. Cover the plate with the plate sealer
8. **INCUBATION OPTIONS**
   either:
   8.a. Shake at room temperature for 2 hours.
   If the plate is not shaken signals bound will be approximately 40% lower.

   _or:_
   8.b. Shake the plate in a plate shaker at room temperature for 15 minutes to ensure adequate mixing of the reagents. Incubate at 4 °C for 16 - 18 hours.
9. If using Option 8b., the following day remove the TMB Substrate from the refrigerator and allow to come to room temperature for at least 30 minutes. **Addition of cold Substrate will cause depressed signal.**
10. Aspirate the plate and wash each well 4 times with 300 µL wash buffer. Tap the plate dry on clean absorbent towels.
11. Add 100 µL of TMB Substrate to each well, using a repeater pipet.
12. Incubate the plate at room temperature for 30 minutes without shaking.
13. Add 50 µL of the Stop Solution to each well, using a repeater pipet.
14. Read the optical density generated from each well in a plate reader capable of reading at 450 nm.
15. Use the plate reader's built-in 4PLC software capabilities to calculate prostaglandin E_2 concentration for each sample.

13 LOW SAMPLE VOLUME FORMAT

13.1 Standard Preparation - Low Sample Volume Format

Label test tubes as #1 through #7.
Pipet 380 µL of Assay Buffer into tube #1 and 200 µL into tubes #2 to #7.
The Prostaglandin E_2 stock solution contains an organic solvent. Prerinse the pipet tip several times to ensure accurate delivery.
Carefully add 20 µL of the Prostaglandin E_2 stock solution to tube #1 and vortex completely.
Take 200 µL of the Prostaglandin E_2 solution in tube #1 and add it to tube #2 and vortex completely.
Repeat the serial dilutions for tubes #3 through #7.
The concentration of Prostaglandin E_2 in tubes 1 through 7 will be 1,000, 500, 250, 125, 62.5, 31.25, and 15.625 pg/mL.

Use all Standards within 2 hours of preparation.

	Std 1	Std 2	Std 3	Std 4	Std 5	Std 6	Std 7
Assay Buffer (µL)	**380**	200	200	200	200	200	200
Addition	Stock	Std 1	Std 2	Std 3	Std 4	Std 5	Std 6
Vol of Addition (µL)	**20**	200	200	200	200	200	200
Final Conc (pg/mL)	1,000	500	250	125	62.5	31.25	15.625

13.2 Assay Protocol - Low Sample Volume Format

1. Use the plate layout sheet on the back page to aid in proper sample and standard identification. Determine the number of wells to be used and return unused wells to the foil pouch with desiccant. Seal the ziploc plate bag and store at 4 °C.
2. Pipet 25 µL of samples or standards into wells in the plate.
3. Pipet 50 µL of Assay Buffer into the non-specific binding (NSB) wells.
4. Pipet 25 µL of Assay Buffer into wells to act as maximum binding wells (B0 or 0 pg/mL).
5. Add 25 µL of the Prostaglandin E_2 Conjugate to each well using a repeater pipet.
6. Add 25 µL of the Prostaglandin E_2 Antibody to each well, **except the NSB wells**, using a repeater pipet.
7. Gently tap the sides of the plate to ensure adequate mixing of the reagents. Cover the plate with the plate sealer
8. **INCUBATION OPTIONS**
   either:
   8.a. Shake at room temperature for 2 hours.
   If the plate is not shaken signals bound will be approximately 40% lower.

   _or:_
   8.b. Shake the plate in a plate shaker at room temperature for 15 minutes to ensure adequate mixing of the reagents. Incubate at 4 °C for 16 - 18 hours.
9. If using Option 8b., the following day remove the TMB Substrate from the refrigerator and allow to come to room temperature for at least 30 minutes. **Addition of cold Substrate will cause depressed signal.**
10. Aspirate the plate and wash each well 4 times with 300 µL wash buffer. Tap the plate dry on clean absorbent towels.
11. Add 100 µL of TMB Substrate to each well, using a repeater pipet.
12. Incubate the plate at room temperature for 30 minutes without shaking.
13. Add 50 µL of the Stop Solution to each well, using a repeater pipet.
14. Read the optical density generated from each well in a plate reader capable of reading at 450 nm.
15. Use the plate reader's built-in 4PLC software capabilities to calculate prostaglandin E_2 concentration for each sample.

