Product Manual

Direct 8-iso-PGF$_{2\alpha}$ ELISA kit

Catalog #: ADI-901-091

480 Well (5 by 96 well) Kit
For use with serum, plasma, and tissue.

NOTE: This version corrects an error in the “Reagent Preparation” section for the Sample Diluent.

For the latest product information, including support documentation, visit us online:
www.enzolifesciences.com

Technical Support (US): 800-942-0430
Technical Support (EU): +41 61 926 8989

USE FOR RESEARCH PURPOSES ONLY

Unless otherwise specified expressly on the packaging, all products sold hereunder are intended for and may be used for research use only. Not for use in diagnostic procedures. Purchase does not include any right or license to use, develop or otherwise exploit these products commercially. Any commercial use, development or exploitation of these products or development using these products without the express written authorization of Enzo Life Sciences, Inc. is strictly prohibited. Buyer assumes all risk and liability for the use and/or results obtained by the use of the products covered by this invoice whether used singularly or in combination with other products.

LIMITED WARRANTY; DISCLAIMER OF WARRANTIES

These products are offered under a limited warranty. The products are guaranteed to meet all appropriate specifications described in the package insert at the time of shipment. Enzo Life Sciences' sole obligation is to replace the product to the extent of the purchasing price. All claims must be made to Enzo Life Sciences, Inc., within five (5) days of receipt of order. THIS WARRANTY IS EXPRESSLY IN LIEU OF ANY OTHER WARRANTIES OR LIABILITIES, EXPRESS OR IMPLIED, INCLUDING WARRANTIES OF MERCHANTABILITY, FITNESS FOR A PARTICULAR PURPOSE, AND NONINFRINGEMENT OF THE PATENT OR OTHER INTELLECTUAL PROPERTY RIGHTS OF OTHERS, AND ALL SUCH WARRANTIES (AND ANY OTHER WARRANTIES IMPLIED BY LAW) ARE EXPRESSLY DISCLAIMED.

TRADEMARKS AND PATENTS

Several Enzo Life Sciences products and product applications are covered by US and foreign patents and patents pending.

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

Please read entire booklet before proceeding with the assay.

Carefully note the handling and storage conditions of each kit component.

Please contact Enzo Life Sciences Technical Support if necessary.

Table of Contents

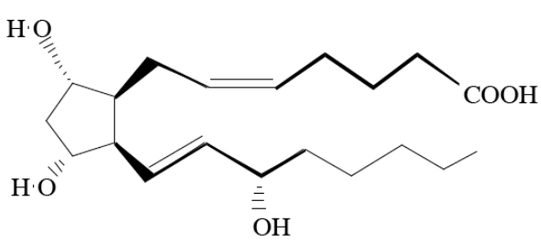
DESCRIPTION

The Direct 8-iso-$PGF_{2\alpha}$ ELISA kit is a competitive immunoassay for the quantitative determination of 8-iso-Prostaglandin $F_{2\alpha}$ in biological fluids. Please read the complete kit insert before performing this assay. The kit uses a polyclonal antibody to 8-iso-$PGF_{2\alpha}$ to bind, in a competitive manner, the 8-iso-$PGF_{2\alpha}$ in the sample or an alkaline phosphatase molecule which has 8-iso-$PGF_{2\alpha}$ covalently attached to it. After a simultaneous incubation at either room temperature or 4°C, the excess reagents are washed away and substrate is added. After a short incubation time the enzyme reaction is stopped and the yellow color generated read on a microplate reader at 405nm. The intensity of the bound yellow color is inversely proportional to the concentration of 8-iso-$PGF_{2\alpha}$ in either standards or samples. The measured optical density is used to calculate the concentration of 8-iso-$PGF_{2\alpha}$. For further explanation of the principles and practice of immunoassays please see the excellent books by Chard[1] or Tijssen[2].

