



800-656-7625

fax 610-367-7825

Certificate of Analysis

Product: Anti-Mouse Angiopoietin 1 (Rabbit)

Code: 100-401-403

Lot #: 18804

Size: 200 µl

Physical State: Liquid (sterile filtered)

Protein Concentration: 75 mg/ml (by Refractometry)

Buffer: 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2

Stabilizer: None

Preservative: 0.01% (w/v) Sodium Azide

Storage Conditions: Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use. Expiration date is one (1) year from date of opening.

Background Information: Angiopoietin-1 (Ang-1) and Angiopoietin-2 (Ang-2) have importance in the development of the endothelium by regulating tyrosine phosphorylation of the membrane receptor Tie-2/Tek. Ang-1 binding to Tie-2/Tek causes phosphorylation of the receptor. Ang-2 competes for this binding, and thus blocks receptor phosphorylation. Ang-1 has a potential fibrinogen-like domain at the carboxyl terminus and coiled-coil regions in the amino terminus. Ang-1 is prominently expressed in the myocardium of atrium and ventricle, mesenchymal and smooth muscle cells surrounding most blood vessels, and lung. In the adult, Ang-1 is also expressed in the heart and liver.

Application Note(s): Suitable for western blotting and other antibody based assays. A 1:500 dilution is recommended for western blotting. The reaction of this antiserum directly with cell supernatants may result in high background due to reactivity of components in the serum. This can be alleviated by first immunoprecipitating the antibody:antigen complex and then detecting the antigen. This method results in a very clean and strong signal. Both Ang-1 and Ang-2 proteins have predicted molecular weights of approximately 57 kDa and appear on western blots close to their predicted molecular weights. In some instances additional bands may be seen at approximately 75 kDa which represent highly glycosylated forms of the protein that migrate at a higher apparent molecular weight.

Recommended Dilutions:

ELISA	1:5,000 - 1:25,000
WESTERN BLOT	1:500 - 1:2,000
IMMUNOHISTOCHEMISTRY	User Optimized
OTHER APPLICATIONS	User Optimized

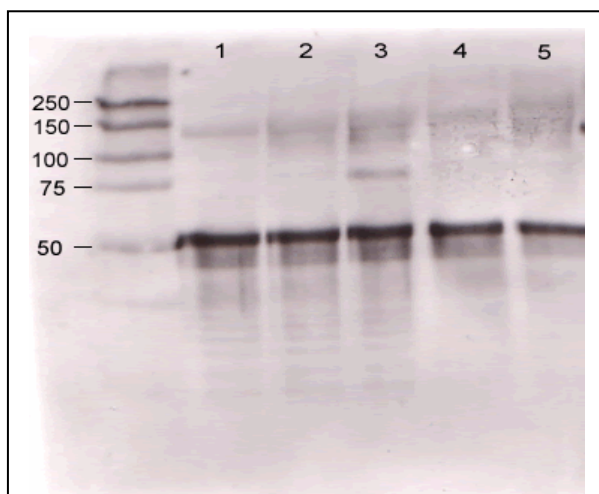


Figure 1. Immunoblotting. Rabbit anti-Ang-1 was used at a 1:500 dilution to detect mouse Ang-1 by western blot against supernatants of mouse angiopoietin-expressing endothelial cells. Lane 1 - wt endothelial cells. Lane 2 - mouse Ang-1 (clone 1-8) expressing cells. Lane 3 - mouse Ang-1 (clone 1-15) expressing cells. Lane 4 - mouse Ang-2 (clone 2-9) expressing cells. Approximately 20 µg of each lysate was used for 10% SDS-PAGE. Immunoprecipitation preceded the reaction with primary antibody at room temperature for 1 h. After subsequent washing, a 1:5,000 dilution of HRP conjugated Gt-a-Rabbit IgG (611-103-122) preceded color development.

Purity and Specificity: This antiserum is directed against mouse angiotensin 1 and shows no reactivity with mouse angiotensin 2. A 57 kDa band is detected when assayed by immunoblot. This reagent cross-reacts with human angiotensin 1. Partially cross reactivity is noted with human angiotensin 2.