14 HIGH SENSITIVITY FORMAT

14.1 Standard Preparation - High Sensitivity Format

Label test tubes as #1 through #9.
Pipet 585 µL of Assay Buffer into tube #1 and 300 µL into tubes #2 to #9. **The Prostaglandin E_2 stock solution contains an organic solvent. Prerinse the pipet tip several times to ensure accurate delivery.**
Carefully add 15 µL of the Prostaglandin E_2 stock solution to tube #1 and vortex completely.
Take 300 µL of the Prostaglandin E_2 solution in tube #1 and add it to tube #2 and vortex completely. Repeat the serial dilutions for tubes #3 through #9.
The concentration of Prostaglandin E_2 in tubes 1 through 9 will be 500, 250, 125, 62.5, 31.25, 15.625, 7.813, 3.906, and 1.953 pg/mL.

Use all Standards within 2 hours of preparation.

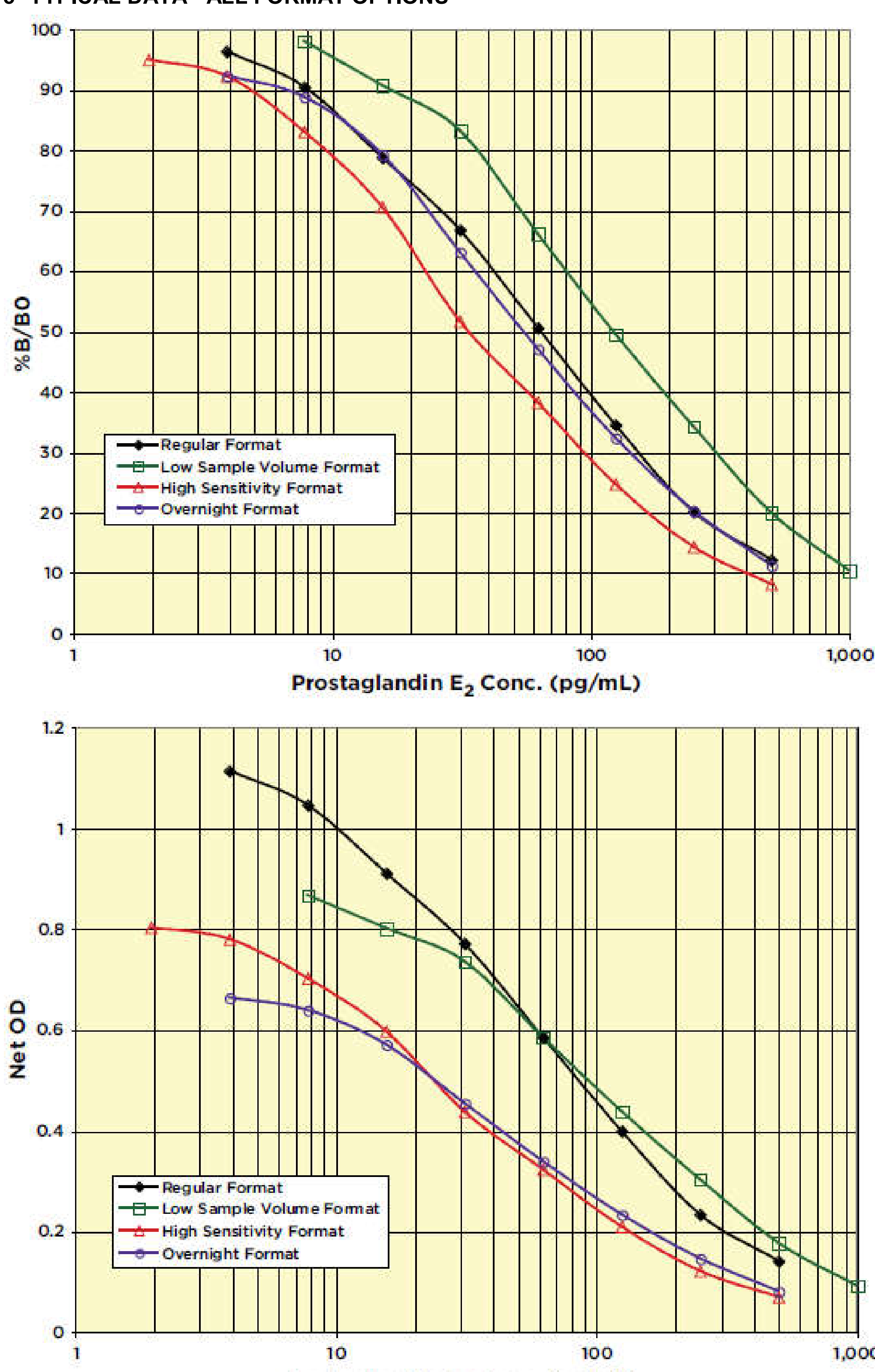
	Std 1	**Std 2**	**Std 3**	**Std 4**	**Std 5**	**Std 6**	**Std 7**	**Std 8**	**Std 9**
Assay Buffer (µL)	**585**	300	300	300	300	300	300	300	300
Addition	Stock	Std 1	Std 2	Std 3	Std 4	Std 5	Std 6	Std 7	Std 8
Vol of Addition (µL)	**15**	300	300	300	300	300	300	300	300
Final Conc (pg/mL)	500	250	125	62.5	31.25	15.625	7.813	3.906	1.953