INTRODUCTION

The 8-epimer of Prostaglandin $F_{2\alpha}$ (8-iso-$PGF_{2\alpha}$) is produced in vivo by both non-cyclooxygenase and cyclooxygenase dependent mechanisms from arachidonic acid[3-5]. 8-iso-$PGF_{2\alpha}$ has been shown to be a potent vasoconstrictor[5,6], a potential mediator of hepatorenal syndrome and atherosclerosis[5] and a mutagen in 3T3 cells and in vascular smooth muscle cells[5,6,7]. It has also been postulated to participate as a pathophysiological mediator and is able to modify the fluidity and integrity of membranes[7]. 8-iso-$PGF_{2\alpha}$ has been shown to circulate in plasma and is excreted in urine[6]. Methods for assessing total 8-iso-$PGF_{2\alpha}$ typically require the alkaline hydrolysis of 8-iso-$PGF_{2\alpha}$ esters from tissues, followed by length procedures involving extractions, phase separations and thin layer chromatography.

8-iso-Prostaglandin $F_{2\alpha}$



The Direct 8-iso-PGF2α ELISA kit is compatible with 8-iso-PGF2α in serum, plasma, and tissue samples. Urine is not suitable for analysis in this kit and should be measured in the 8-iso-PGF2α ELISA Kit (catalog number ADI-900-010).

PRECAUTIONS

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

1. Some kit components contain azide, which may react with lead or copper plumbing. When disposing of reagents always flush with large volumes of water to prevent azide build-up.

Handle with care

2. Some solutions supplied in this kit are caustic: care should be taken in their use.
3. The activity of the alkaline phosphatase conjugate is dependent on the presence of Mg^{2+} and Zn^{2+} ions. The activity of the conjugate is affected by concentrations of chelators (>10 mM) such as EDTA and EGTA.
4. We test this kit's performance with a variety of samples, however it is possible that high levels of interfering substances may cause variation in assay results.
5. The 8-iso-Prostaglandin $F_{2\alpha}$ Standard provided, Catalog No. 80-0110, is supplied in ethanolic buffer at a pH optimized to maintain 8-iso-$PGF_{2\alpha}$ integrity. Care should be taken handling this material because of possible effects of prostaglandins.

Note: Protocol provided covers applications [1-2]. Assay set-up can be readily modified for alternative applications by inclusion, omission or substitution of specific components.

MATERIALS SUPPLIED

1. **Goat anti-Rabbit IgG Microtiter Plate, Five Plates of 96 Wells; Catalog No. 80-0060:**

   Plates using break-apart strips coated with goat antibody specific to rabbit IgG.

2. **8-iso-PGF$_{2\alpha}$ Conjugate, 27ml; Catalog No. 80-0119:**

   A blue solution of alkaline phosphatase conjugated with 8-iso-PGF$_{2\alpha}$.

3. **Direct 8-iso-PGF$_{2\alpha}$ ELISA Antibody, 27ml; Catalog No. 80-1173:**

   A yellow solution of a rabbit polyclonal antibody to 8-iso-PGF$_{2\alpha}$.

4. **Direct 8-iso-PGF$_{2\alpha}$ Sample Diluent Concentrate, 125ml; Catalog No. 80-0843:**

   A proprietary solution containing sodium azide as a preservative. See page 8 for preparation before use.

5. **Neutralizing Reagent, 30ml**

   Catalog No. 80-1476.

6. **Wash Buffer Concentrate, 105ml; Catalog No. 80-1287:**

   Tris buffered saline containing detergents.

7. **8-iso-Prostaglandin F2α Standard, 3 x 0.5ml; Catalog No. 80-0110;**
   A 1,000,000 pg/ml solution of 8-iso-PGF$_{2\alpha}$ per vial.

8. **pNpp Substrate, 105ml; Catalog No. 80-0076:**

   A solution of p-nitrophenyl phosphate in buffer. Ready to use.

9. **Stop Solution, 32ml; Catalog No. 80-0248:**

   A solution of trisodium phosphate in water. Keep tightly capped. Caution: Caustic.

10. **Direct 8-iso-PGF2α Assay Layout Sheet, 1 each; Catalog No. 30-0149**

11. **Plate Sealers, 5 each**

Store kit at 4°C.

STORAGE

All components of this kit are stable at 4°C until the kit's expiration date.

