PRODUCT INFORMATION

Thromboxane A Synthase (human, recombinant)

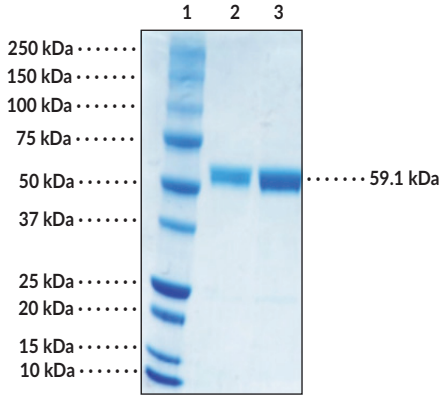
Item No. 33678

Overview and Properties

Synonyms:	CYP5A1, Cytochrome P450 5A1, TBXAS1, TXAS, TXA Synthase
Source:	Active recombinant C-terminal His-tagged thromboxane A synthase expressed in *E. coli*
Amino Acids:	30-533 with a modified N-terminus and F-G loop
Uniprot No.:	P24557
Molecular Weight:	59.1 kDa
Storage:	-80°C (as supplied)
Stability:	≥6 months
Purity:	***batch specific*** (≥90% estimated by SDS-PAGE)
Supplied in:	50 mM Tris, pH 7.4, with 5% glycerol, 0.2 mM DTT, and 0.2 mM EDTA
Protein Concentration:	***batch specific*** mg/ml
Activity:	Confirmed

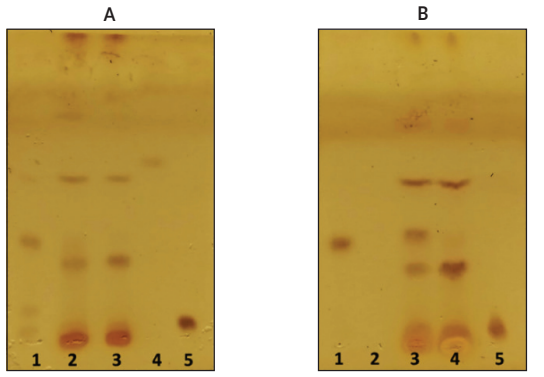
Information represents the product specifications. Batch specific analytical results are provided on each certificate of analysis.

Images



Lane 1: MW Markers
Lane 2: Thromboxane A Synthase (2 µg)
Lane 3: Thromboxane A Synthase (4 µg)

SDS-PAGE Analysis of Thromboxane A Synthase.



Panel A: Assay using 18 µM TXAS

Lane 1: PGH_2
Lane 2: PGH_2 conversion for 30 min.
Lane 3: PGH_2 conversion for one hour.
Lane 4: 12(S)-HHTrE (Item No. 34590)
Lane 5: TXB_2 (Item No. 19030)

Panel B: Assay using 9 µM TXAS

Lane 1: PGH_2
Lane 2: NO substrate control
Lane 3: PGH_2 conversion for 30 min.
Lane 4: PGH_2 conversion for one hour.
Lane 5: TXB_2 (Item No. 19030)

Thin layer chromatographic analysis of Prostaglandin H_2 (PGH_2; Item No. 17020) conversion by recombinant TXAS (human, recombinant). *In vitro* conversion of PGH_2 (50 µM) was done using 18 µM (Panel A) and 9 µM (Panel B) in 250 µL of assay buffer (10 mM potassium phosphate buffer, pH 7.4, with 0.2 mM DTT, 0.5 mM EDTA and 10% glycerol). The concentration of the protein was based on the Ferrous-CO complexed form of the protein. Analysis of the product was performed in a TLC plate using solvent 40:60:1 ethyl acetate:heptane:acetic acid at 4°C. The plates were visualized by spraying 5% sulfuric acid in methanol.

CAYMAN CHEMICAL
1180 EAST ELLSWORTH RD
ANN ARBOR, MI 48108 · USA
PHONE: [800] 364-9897
[734] 971-3335
FAX: [734] 971-3640
CUSTSERV@CAYMANCHEM.COM
WWW.CAYMANCHEM.COM

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Description

Thromboxane A synthase (TXAS), also known as cytochrome P450 (CYP) isoform CYP5A1, is the enzyme that catalyzes the isomerization of prostaglandin H_2 (PGH_2; Item No. 17020) into thromboxane A_2 (TXA_2), a vasoconstrictor and an inducer of platelet aggregation.[1,2] It also catalyzes the cleavage of PGH_2 into malondialdehyde (MDA) and 12(S)-hydroxyheptadecatrienoic acid (12(S)-HHT; Item No. 34590), a leukotriene B_4 (LTB_4) receptor 2 (BLT_2) agonist that also has a role in platelet aggregation.[1] TXAS exists as a monomer and is composed of an N-terminal membrane anchor domain, a heme-binding catalytic residue, and several substrate-binding residues.[3-5] It is expressed in numerous cells, including platelets, monocytes, and macrophages, as well as several tissues, and is localized to the endoplasmic reticulum.[3] TXAS undergoes suicide inactivation during catalysis. Mice deficient in TXAS exhibit prolonged bleeding time and defective platelet aggregation.[6] Cayman's Thromboxane A Synthase (human, recombinant) protein can be used for enzyme activity assay and Western blot (WB) applications. To construct this protein, the N-terminal amino acids 1-29 of human CYP5A1 were removed and replaced with a hydrophilic sequence. The F-G loop of TXAS was modified using sequences from rabbit CYP2C5 and CYP2C3. The C-terminal of this protein contains 6x-His tag followed by a stop codon.

References

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2. Hajeyah, A.A., Griffiths, W.J., Wang, Y., *et al.* The biosynthesis of enzymatically oxidized lipids. *Front. Endocrinol. (Lausanne)* **11**, 591819 (2020).
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5. Seo, M.-J. and Oh, D.-K. Prostaglandin synthases: Molecular characterization and involvement in prostaglandin biosynthesis. *Prog. Lipid Res.* **66**, 50-68 (2017).
6. Yu, I.-S., Lin, S.-R., Huang, C.-C., *et al.* TXAS-deleted mice exhibit normal thrombopoiesis, defective hemostasis, and resistance to arachidonate-induced death. *Blood* **104(1)**, 135-142 (2004).

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