14.2 Assay Protocol - High Sensitivity Format

1. Use the plate layout sheet on the back page to aid in proper sample and standard identification. Determine the number of wells to be used and return unused wells to the foil pouch with desiccant. Seal the ziploc plate bag and store at 4 °C.
2. Pipet 100 µL of samples or standards into wells in the plate.
3. Pipet 125 µL of Assay Buffer into the non-specific binding (NSB) wells.
4. Pipet 100 µL of Assay Buffer into wells to act as maximum binding wells (B0 or 0 pg/mL).
5. Add 25 µL of the Prostaglandin E_2 Conjugate to each well using a repeater pipet.
6. Add 25 µL of the Prostaglandin E_2 Antibody to each well, **except the NSB wells**, using a repeater pipet.
7. Gently tap the sides of the plate to ensure adequate mixing of the reagents. Cover the plate with the plate sealer
8. **INCUBATION OPTIONS**
   <u>either:</u>
   8.a. Shake at room temperature for 2 hours.
   If the plate is not shaken signals bound will be approximately 40% lower.

   <u>or:</u>
   8.b. Shake the plate in a plate shaker at room temperature for 15 minutes to ensure adequate mixing of the reagents. Incubate at 4 °C for 16 - 18 hours.
9. If using Option 8b., the following day remove the TMB Substrate from the refrigerator and allow to come to room temperature for at least 30 minutes. **Addition of cold Substrate will cause depressed signal.**
10. Aspirate the plate and wash each well 4 times with 300 µL wash buffer. Tap the plate dry on clean absorbent towels.
11. Add 100 µL of TMB Substrate to each well, using a repeater pipet.
12. Incubate the plate at room temperature for 30 minutes without shaking.
13. Add 50 µL of the Stop Solution to each well, using a repeater pipet.
14. Read the optical density generated from each well in a plate reader capable of reading at 450 nm.
15. Use the plate reader's built-in 4PLC software capabilities to calculate prostaglandin E_2 concentration for each sample.

15 TYPICAL DATA - ALL FORMAT OPTIONS



Overnight Data is from the <u>Regular Format</u>.

16 CALCULATION OF RESULTS

Average the duplicate OD readings for each standard and sample. Create a standard curve by reducing the data using the 4PLC fitting routine on the plate reader, after subtracting the mean OD's for the NSB. The sample concentrations obtained, calculated from the %B/B0 curve, should be multiplied by the dilution factor to obtain neat sample values.

Typical Data - *2 Hour Regular Format*

Sample	Mean OD	Net OD	% B/B0	PGE_2 Conc. (pg/mL)
NSB	0.061	0	-	-
Standard 1	0.202	0.141	12.19	500
Standard 2	0.294	0.233	20.14	250
Standard 3	0.460	0.399	34.49	125
Standard 4	0.646	0.585	50.56	62.5
Standard 5	0.833	0.772	66.72	31.25
Standard 6	0.972	0.911	78.74	15.625
Standard 7	1.107	1.046	90.41	7.813
Standard 8	1.175	1.114	96.28	3.906
B0	1.218	1.157	100	0
Sample 1	0.464	0.403	34.83	121.8
Sample 2	1.030	0.969	83.75	12.28

Always run your own standard curve for calculation of results. Do not use this data.

Conversion Factor: 100 pg/mL of prostaglandin E_2 is equivalent to 283.7 pM.

Typical Standard Curve - *2 Hour Regular Format*

Always run your own standard curve for calculation of results. Do not use this data.

17 VALIDATION DATA

Generated in 2 Hour Regular Format

17.1 Sensitivity and Limit of Detection

Sensitivity was calculated by comparing the OD's for nineteen wells run for each of the B0 and standard #8. The detection limit was determined at two (2) standard deviations from the B0 along the standard curve.

Sensitivity was determined as 3.07 pg/mL.

The Limit of Detection for the assay was determined in a similar manner by comparing the OD's for twenty runs for each of the zero standard and a low concentration human sample.

Limit of Detection was determined as 3.25 pg/mL.

We expect the *High Sensitivity Format* to give enhanced Sensitivity and LoD.