OTHER MATERIALS NEEDED

1. Deionized or distilled water.
2. 10N NaOH, for serum and plasma, see page 6.
3. Concentrated HCl (12.1N) for serum and plasma see page 6.
4. 2N NaOH, for tissue, see page 6.
5. 2N HCl, for tissue, see page 6.
6. Precision pipets for volumes between 5µl and 1,000µl.
7. Repeater pipets for dispensing 50µl and 200µl.
8. A disposable beaker for diluting buffer concentrates.
9. Graduated cylinders.
10. 45°C water bath or incubator.
11. Microcentrifuge.
12. A microplate shaker.
13. Adsorbent paper for blotting.
14. Microplate reader capable of reading at 405nm, preferably with correction between 570 and 590nm.

SAMPLE HANDLING

Hydrolysis of lipoprotein or phospholipid coupled 8-iso-Prostaglandin $F_{2\alpha}$ (8-iso-$PGF_{2\alpha}$) is required to ensure that the measured 8-iso-$PGF_{2\alpha}$ is a true reflection of both free and esterified isoprostane. To hydrolyze this ester bond, the sample is treated with NaOH at 45°C for 2 hours. Dilution of the liberated 8-iso-$PGF_{2\alpha}$ is minimized in liquid samples (e.g., serum and plasma) when 4 parts of sample are treated with 1 part of 10N NaOH. Tissue samples are hydrolyzed in an excess of 2N NaOH. Samples are then neutralized. See below for full details. **It is important that all standards and diluted samples be in the same matrix. All dilutions of samples and standards must be made with the Direct 8-iso Sample Diluent solution provided.**

Please note that the hydrolysis step will destroy some of the liberated 8-iso-$PGF_{2\alpha}$ due to the effects of strong base on the isoprostane structure. However, the relative change in 8-iso-$PGF_{2\alpha}$ should be identical from sample to sample. If you wish to determine the percentage of endogenous 8-iso-$PGF_{2\alpha}$ destroyed during hydrolysis, we suggest adding a known amount of 8-iso-$PGF_{2\alpha}$ to a sample prior to hydrolysis and determine the percent recovery of added 8-iso-$PGF_{2\alpha}$.

Tissue Samples

Prior to hydrolysis, samples should be stored at -20℃ or lower. Tissue samples should be powdered prior to hydrolysis. Homogenization or other methods of cell disruption may be used. An appropriate excess volume of 2N NaOH should be used. We recommend from 10 µg to 1 mg of tissue perml of 2N NaOH. Samples in 2N NaOH should be covered and heated at 45℃ for 2 hours to ensure hydrolysis. After hydrolysis, the samples should be cooled and neutralized with an equal volume of 2N HCl. For example, if 2 mg of tissue are hydrolyzed in 2ml of 2N NaOH, 2ml of 2N HCl would be added after the hydrolysis step. Centrifuge the neutralized samples at 3,000 rpm in a microcentrifuge. If necessary, check the pH of the neutralized samples. The pH should be in the range of 6-8. If it is not, adjust the pH to this range. The clear supernatant can be used in the assay or stored at ≤ -20℃ for future use. **Prior to analysis, dilutions are made with the Direct 8-iso Sample Diluent.**

Serum, EDTA or heparinized Plasma

Samples should be kept frozen at -20℃ or lower. Use 1 part of 10N NaOH for every 4 parts of liquid sample. Cap and heat the sample at 45℃ for 2 hours. Cool, then adjust the pH of the sample by adding 100µl of concentrated (12.1N) HCl per 500µl of hydrolyzed sample. The sample should appear milky after this

addition. Centrifuge the samples for 5 minutes at 14,000 rpm in a microcentrifuge. The appearance and removal of a white precipitate in the pellet is expected and necessary. The clear supernatant can be used in the assay or stored at ≤ -20°C for future use. If necessary check the pH of the neutralized samples. The pH should be in the range of 6-8. If it is not, adjust the pH to this range. **Consult the Sample Recovery section on page 16 for the appropriate dilution of the hydrolyzed, neutralized sample with the Direct 8-iso Sample Diluent solution prior to analysis.**

Remember to correct any measured 8-iso-PGF2α concentrations for the dilution of the original sample by addition of added base and the HCl used for neutralization. Based on this protocol, the total dilution for your liquid samples would be 1:1.5. However the end-user must verify that the recommended dilutions are appropriate for their samples.

