



8-Isoprostane Affinity Purification Kit

Item No. 501110

www.caymanchem.com

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GENERAL INFORMATION

Materials Supplied

Item Number	Item	1 ea Quantity/Size	5 ea Quantity/Size
400220	Eicosanoid Affinity Column Buffer (5X)	1 vial/50 ml	1 vial/50 ml
400230	Eicosanoid Affinity Column Elution Solution	1 vial/25 ml	2 vials/25 ml
401111	8-Isoprostane Affinity Column	1 column/ 0.5 ml sorbent	5 columns/ 0.5 ml sorbent

If any of the items listed above are damaged or missing, please contact our Customer Service department at (800) 364-9897 or (734) 971-3335. We cannot accept any returns without prior authorization.



WARNING: THIS PRODUCT IS FOR RESEARCH ONLY - NOT FOR HUMAN OR VETERINARY DIAGNOSTIC OR THERAPEUTIC USE.

Safety Data

This material should be considered hazardous until further information becomes available. Do not ingest, inhale, get in eyes, on skin, or on clothing. Wash thoroughly after handling. Before use, the user must review the complete Safety Data Sheet, which has been sent *via* email to your institution.

Precautions

Please read these instructions carefully before beginning this assay.

If You Have Problems

Technical Service Contact Information

Phone: 888-526-5351 (USA and Canada only) or 734-975-3888

Fax: 734-971-3641

Email: techserv@caymanchem.com

Hours: M-F 8:00 AM to 5:30 PM EST

In order for our staff to assist you quickly and efficiently, please be ready to supply the lot number of the kit (found on the outside of the box).

Storage and Stability

This kit will perform as specified if stored as directed at 4°C and used before the expiration date indicated on the outside of the box.

ASSAY PROTOCOL

Reagent Preparation

1. Eicosanoid Affinity Column Buffer (5X) (Item No. 400220)

Dilute 1:5 with UltraPure water (*i.e.*, dilute 50 ml vial to a total volume of 250 ml).

2. Eicosanoid Affinity Column Elution Solution (Item No. 400230)

This solution is ready to use.

3. ELISA Buffer

Combine 13.3 g potassium phosphate (dibasic), 3.2 g potassium phosphate (monobasic), 23.4 g sodium chloride, 370 mg tetrasodium EDTA, 100 mg sodium azide, and 1 g bovine serum albumin (Sigma A7030 or equivalent) in a final volume of 1,000 ml. Stir at room temperature until completely dissolved. The pH will be 7.0-7.4. This buffer may be purchased as a 10X concentrated buffer (Item No. 400060).

Purification Protocol

1. Remove the top cap from the column and then gently remove the bottom plug. Be certain to remove the top cap first to avoid air bubbles being drawn into the settled sorbent.
2. Allow the storage solution to pass through the sorbent and then wash the column with 2 ml of Column Buffer. Repeat wash.
3. Apply the sample to the column and allow the entire sample to pass through the sorbent.
4. Wash the column with 2 ml Column Buffer, followed by 2 ml UltraPure Water. Allow all the water to pass through the sorbent. Discard both of these washes.
5. Place a test tube capable of holding at least 5 ml under the column. Elute the 8-isoprostane from the column by adding 2 ml Elution Solution and allowing it to pass through the sorbent. Repeat with an additional 2 ml Elution Solution. If the analysis cannot be performed immediately, store the sample in the Elution Solution at -80°C; it will be stable for at least one year.
6. Evaporate the Elution Solution to dryness either by vacuum centrifugation or under a stream of dry nitrogen. It is important that all the organic solvent be removed as even trace quantities may adversely affect an immunoassay.
7. Immediately dissolve the 8-isoprostane in buffer or solvent appropriate for your application. If you are assaying the sample with one of our 8-Isoprostane ELISA Kits (Item No. 516351 or Item No. 516360), dissolve the sample in ELISA Buffer. The amount of ELISA Buffer depends on the original sample volume and the expected concentration of 8-isoprostane in the sample. A dilute sample may be concentrated by dissolving the residue in a smaller volume of ELISA Buffer than the original sample volume.

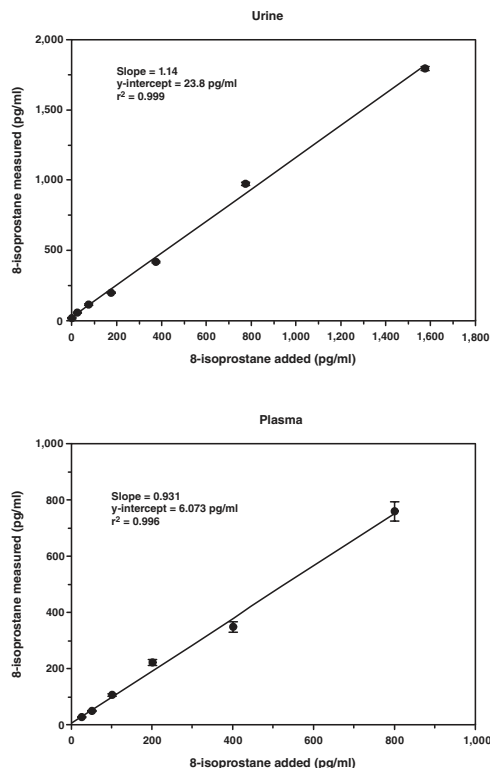
Recovery

Figure 1. Recovery of 8-isoprostane from urine (top) and plasma (bottom). Urine and plasma were spiked with the indicated amount of 8-isoprostane and purifications were performed for each concentration. Samples were analyzed by ELISA. Urine and plasma recoveries average >90%.

NOTESWarranty and Limitation of Remedy

Buyer agrees to purchase the material subject to Cayman's Terms and Conditions. Complete Terms and Conditions including Warranty and Limitation of Liability information can be found on our website.

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