17.2 Linearity

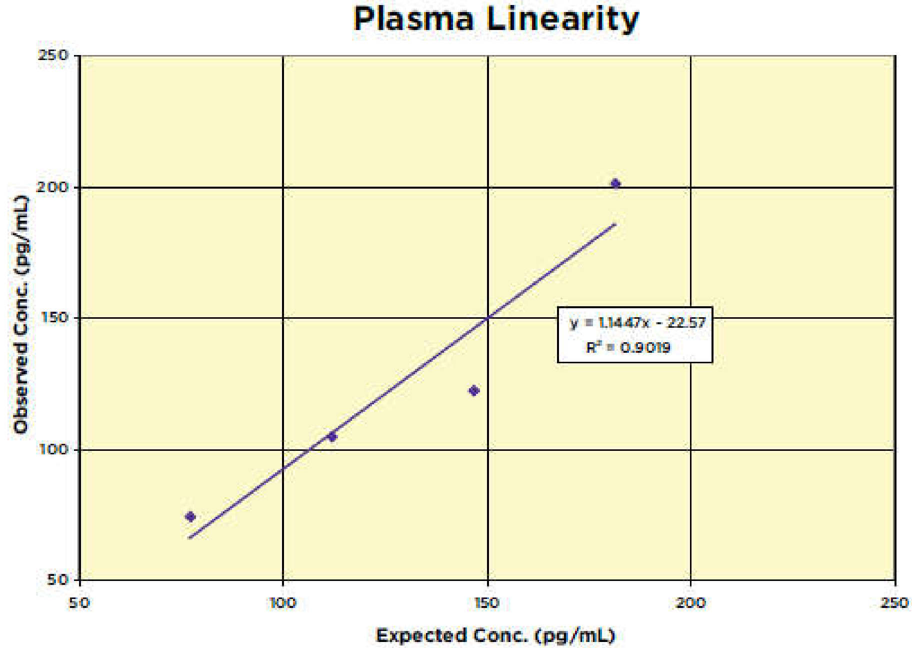
Linearity was determined in human plasma and urine samples by taking two diluted samples with known PGE_2 concentrations. A plasma sample with a high PGE_2 concentration of 216.4 pg/mL was mixed with one with a lower value of 42.5 pg/mL. A urine sample with a high PGE_2 concentration of 32.6 pg/mL was mixed with one with a lower value of 8.6 pg/mL. They were mixed in the ratios given below. The measured concentrations were compared to the expected values based on the ratios used.

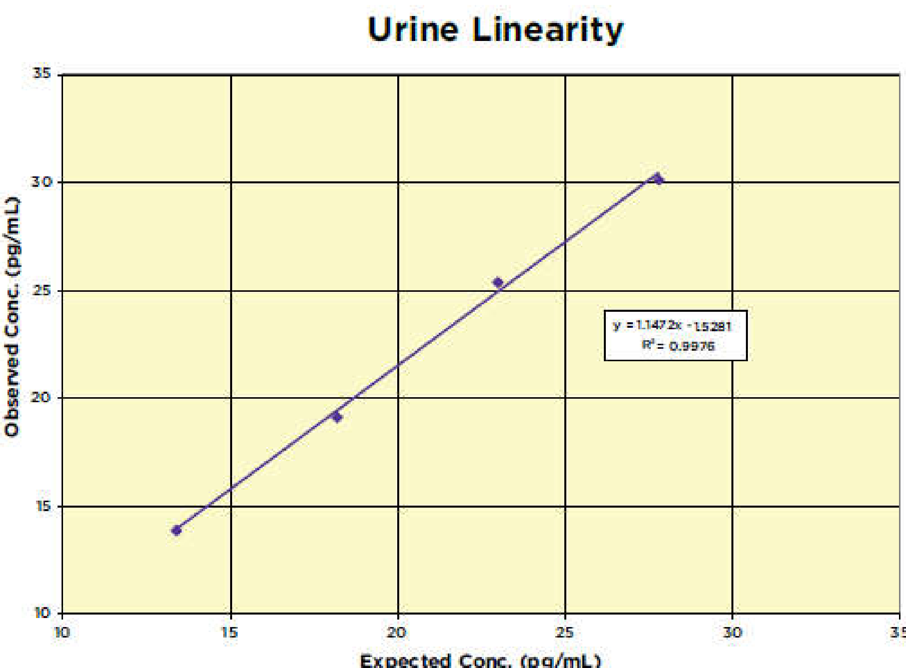
Plasma Linearity

High Sample	Low Sample	Observed Conc.	Expected Conc.	% Recovery
80%	20%	201.1	181.6	110.8%
60%	40%	122.3	146.8	83.3%
40%	60%	104.7	112.0	93.5%
20%	80%	74.3	77.3	96.1%
			Mean Recovery	**95.9%**