PROCEDURAL NOTES

1. Do not mix components from different kit lots or use reagents beyond the kit expiration date.
2. Allow all reagents to warm to room temperature for at least 30 minutes before opening.
3. Standards can be made up in either glass or plastic tubes.
4. Pre-rinse the pipet tip with reagent, use fresh pipet tips for each sample, standard and reagent.
5. Pipet standards and samples to the bottom of the wells.
6. Add the reagents to the side of the well to avoid contamination.
7. This kit uses break-apart microtiter strips, which allow the user to measure as many samples as desired. Unused wells must be kept desiccated at 4°C in the sealed bag provided. The wells should be used in the frame provided.
8. Care must be taken to minimize contamination by endogenous alkaline phosphatase. Contaminating alkaline phosphatase activity, especially in the substrate solution, may lead to high blanks. Care should be taken not to touch pipet tips and other items that are used in the assay with bare hands.
9. Prior to addition of substrate, ensure that there is no residual wash buffer in the wells. Any remaining wash buffer may cause variation in assay results.

REAGENT PREPARATION

1. **Direct 8-iso-PGF2α Sample Diluent**
   Prepare the Direct 8-iso-PGF2α Sample Diluent by diluting the supplied concentrate with an equal volume of deionized water. This can be stored at room temperature until the kit's expiration.

2. **8-iso-PGF2α Standard**

   Allow the 1,000,000 pg/ml 8-iso-PGF2α standard solution to warm to room temperature. Label five 12 x 75 mm glass tubes #1 through #5. Pipet 900 µL of Direct 8-iso Sample Diluent into tube #1. Pipet 800 µL of Direct 8-iso Sample Diluent into tubes #2 through #5. Add 100 µL of the 1,000,000 pg/ml standard to tube #1. Vortex thoroughly. Add 200 µL of tube #1 to tube #2 and vortex thoroughly. Add 200 µL of tube #2 to tube #3 and vortex. Continue this for tubes #4 through #5.

   The concentration of 8-iso-PGF2α in tubes #1 through #5 will be 100,000, 20,000, 4,000, 800 and 160 pg/ml respectively. See the Direct 8-iso-PGF2α Assay Layout Sheet for dilution details.

   Diluted standards should be used within 60 minutes of preparation.

3. **Conjugate 1:10 Dilution for Total Activity Measurement**

   Prepare the conjugate 1:10 Dilution by diluting 50µL of the supplied conjugate with 450µL of the Sample Diluent. The dilution should be used within 3 hours of preparation. **This 1:10 dilution is intended for use in the Total Activity Wells ONLY.**

4. **Wash Buffer**

   Prepare Wash Buffer by diluting 5ml of the supplied concentrate with 95ml of deionized water. This can be stored at room temperature until the kit expiration or for 3 months, whichever is earlier.

ASSAY PROCEDURE

Bring all reagents to room temperature for at least 30 minutes prior to opening. All standards and samples should be run in duplicate.

1. Refer to the Assay Layout Sheet to determine the number of wells to be used and put any remaining wells with the desiccant back into the pouch and seal the ziploc. Store unused wells at 4°C.
2. Pipet 50µL of Neutralizing Reagent into all wells, except the Total Activity (TA) and Blank wells.
3. Pipet 50µL of Direct 8-iso Sample Diluent into the NSB and the Bo (0 pg/ml Standard) wells.
4. Pipet 50µL of Standards #1 through #5 into the appropriate wells.
5. Pipet 50µL of the hydrolyzed/neutralized Samples into the appropriate wells.
6. Pipet 50µL of Direct 8-iso Sample Diluent into the NSB wells.
7. Pipet 50µL of blue Conjugate into each well, except the Total Activity (TA) and Blank wells.
8. Pipet 50µL of yellow Antibody into each well, except the Blank, TA and NSB wells.

NOTE: Every well used should be **Green** in color except the NSB wells which should be **Blue**. The Blank and TA wells are empty at this point and have no color.

9. Incubate the plate at room temperature on a plate shaker for 2 hours at ~500 rpm* or 18-24 hours at 4°C. The plate may be covered with the plate sealer provided, if so desired.
10. Empty the contents of the wells and wash by adding 400µL of wash solution to every well. Repeat the wash 2 more times for a total of 3 washes.
11. After the final wash, empty or aspirate the wells, and firmly tap the plate on a lint free paper towel to remove any remaining wash buffer.
12. Add 5µL of the light blue Conjugate 1:10 Dilution (see Step 2, Reagent Preparation, on page 8) to the TA wells.
13. Add 200µL of the pNpp Substrate solution to every well. Incubate at room temperature for 45 minutes without shaking.
14. Add 50µL of Stop Solution to every well. This stops the reaction and the plate should be read immediately.

