ALPHA DIAGNOSTIC INTERNATIONAL

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Product Specification Sheet

EP4 Receptor Antibodies

Cat. # EP41-P	Human EP4 Control Peptide # 1	**SIZE**: 100 ug
Cat. # EP41-S	Rabbit Anti-Human EP4 antiserum # 1	**SIZE**: 100 ul
Cat. # EP41-A	Rabbit Anti-Human EP4 IgG # 1(affinity pure)	**SIZE**: 100 ug

Prostanoids are the cyclooxygenase metabolites derived from C-20 unsaturated fatty acids (arachidonic acid) and include prostaglandin (PG) D2, PGE2, PGF2 alpha, PGI2, and thrombaxane (Tx) A2. The fatty acids precursors are released from the membrane phosphoplipids in response to various physiological and pathological stimuli by the action of phospholipase A2 and are converted to various prostanoids by the sequential actions of cyclooxygenases and the respective synthases. **Prostaglandin PGE2** is one of the major prostaglandin produced during inflammation. A variety of PGE2-mediated effects on vascular smooth muscle tonus, glomerular cell function, renin release, and renal salt and water transport have been described. The actions of PGE2 are mediated by rhodopsin-type; G-protein coupled membrane receptors, termed **E-prostanoid (EP) receptors or PTGERs**. There are four subtypes of PGE receptors designated as **EP1, EP2, EP3, and EP4** that are encoded by different genes and expressed differently in each tissue. The intracellular signaling also differs among the receptor subtypes. In general, EP receptors display a protein topology typical of GPCR - 7 TM domain, an extracellular N-terminus, and a large intracellular C-terminus.

EP4 (mouse 513 aa, rat 488 aa; human 488 aa, chromosome 5p13.1, ~ 90% interspecies homology) is expressed in intestine, lung, thymus, kidney, uterus, pancreas, spleen, heart, stomach, and brain. EP4 knockout mice show patent ductus arteriosus, and decreased inflammatory bone resorption. EP4 activity is mediated by Gs that stimulate adenyl cyclase. It has a relaxing effect on smooth muscle and implicated in renal hemodynamics, intestinal epithelial transport, adrenal aldosterone secretion, and uterine functions. It was originally reported as EP2 subtype.

Source of Antigen and Antibodies

Antigen	16-aa peptide of Human EP4; **Designated (EP41-P or control peptide).;** epitope location ~ N-terminus, Extracellular domain
Ab Host/type	Rabbit, polyclonal Unpurified antiserum (cat #EP41-S) Aff pure IgG (**cat #EP41-A**
2-ab	**Goat Anti-rabbit IgG-HRP** cat # 20320 (AP, biotin, FITC conjugates also available)
-ve control IgG	**# 20009-1, Rabbit (non-immune) IgG, purified, suitable for ELISA, Western, IHC as –ve control**

Form & Storage of Antibodies/Peptide Control

Antiserum (unpurified)
100ul solution lyophilized powder
Supplied 0.05% azide, **Reconstitute** powder in 100 ul PBS

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.

Recommended Usage

Western Blotting (1:1K-5K for neat serum and 1-10 ug/ml for affinity pure antibody using ECL technique). An antibody made to this epitope has been shown to recognize 47 and 78 kDa EP4 bands in the kidney (2-3).

ELISA: Control peptide can be used to coat ELISA plates at 1 ug/ml and detected with antibodies (1:10-50K for neat serum and 0.5-1 ug/ml for affinity pure).

Histochemistry & Immunofluorescence: Not tested. We recommend the use of affinity purified antibody at 2-20 ug/ml as described (2) or paraformaldehyde fixed tissues.

Specificity & Cross-reactivity

The human EP41-P control peptide is 100% conserved in monkey, 87% conserved in rabbit, ovine and canine, 81% in mouse and rat EP4. No significant homology of EP41-P exist with other EP (1, 2 or 3) receptors. Antibody cross-reactivity in various species is not established. The EP41-P 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**. Bastien L et al (1994) J. Biol. Chem. 269, 11873; An S et al (1993) BBRC 197, 263; Foord SM et al (1996) Genomics 35, 182; Mori K et al (199J. Mol. Med. 74, 333-336; Narumiya S et al (1999) Physiol. Rev. 79, 1193 (review).

2. Citations of for ADI Antibodies (see updates at the web site)
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*This product is for In vitro research use only.

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