ALPHA DIAGNOSTIC INTERNATIONAL

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Product Specification Sheet

Cyclooxygenase-2 (PGHS-2; PGHS-2; Prostaglandin-endoperoxide synthase-2; COX-2)

Cat. COX22-A	Goat Anti-Rat COX-2 IgG # 2, aff pure	**SIZE**: 100 ug
Cat. COX22-P	Rat COX-2 Control peptide # 2	**SIZE**: 100 ug
Cat. COX22-C	Ovine COX-2 Protein WB +ve control	**SIZE**: 100 ul

Prostaglandins are important regulators of immune responses, fever, and pain. Two isoforms of Prostaglandin H synthase are well characterized, namely COX1 (also called PGHS-1; PHS-1; Prostaglandin-endoperoxide synthase-1) and COX2 (also called PGHS-2; Prostaglandin-endoproxide synthase-2 and PHSII). Both forms of COX proteins are membrane associated heme proteins containing Cyclooxygenase and peroxidase activities. These enzymes are targets of NSAID (Non steroidal anti-inflammatory drugs) such as aspirin. Cox-2 (rat/mouse/human 604 aa) is homodimer of 70KD subunits (1). COX2 is induced by cytokines and mitogens.

COX-2 may play an important role in regulating or promoting cell proliferation in some normal and nepotistic cells. It catalyzes arachidonate + AH(2) + 2 O(2) = PROSTAGLANDIN H2 + A + H(2)O. This enzyme acts both as a dioxygenase and as a peroxidase. It acts as the first step in the formation of prostaglandins and thromboxanes. Cox-2 contains 1 EGF-like domain.

Source of Antigen and Antibodies

Antigen	A 19 amino peptide sequence (designated as COX22-P; **control peptide**) near the C-terminus of rat Cox-2 (1) was synthesized, coupled to KLH
Ab Host/type	Goat, Polyclonal purified IgG, purified over antigen-agarose (Cat # COX22-A
2-Ab	**Rabbit Anti-goat IgG-HRP conjugate** Cat # 30220 (AP, biotin, FITC conjugates also available)
-ve control IgG	**# 20011-1, Goat (non-immune) IgG, purified, suitable for ELISA, Western, IHC as –ve control**

Cox-2 western blot +ve control (# COX22-C) is prepared from **ovine** placenta. For Western blot +ve control (**Cat # COX22-C**) is supplied in SDS-PAGE sample buffer (reduced). Load 10 ul/lane of **COX22-C** for good visibility with antibody Cat # **COX22**-A or other antibodies. Store at –20oC in suitable size aliquots. SDS may crystallize in cold conditions. It should redissolve by warming before taking it from the stock. It should be heated once prior to loading on gels. If the product has been stored for several weeks, then it may be preferable to add 5 ul of fresh 2x sample buffer per 10 ul of the **COX22-C** solution prior to heating and loading on gels. This preparation is not biologically active. It is not suitable for ELISA or other applications where native protein is required. Do not freeze, thaw, or heat repeatedly.

Form & Storage of Antibodies/Peptide Control

Affinity pure IgG

100 ug/100ul solution lyophilized powder

Supplied in **Buffer**: PBS+0.1% BSA

Reconstitute powder in PBS at 1mg/ml

Control/blocking peptide

100 ug/100 ul solution lyophilized powder

Supplied in Buffer: PBS pH 7.5,

Reconstitute powder in PBS at 1 mg/ml.

Storage

Short-term: unopened, undiluted liquid vials at -20OC and powder at 4oC or -20oC..

Long-term: at –20C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

Stability: 6-12 months at –20oC or below.
Shipping: 4oC for solutions and room temp for powder.

Recommended Usage

Western Blotting (1:500-1:3000 for affinity pure serum using Chemiluminescence technique). COX22-A detected ~70 kDa band in LPS+PMA induced RAW 264.7 cells and not in non-induced cells.

ELISA (1:10K-1:30K).

Histochemistry & Immunofluorescence: not tested. We recommend the use of aff pure antibody at 2-20 ug/ml. Cells can be induced with LPS+PMA prior to fixing.

Specificity & Cross-reactivity

The rat COX22-P peptide sequences has significant homology with mouse (100%), rabbit (84%), human (84%), pig (84%), bovine (84%) horse (78%) and chicken (73%) Cox-2. Antibodies show crossreactivity with mouse, rat, and human Cox-2. No cross-reactivity with COX1 was observed. Antibody crossreactivity in various species is not established. Control peptide, because of its low mol. Wt (<3 kDa), is not suitable for Western. It should be used for ELISA or antibody blocking experiments (use 5-10 ug control peptide per 1 ug of aff pure IgG or 1 ul antiserum) to confirm antibody specificity (see detailed protocol at the web site).

General References: (1). Hla, T. & Neilson, K. (1992) PNAS USA, 89, 7384-7388; Jones DA (1993) JBC, 268, 9049-9054; Yamagata,K et al (1993) Neuron 11, 371-386; Ryseck,R.P. et al (1992) Cell Growth Differ. 3 (7), 443.

2. Citations of for ADI Antibodies (see updates at the web site)
DiPerna CA, 2003, J. Thoracic Cardiovascular Surgery, 126, 1129-1133, , IHC
DiPerna CA, 2002, Chest. 122(4) Supplement:165S-166S, , IHC
Carlson NG, 2006, Journal of Neuroimmunology, 174, 21-31, , IHC
Castle PE, 2003, Cancer Epidemiol. Biomarkers Prev., 12: 1449 - 1456, WB,

*This Product is for *in vitro* research use only.

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