15. Blank the plate reader against the Blank wells, read the optical density at 405nm, preferably with correction between 570 and 590nm. If the plate reader is not able to be blanked against the Blank wells, manually subtract the mean optical density of the Blank wells from all readings.

    * The plate shaker speed was based on a BellCo Mini Orbital Shaker (model no. 7744-08096). The actual speed of the plate shaker should be such that the liquid in the plate wells mixes thoroughly, but does not splash out of the well.

CALCULATION OF RESULTS

Several options are available for the calculation of the concentration of 8-iso-PGF2α in the samples. We recommend that the data be handled by an immunoassay software package utilizing a four parameter logistic curve fitting program (such as AssayBlaster™, ENZO catalog number ADI-28-0002). Such software is often supplied by plate reader manufacturers. If data reduction software is not readily available, the concentration of 8-iso-$PGF_{2\alpha}$ can be calculated as follows:

1. Calculate the average Net Optical Density (OD) bound for each standard and sample by subtracting the average NSB OD from the average OD bound:

   Average Net OD = Average Bound OD - Average NSB OD

2. Calculate the binding of each pair of standard wells as a percentage of the maximum binding wells (Bo), using the following formula:

   $$\text{Percent Bound} = \frac{\text{Net OD}}{\text{Net Bo OD}} \times 100$$

3. Approximate a straight line through the points. The concentration of 8-iso-$PGF_{2\alpha}$ in the unknowns can be determined by interpolation.

Remember to correct any measured 8-iso-$PGF_{2\alpha}$ concentrations for the dilution of the original sample by addition of added acid/base used for neutralization.

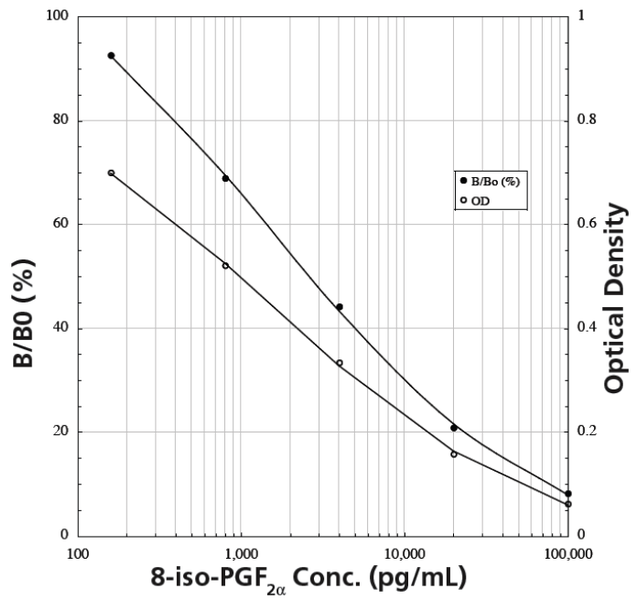
TYPICAL RESULTS

The results shown below are for illustration only and should not be used to calculate results.

	2 Hour Format			Overnight Format		
Sample	**Average Net OD**	**Percent Bound**	**8-iso-$PGF_{2\alpha}$ (pg/ml)**	**Average Net OD**	**Percent Bound**	**8-iso-$PGF_{2\alpha}$ (pg/ml)**
TA	0.398			0.312		
NSB	0.001	0.00%		-0.003	0.00%	
Bo	0.755	100%	0	1.040	100%	
S1	0.062	8.2%	100,000	0.073	7.0%	100,000
S2	0.157	20.8%	20,000	0.174	16.7%	20,000
S3	0.334	44.2%	4,000	0.388	37.3%	4,000
S4	0.521	69.0%	800	0.667	64.1%	800
S5	0.700	92.7%	160	0.921	88.6%	160
Unknown 1	0.238	31.5%	8,949	0.282	27.1%	8,005
Unknown 2	0.563	74.6%	1,245	0.661	63.6%	836
TA	0.398			0.312		