Urine Linearity

High Sample	Low Sample	Observed Conc.	Expected Conc.	% Recovery
80%	20%	30.1	27.8	108.3
60%	40%	25.4	23.0	110.2
40%	60%	19.1	18.2	105.0
20%	80%	13.9	13.4	103.3
			Mean Recovery	**106.7%**





17.3 Intra Assay Precision

Three human samples were diluted with Assay Buffer and run in replicates of 20 in an assay. The mean and precision of the calculated Prostaglandin E_2 concentrations were:

Sample	Prostaglandin E_2 Conc. (pg/mL)	%CV
1	11.7	12.3
2	98.6	6.3
3	131.2	4.9

17.4 Inter Assay Precision

Three human samples were diluted with Assay Buffer and run in duplicates in seventeen assays run over multiple days by four operators. The mean and precision of the calculated Prostaglandin E_2 concentrations were:

Sample	Prostaglandin E_2 Conc. (pg/mL)	%CV
1	12.3	8.8
2	100.5	8.1
3	134.7	9.8

17.5 Sample Values

Eight human serum samples that did not contain COX inhibitors were tested in the assay.

Neat sample were diluted 1:20-1:50 in Assay Buffer and adjusted values ranged from 652 to 4,170 pg/mL with an average of 2,126 pg/mL.

Ten human plasma samples that did not contain COX inhibitors were tested in the assay.

Neat sample were diluted 1:20-1:50 in Assay Buffer and adjusted values ranged from 219 to 4,328 pg/mL with an average of 1,717 pg/mL.

Eight normal human urine samples were diluted 1:10- 1:20 in Assay Buffer and adjusted values ranged from 56.9 to 326 pg/mL with an average of 149.9 pg/mL

17.6 Cross Reactivity

The following cross reactants were tested in the assay and calculated at the 50% binding point.

Eicosanoid	Cross Reactivity (%)
Prostaglandin E_2	100%
Prostaglandin E_1	27.28%
Prostaglandin $F_{2\alpha}$	0.33%
Thromboxane B_2	< 0.02%
6-keto-Prostaglandin $F_{1\alpha}$	< 0.02%
15-keto-Prostaglandin E_1	< 0.02%
16,16-dimethyl-Prostaglandin E_2	< 0.02%
Arachidonic Acid	< 0.02%

17.7 Interferents

A variety of solvents were tested as possible interfering substances in the assay. Organic solvents such as DMSO, Dimethylformamide (DMF), methanol and ethanol were tested in the assay at 0.1%. DMSO and DMF caused a 1.2% and 0.8% decrease in measured PGE_2 levels, whereas methanol and ethanol caused an increase of 2.5% and 4.6% in measured PGE_2 levels. A solvent only control should be run by the end user when appropriate.

Hemoglobin at 0.02 mg/dL caused a 1% decrease in measured PGE_2 levels.

Elevated lipids will also interfere with the measurement of PGE_2. Follow the extraction recommendations described in chapter 10 SAMPLE PREPARATION.

LIMITED WARRANTY

DRG warrants that at the time of shipment this product is free from defects in materials and workmanship. This warranty is in lieu of any other warranty expressed or implied, including but not limited to, any implied warranty of merchantability or fitness for a particular purpose.

18 PLATE LAYOUT SHEET

	1	2	3	4	5	6	7	8	9	10	11	12
A												
B												
C												
D												
E												
F												
G												
H												

SYMBOLS USED

Symbol	English	Deutsch	Français	Español	Italiano
	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-Konformitäts-kennzeichnung	Conformité aux normes européennes	Conformidad europea	Conformità europea
IVD	In vitro diagnostic device	In-vitro-Diagnostikum	Usage 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
	Contains sufficient for <n> tests/	Ausreichend für "n" Ansätze	Contenu suffisant pour "n" tests	Contenido suficiente para <n> ensayos	Contenuto sufficiente per "n" saggi
	Storage Temperature	Lagerungstemperatur	Température de conservation	Temperatura de conservación	Temperatura di conservazione
	Expiration Date	Mindesthaltbarkeits-datum	Date limite d'utilisation	Fecha de caducidad	Data di scadenza
	Manufacturer	Hersteller	Fabricant	Fabricante	Fabbricante
Distributed by	Distributor	Vertreiber	Distributeur	Distribuidor	Distributore
Content	Content	Inhalt	Conditionnement	Contenido	Contenuto
Volume/No.	Volume / No.	Volumen/Anzahl	Volume/Quantité	Volumen/Número	Volume/Quantità