

Anti-Aldolase ALDOA Antibody

Catalog Number: A05022-1

About ALDOA

Aldolase plays a key role in glycolysis and gluconeogenesis. In addition, it may also function as scaffolding protein. In vertebrates, three forms of this ubiquitous glycolytic enzyme are found, aldolase A in muscle, aldolase B in the liver, and aldolase C in the brain. Alkylation of Arg-43 inactivates the enzyme. Aldolase is involved in step 4 of the subpathway that synthesizes D-glyceraldehyde 3-phosphate and glyceraldehyde phosphate from D-glucose.

Overview

Product Name	Anti-Aldolase ALDOA Antibody
Reactive Species	Human, Rabbit
Description	Boster Bio Anti-Aldolase ALDOA Antibody (Catalog # A05022-1). Tested in ELISA, IP, WB applications. This antibody reacts with Human, Rabbit.
Application	ELISA, IP, WB
Clonality	Polyclonal
Formulation	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2, 0.01% (w/v) Sodium Azide
Storage Instructions	Store vial at 4°C prior to restoration. For extended storage aliquot contents and freeze at -20°C or below. 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. (Ship at ambient temperature.)
Host	Goat
Uniprot ID	P00883

Technical Details

Immunogen	Aldolase [Rabbit Muscle]
Predicted Reactive Species	Bovine, Chicken, Chimpanzee
Isotype	IgG
Form	Lyophilized
Concentration	1 mg/ml by UV absorbance at 280 nm
Purification	Anti-ALDOLASE was prepared from monospecific antiserum by a delipidation, salt fractionation and ion exchange chromatography. Assay by immunoelectrophoresis resulted in a single precipitin arc against anti-Goat Serum, purified and partially purified Aldolase [Rabbit Muscle].

Suggested Dilutions

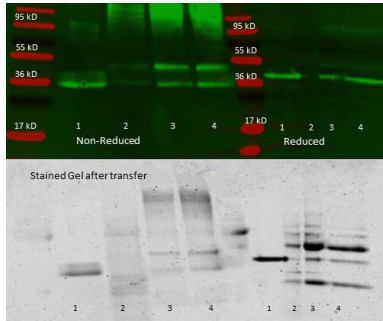
ELISA: 1:7,000

IP: 1:100

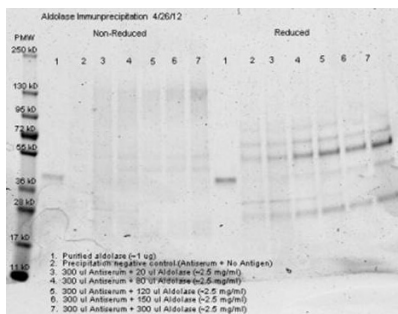
WB: 1:1,000 - 1:5,000

Anti-Aldolase Antibody has been tested by ELISA, immunoprecipitation, and western blot. This product is assayed against 1.0 µg of Aldolase [Rabbit Muscle] in a standard ELISA using Peroxidase conjugated Affinity Purified anti-Goat IgG [H&L] (Rabbit) code #605-4302 and (ABTS (2,2'-azino-bis-[3-ethylbenthiazoline-6-sulfonic acid]) code # ABTS-100 as a substrate for 30 minutes at room temperature. A working dilution of 1:10,000 to 1:40,000 is suggested for this product. Use approximately 5 ul of antibody to immunoprecipitate 50 ul of protein lysate.

