Product Information

Prostaglandin E₂-MOX EIA Antiserum

Catalog No. 414732



Water used to prepare all EIA reagents and buffers must be deionized and free of trace organic contaminants ("UltraPure"). Use activated carbon filter cartridges or other organic scavengers. Glass distilled water (even if double distilled), HPLC-grade water, and sterile water (for injections) are not adequate for EIA. UltraPure water may also be purchased (Catalog No. 400000).

Laboratory Procedures

This vial contains lyophilized Prostaglandin E2-MOX EIA Antiserum (anti-PGE2-MOX rabbit IgG). For long term storage, we suggest that the antiserum be stored as supplied at -20°C; it will be stable for at least one year. Reconstitute 100 determination vials with 6 ml EIA Buffer or 500 determination vials with 30 ml EIA Buffer (see buffer preparation instructions below). Store the reconstituted antiserum at 4°C and use within four weeks. For your convenience, we have supplied a 20% surplus of antiserum.

Sample and Standard Preparation

This assay measures the methoximated derivative of PGE₂ (PGE₂-MOX). All standards and samples must be methoximated prior to performing the assay. See the methoximation protocol on the following page for details.

Buffer Preparation

1. Phosphate Buffer

Prepare a 1.0 M phosphate buffer solution by combining 133 g K₂HPO₄ and 32.15 g KH₂PO₄ and diluting to a total volume of 1.0 liter with UltraPure water. The pH of this solution will be 7.4.

2. EIA Buffer

Combine 100 ml of the phosphate buffer prepared above with 100 mg NaN₃, 23.4 g NaCl, 370 mg tetrasodium EDTA, and 1 g bovine serum albumin (Sigma A7030 or equivalent). Stir at room temperature until completely dissolved and dilute to a total volume of 1.0 liter with UltraPure water. Add 0.1 ml Tween 20 and mix thoroughly. This EIA Buffer may also be purchased from Cayman as a 10X concentrate buffer (Catalog No. 400060).

Combine 10 ml of the phosphate buffer prepared above with 0.5 ml Tween 20. Bring to a final volume of 1.0 liter with UltraPure water. This Wash Buffer may also be purchased from Cayman as a 400X concentrate buffer (Catalog

Specificity

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Prostaglanidn E ₃	82%	6-keto Prostaglandin F _{1α} -MOX	<0.01%
8-iso Prostaglandin E ₂ -MOX	21%	Thromboxane B ₂ -MOX	<0.01%
15-keto Prostaglandin E ₂ -MOX	7.8%	Prostaglandin D ₂	<0.01%
Prostaglandin E ₁ -MOX	5.4%	6-keto Prostaglandin F ₁ ,	<0.01%
Prostaglandin E ₂	0.6%	Thromboxane B ₂	<0.01%
Tetranor PGEM-MOX	0.06%	Tetranor PGEM	<0.01%
Prostaglandin D ₂ -MOX	<0.01%	Prostaglandin F _{2a}	<0.01%

When used in conjunction with the ACE $^{\text{\tiny{TM}}}$ reagents for the 17-phenyl trinor PGF $_{2\alpha}$ EIA kit, this antiserum exhibits a detection limit (80% B/B $_0$) of ~3 pg/ml and an IC $_{50}$ (50% B/B $_0$) of ~16 pg/ml for 17-phenyl trinor PGF $_{2\alpha}$.

WARNING: This product is for laboratory research only: not for administration to humans. Not for human or veterinary DIAGNOSTIC OR THERAPEUTIC USE.

This material should be considered hazardous until information to the contrary becomes available. Do not ingest, swallow, or inhale. Do not get in eyes, on skin, or on clothing. Wash thoroughly after handling. This information contains some, but not all, of the information required for the safe and proper use of this material. Before use, the user must review the complete Material Safety Data Sheet, which has been sent under separate cover to the MSDS supervisor at your institution.

WARRANTY AND LIMITATION OF REMEDY Cayman Chemical Company makes **no warranty or guarantee** of any kind, whether written or oral, expressed or implied, including without limitation, any warranty of fitness for a particular ose, suitability and merchantability, which extends beyond the description of the chemicals hereof. Cayman warrants only to the original customer that the material will meet our specifications

Cayman will carry out its delivery obligations with due care and skill. Thus, in no event will Cayman have any obligation or liability, whether in tort (including negligence) or in contract, for

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Said refund or replacement is conditioned on Buyer giving written notice to Cayman within thirty (30) days after arrival of the material at its destination. Failure of Buyer to give said notice within thirty (30) days shall constitute a waiver by Buyer of all claims hereunder with respect to said material.

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Product Information



Prepare the methyloximating reagent by adding 0.82 g sodium acetate and 0.1 g methoxylamine HCl to 10 ml of a 10:90 ethanol/water solution (both sodium acetate and methoxylamine are available from Cayman Chemical, see front for catalog numbers). This reagent is not stable so it should all be used at one time.

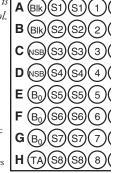
Heat a 1:1 solution of PGE_2 standard or sample and the methoximating reagent at 60°C for 30 minutes. Centrifuge the samples (standards do not need to be centrifuged) and dilute the standard or sample supernatant at least 1:5 with EIA Buffer before addition to the assay well.

Suggested Assay Protocol

This antiserum has been tested and formulated to work exclusively with ACETM reagents. This antiserum may not perform as described if used with other assay reagents or protocols. *NOTE: this is an abbreviated protocol. If you are not familiar with this assay, please contact us for a complete protocol.*

- 1. Add 100 μ l of EIA Buffer to NSB wells and 50 μ l to B₀ wells.
- 2. Add 50 µl of Standard/sample to the appropriate wells.
- 3. Add 50 µl Tracer (Catalog No. 414730) to all wells except Blk and TA.
- 4. Add 50 μl Antiserum to all wells except Blk, TA, and NSB.
- 5. Incubate overnight at 4°C.
- 6. Wash the plate five times with Wash Buffer.
- 7. Add 200 µl Ellman's Reagent to each well.
- 8. Add 5 µl Tracer to the TA well.
- 9. Develop for approximately 90-120 minutes ($B_0 = 0.3-0.8 \text{ AU}$).
- 10. Read absorbance at a wavelength between 405 and 420 nm.

Blk-Blank; **NSB**-Non-specific Binding; **B**₀-Maximum Binding; **TA**-Total Activity; **S1-S8**-Standards; **1-8**-Samples



Related Products

Prostaglandin E_2 - Cat. No. 14010 • UltraPure Water - Cat. No. 400000 • Mouse Monoclonal Anti-Rabbit IgG - Cat. No. 400003 • Precoated (Mouse Anti-Rabbit IgG) EIA 96-Well Strip Plates - Cat. No. 400005 • Precoated (Mouse Anti-Rabbit IgG) EIA 96-Well Plates - Cat. No. 400007 • 96-Well Cover Sheets - Cat. No. 400012 • SPE Cartridges (C-18) - Cat. No. 400020 • Tween 20 - Cat. No. 400035 • Methoxylamine HCl - Cat. No. 400036 • Sodium Acetate - Cat. No. 400037 • Ellman's Reagent - Cat. No. 400050 • EIA Buffer Concentrate (10X) - Cat. No. 400060 • Wash Buffer Concentrate (400X) - Cat. No. 400062 • Prostaglandin E_3 -MOX AChE Tracer - Cat. No. 414730

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