Immunogen: This whole rabbit serum was prepared by repeated immunizations with a synthetic peptide N-Q-R-R-N-P-E-N-G-G-R-R-Y-N-R-I-Q-H-G-Q corresponding to a region (aa 21-40) near the N-terminus of mouse angiotensin 1 protein conjugated to KLH using maleimide. A residue of cysteine was added to the amino terminal end to facilitate coupling.

Relevant Links: NCBI [NP_033770](#) (Ang1) NCBI [NP_031452](#) (Ang2)

Protein Sequence: Mouse Angiotensin 1, 496 aa, predicted MW 57.5 kDa

1	mtvflsfaff	aailthigcs	nqrrnpengg	rrynrihqgq	caytfilpeh	dgn cresate
61	qyntnalqrd	aphvepdfss	qklqhlehm	enytwlqkl	enyivenmks	emaqiqnav
121	qnhtatmlei	gtsllsqtae	qtrkldvet	qvlntsrle	iqllenslst	yklekllqq
181	tneilkihek	nsllhekile	megkhkeeld	tlkeekenlq	glvsrqtfii	qelekqlsra
241	tnnnsilqkq	qlelmdtvhn	lislctkegv	llkggkreee	kpfrdcadvy	qagfnksgiy
301	tiyfnmpep	kkvfcnmdiv	gggwtviqhr	edgslfdqrg	wkeykmgfng	psgeywigne
361	fifaitsqrq	ymlrieldw	egnraysqyd	rfhignekqn	yrlylkghtg	tagkqssll
421	hgadfstkda	dndncmckca	lmltggwwfd	acgpsnlngm	fytagqnhgk	lmgikwhyfk
481	gpsyslrstt	mmirpldf				

Related Products:

#100-401-403	Anti-Angiotensin 1 (Mouse specific) (Rabbit)
#100-401-402	Anti-Angiotensin 2 (Mouse specific) (Rabbit)
#611-703-127	Peroxidase Conjugated Affinity Purified Anti-RABBIT IgG (H&L) (DONKEY) MX10
#611-132-122	IRDye800 Conjugated Affinity Purified Anti-RABBIT IgG (H&L) (GOAT) MX10
#KIA-003	MaxTag™ Anti-RABBIT IgG Kit for Immunoblotting
#MB-070	Blocking Buffer for Fluorescent Western Blotting
#B501-0500	BLOTTO
#BSA-30	30% BOVINE SERUM ALBUMIN SOL'N in 0.85% sodium chloride (no preservative or stabilizer)

General References:

Suri, C., Jones, P.F., Patan, S., Bartunkova, S., Maisonpierre, P.C., Davis, S., Sato, T.N. and Yancopoulos, G.D. (1996) Requisite role of angiotensin-1, a ligand for the TIE2 receptor, during embryonic angiogenesis. *Cell*, **87**(7): 1171-1180.

Davis, S., Aldrich, T.H., Jones, P.F., Acheson, A., Compton, D.L., Jain, V., Ryan, T.E., Bruno, J., Radziejewski, C., Maisonpierre, P.C. and Yancopoulos, G.D. (1996) Isolation of angiotensin-1, a ligand for the TIE2 receptor, by secretion-trap expression cloning. *Cell*, **87**(7): 1161-1169.

Maisonpierre, P.C., Suri, C., Jones, P.F., Bartunkova, S., Wiegand, S.J., Radziejewski, C., Compton, D., McClain, J., Aldrich, T.H., Papadopoulos, N., Daly, T.J., Davis, S., Sato, T.N. and Yancopoulos, G.D. (1997) Angiotensin-2, a natural antagonist for Tie2 that disrupts *in vivo* angiogenesis. *Science*, **277**(5322): 55-60.

USDA Certification: All products of animal origin manufactured by Rockland Immunochemicals are derived from starting materials of North American origin. Collection was performed in United States Department of Agriculture (USDA) inspected facilities and all materials have been inspected and certified to be free of disease and suitable for exportation.

Note: This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information.