Anti-Aldolase ALDOA Antibody (A05022-1) Images

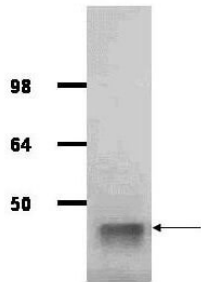
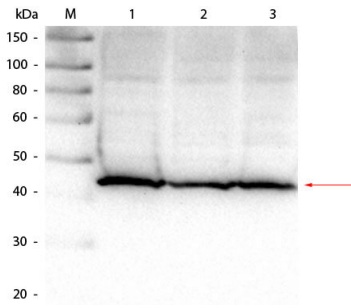


Anti aldolase antibody – Immunoprecipitation and Western Blot. 300 µl aliquots of whole anti-aldolase antiserum (100-1141) were used to precipitate varying amounts of purified aldolase and precipitates with controls were compared by SDS-PAGE and Western blot. Samples shown in the image are: 1. Purified aldolase 2. 300 µl antiserum with no antigen (negative control) 3. 300 µl antiserum with ~100 µl aldolase (2.5 mg/ml) 4. 300 µl antiserum with ~200 µl aldolase (2.5 mg/ml) For the precipitation, 300 ul of antiserum and an equal volume of aldolase antigen in PBS was incubated ~24 hrs at 4°C, centrifuged for 6 minutes at 13K RPM, washed once with PBS, centrifuged and dissolved in 60 ul 0.1 N NaOH. 90 ul of PBS was added, the sample was divided in 2 portions, and an equal volume of reducing (+4% BME) or non-reducing 2X sample buffer was added. The reduced samples were boiled for five minutes, and all samples were run at 140 V for ~45 minutes on a 4-20% tris/glycine gradient gel. Gel was stained, destained and imaged(see attached) using standard protocols. Precipitation of aldolase was confirmed by comparison of increasing amounts of antigen with the control protein by SDS PAGE and observation of a 40-45 kD MW band corresponding to Aldolase. Additional higher and lower molecular weight bands correspond to serum proteins.



Anti aldolase antibody- Immunoprecipitation- Immunoprecipitation was performed with 300 ul of anti aldolase antiserum and an equal volume of varied amounts (diluted from a stock solution of ~2.5 mg/ml) of purified aldolase in PBS. Antibody/Antigen mixture was incubated ~24 hrs at 4°C, centrifuged for 6 minutes at 13K RPM, washed once with PBS, centrifuged and dissolved in 60 ul 0.1 N NaOH. 90 ul of PBS was added, the sample was divided in 2 portions, and an equal volume of reducing (+4% BME) or non-reducing 2X sample buffer was added. The reduced samples were boiled for five minutes, and all samples were run at 140 V for ~45 minutes on a 4-20% tris/glycine gradient gel. Gel was stained, destained and imaged(see attached) using standard protocols. Precipitation of aldolase was confirmed by comparison of increasing amounts of antigen with the control protein by SDS PAGE and observation of a 40-45 kD MW band corresponding to Aldolase. Additional higher and lower molecular weight bands correspond to serum proteins.

Western Blot of Goat anti-Aldolase Antibody. Lane 1: Hela lysate . Lane 2: HEK293 lysate . Lane 3: Jurkat lysate . Load: 25 µg per lane. Primary antibody: Goat anti-Aldolase Antibody at 1:1,000 overnight at 4°C. Secondary antibody: HRP Dk-a-Gt IgG secondary antibody at 1:40,000 for 30 min at RT. Block: for 30 min at RT. Predicted/Observed size: 39 kDa, 41 kDa for Aldolase.



IgG purified antibody to rabbit muscle aldolase (100-1141, 200-1141 and 200-1341) was used at a 1:1000 dilution to detect human aldolase by Western blot. A whole cell lysate prepared from human derived A293 cells was loaded on a 4-12% tris glycine gradient gel for SDS-PAGE. The gel was transferred to nitro-cellulose using standard techniques. Antibody reaction with the membrane occurred overnight at 4° C in TTBS supplemented with 2% non-fat dry milk. Color was allowed to develop using SuperSignal West Pico Chemiluminescent Substrate (PIERCE). Other detection methods will yield similar results. This antibody clearly detects a band at ~41 kDa consistent with human aldolase.

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Anti-Aldolase ALDOA Antibody

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