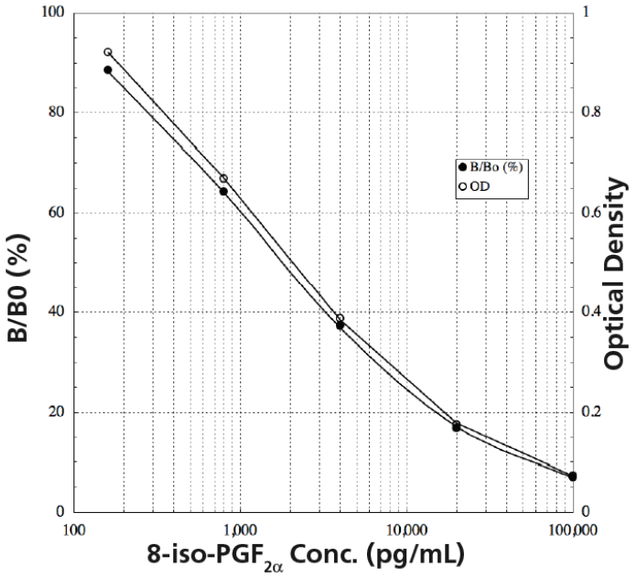
TYPICAL STANDARD CURVES

Typical standard curves are shown below. These curves must not be used to calculate 8-iso-PGF$_{2\alpha}$ concentrations; each user must run a standard curve for each assay.



Typical Quality Control Parameters

Total Activity Added	=	0.398 x 10 x 10	=	39.8
%NSB	=	0.0%		
%Bo/TA	=	19.0%		
Quality of Fit	=	1.00 (Calculated from 4 parameter logistic curve fit)		
20% Intercept	=	23,192 pg/ml		
50% Intercept	=	2,627 pg/ml		



TYPICAL QUALITY CONTROL PARAMETERS

Total Activity Added	=	0.312 x 10 x 10 =31.2
%NSB	=	0.0%
%Bo/TA	=	3.3%
Quality of Fit	=	1.00 (Calculated from 4 parameter logistic curve fit)
20% Intercept	=	14,802 pg/ml
50% Intercept	=	1,820 pg/ml

PERFORMANCE CHARACTERISTICS

The following parameters for this kit were determined using the guidelines listed in the National Committee for Clinical Laboratory Standards (NCCLS) Evaluation Protocols[8].

Sensitivity

Sensitivity was calculated by determining the average optical density bound for sixteen (16) wells run as Bo, and comparing to the average optical density for sixteen (16) wells run with Standard #5. The detection limit was determined as the concentration of 8-iso-$PGF_{2\alpha}$ measured at two (2) standard deviations from the zero along the standard curve.

2 HOUR FORMAT

Average Optical Density for the Bo = 0.341 ± 0.010 (2.9%)

Average Optical Density for Standard #5q= 0.310 ± 0.012 (4.0%)

Delta Optical Density (0-160 pg/ml) = 0.341 - 0.310= 0.031

2 SD's of the Zero Standard = 2 x 0.010 = 0.020

Sensitivity = $\frac{0.020}{0.031}$ x 160 pg/ml = **103.2 pg/ml**

OVERNIGHT FORMAT

Average Optical Density for the Bo = 0.484 ± 0.011 (1.8%)

Average Optical Density for Standard #5 = 0.396 ± 0.008 (1.5%)

Delta Optical Density (0-160 pg/ml) = 0.484 - 0.396 = 0.088

2 SD's of the Zero Standard = 2 x 0.011 = 0.022

Sensitivity = $\frac{0.022}{0.088}$ x 160 pg/ml = **40.0 pg/ml**

Linearity

A sample containing 50,000 pg/ml 8-iso-$PGF_{2\alpha}$ was diluted 7 times 1:2 in Direct 8-iso Sample Diluent and measured in the assay. The data was plotted graphically as actual 8-iso-$PGF_{2\alpha}$ concentration versus measured 8-iso-$PGF_{2\alpha}$ concentration.

The line obtained had a slope of 0.967 and a correlation coefficient of 0.986.

Precision

Intra-assay precision was determined by taking samples containing low, medium and high concentrations of 8-iso-$PGF_{2\alpha}$ and running these samples multiple times (n=16) in the same assay. Inter-assay precision was determined by measuring three samples with low, medium and high concentrations of 8-iso-$PGF_{2\alpha}$ in multiple assays (n=8).

The precision numbers listed below represent the percent coefficient of variation for the concentrations of 8-iso-$PGF_{2\alpha}$ determined in these assays as calculated by a 4 parameter logistic curve fitting program.

	8-iso-$PGF_{2\alpha}$ (pg/ml)	Intra-assay %CV	Inter-assay %CV
Low	970	11.3	
Medium	3,264	5.7	
High	5,092	5.1	
Low	1,473		5.4
Medium	4,501		5.8
High	6,679		10.4

Cross Reactivities

The cross reactivities for a number of related eicosanoid compounds was determined in the regular 8-iso-$PGF_{2\alpha}$ ELISA assay by dissolving the cross reactant (purity checked by N.M.R. and other analytical methods) in Assay Buffer at concentrations from 100,000 to 6 pg/ml. These samples were then measured in the assay, and the measured 8-iso-$PGF_{2\alpha}$ concentration at 50% B/Bo was calculated. The % cross reactivity was calculated by comparison with the actual concentration of cross reactant in the sample and expressed as a percentage.

Compound	Cross Reactivity
8-iso-$PGF_{2\alpha}$	100%
$PGF_{1\alpha}$	4.6%
$PGF_{2\alpha}$	1.85%
PGE_1	0.19%
TXB_2	0.023%
PGB_1	0.02%
PGE_3	0.012%
6-keto-$PGF_{1\alpha}$	0.008%
13,14-dihydro-15-keto-$PGF_{2\alpha}$	0.008%
6,15-keto-13,14-dihydro-$PGF_{1\alpha}$	0.005%
8-iso-PGE_1	<0.001%
PGA_2	<0.001%
PGJ_2	<0.001%
2-Arachidonoylglycerol	<0.001%
Anandamide	<0.001%

SAMPLE RECOVERIES

Please refer to pages 6-8 for Sample Handling recommendations, and preparation of Direct 8-iso-PGF2α Sample Diluent and Standard.

8-iso-PGF2α concentrations were measured in the samples listed below. 8-iso-PGF2α was spiked into the undiluted samples of these hydrolysed samples which were neutralized and diluted into the Direct 8-iso Sample Diluent and then assayed in the kit. The following results were obtained:

Sample	% Recovery*	Recommended Dilution*
Porcine Serum	93.8	None
Equine Heparinized Plasma	97.2	None
Porcine EDTA Plasma	109.8	1:8

* See Sample Handling instructions on page 6 for details.

Urine is not a suitable sample for analysis in this kit. Urine samples should be measured using the 8-iso PGF2α ELISA Kit, (Catalog Number ADI-900-010).

REFERENCES

1. T. Chard, “An Introduction to Radioimmunoassay & Related Techniques 4th Ed.”, (1990) Amsterdam: Elsevier.
2. P. Tijssen, “Practice & Theory of Enzyme-Linked Immunosorbent Assays”, (1985) Amsterdam: Elsevier.
3. J.D. Morrow, et al., Proc. Natl. Acad. Sci. USA, (1990) 87: 9383.
4. M. Banerjee, et al., Am. J. Physiol., (1992) 263: H660.
5. J.A. Lawson & K.M. Maxey, Cayman Currents, (1996) 2: 1&3.
6. D. Pratico, et al., J.Biol.Chem., (1995) 270: 9800.
7. T. Yura, et al., Adv. Prostag. Thromb. R., (1995) 23: 237.
8. National Committee for Clinical Laboratory Standards Evaluation Protocols, SC1, (1989) Villanova, PA: NCCLS.

NOTES

NOTES

NOTES

NOTES

GLOBAL HEADQUARTERS
Enzo Life Sciences Inc.
10 Executive Boulevard
Farmingdale, NY 11735
Toll-Free:1.800.942.0430
Phone:631.694.7070
Fax: 631.694.7501
info-usa@enzolifesciences.com

EUROPE
Enzo Life Sciences (ELS) AG
Industriestrasse 17
CH-4415 Lausen
Switzerland
Phone:+41/0 61 926 89 89
Fax:+41/0 61 926 89 79
info-ch@enzolifesciences.com

For local distributors and detailed product information visit us online:
www.enzolifesciences.com