

Monoclonal Antibodies Specific for Pyruvate Dehydrogenase

Quick Facts

Storage upon receipt:

All products, except A-31853, A-31854

- $\leq -20^{\circ}\text{C}$
- Desiccate

Products A-31853, A-31854

- $2-6^{\circ}$
- Protect from light

Introduction

Molecular Probes offers a series of mouse monoclonal antibodies against specific subunits of the human pyruvate dehydrogenase (PDH) complex (Table 1). PDH is a large multienzyme assembly residing in the mitochondrial matrix and consisting of three catalytic activities: the tetrameric pyruvate decarboxylase, composed of two 41 kD E1 α subunits and two 34.5 kD E1 β subunits; the 72 kD dihydrolipoyl dehydrogenase (E2); and the 55 kD dihydrolipoyl dehydrogenase (E3). The E2 subunit comprises the core of the PDH complex to which the E1 α and E1 β tetramer subunits, as well as the E3 subunit, bind. The E3 subunit is anchored to the E2 subunit by an E3 binding protein to yield a 72 kD/55 kD complex (E2/E3bp).¹

The PDH complex is responsible for the oxidative decarboxylation of pyruvate to form acetyl coenzyme A, which is in turn fed into the citric acid cycle. Deficiencies in the PDH complex lead to lactic acidosis;² severe cases can lead to developmental defects such as congenital brain malformation.³ The subunit-specific anti-

PDH antibodies available from Molecular Probes should prove useful in research applications where structural abnormalities of the PDH complex, possibly associated with functional anomalies, can be detected.

We also offer the anti-PDH E1 α subunit antibody directly conjugated with our Alexa Fluor 594 dye and the anti-PDH E2 subunit conjugated with Alexa Fluor 488 dye (see Table 2).

Materials

Contents

Molecular Probes' unlabeled monoclonal antibodies specific for subunits of mitochondrial pyruvate dehydrogenase are supplied in unit sizes of 100 μg . To prepare stock solutions, reconstitute these antibodies in 500 μL of phosphate-buffered saline (PBS), pH 7.4, containing 1% bovine serum albumin (BSA).

The Alexa Fluor dye-labeled anti-PDH subunit E1 α (A-31853) and subunit E2 (A-31854) antibodies are supplied in unit sizes of 100 μL as 1 mg/mL solutions in PBS, 0.1% BSA, 5 mM sodium azide. At the time of preparation, the products are certified to be free of unconjugated dyes.

Storage

Upon receipt, store the lyophilized antibodies desiccated at $\leq -20^{\circ}\text{C}$. When properly stored, these products are stable for at least one year. Store solutions of the antibodies for up to two weeks at $2-6^{\circ}\text{C}$ with the addition of 2 mM sodium azide. For longer storage, divide solutions into single-use aliquots and freeze at $\leq -20^{\circ}\text{C}$. AVOID REPEATED FREEZING AND THAWING.

Upon receipt of the Alexa Fluor dye-labeled anti-PDH antibodies, store at $2-6^{\circ}\text{C}$. When properly stored, these products are stable for at least six months. For longer storage, divide solutions into single-use aliquots and freeze at $\leq -20^{\circ}\text{C}$. AVOID REPEATED FREEZING AND THAWING, AND PROTECT FROM LIGHT.

Table 1. Monoclonal antibodies for mitochondrial pyruvate dehydrogenase subunits.

Catalog Number	Subunit Specificity	Clone	Isotype	Suggested Concentration for Westerns	Suggested Concentration for IHC	Species Specificity *		
						Hum	Bov	Rat
A-21323	E1 α subunit of PDH	9H9	IgG _{1,k}	0.04 $\mu\text{g}/\text{mL}$	1 $\mu\text{g}/\text{mL}$	+	ND	ND
A-21324	E1 β subunit of PDH	17A5	IgG _{1,k}	5 $\mu\text{g}/\text{mL}$	NR	+	ND	+
A-21325	E2 subunit of PDH	15D3	IgG _{1,k}	0.01 $\mu\text{g}/\text{mL}$	0.1 $\mu\text{g}/\text{mL}$	+	ND	-
A-21326	E2/E3bp component of PDH	13G2	IgG _{2a,k}	0.5 $\mu\text{g}/\text{mL}$	1 $\mu\text{g}/\text{mL}$	+	+	ND

* Species specificity: Hum = human; Bov = bovine; (+), strong reactivity; (-), no reactivity. ND, Not determined; NR, Not recommended.

Table 2. Alexa Fluor dye conjugates of anti-pyruvate dehydrogenase subunits.

Catalog Number	Subunit Specificity	Fluorophore	Abs †	Em †
A-31854	E2 subunit of PDH	Alexa Fluor 488	495	519
A-31853	E1 α subunit of PDH	Alexa Fluor 594	590	617

† Approximate absorption (Abs) and fluorescence emission (Em) maxima, in nm, for conjugates.

Applications

Suggested concentrations for use of the monoclonal antibodies, where available, are given in Table 1. Typically, optimal antibody concentrations for both Western and immunohistochemistry applications range from 1 to 10 μ g/mL.

References

1. J Biol Chem 272, 5757 (1997); 2. Biochem J 239, 89 (1986); 3. Neurology 53, 612 (1999).

Product List *Current prices may be obtained from our Web site or from our Customer Service Department.*

Cat #	Product Name	Unit Size
A-21323	Anti-pyruvate dehydrogenase E1 α subunit (human mitochondrial), mouse IgG ₁ , monoclonal 9H9 (anti-PDH E1 α subunit)	100 μ g
A-31854	anti-pyruvate dehydrogenase E2 subunit (human mitochondrial), mouse IgG ₁ , monoclonal 15D3, Alexa Fluor® 488 conjugate (anti-PDH E2 subunit, Alexa Fluor® 488 conjugate) *1 mg/mL*	100 μ L
A-31853	anti-pyruvate dehydrogenase E1 α subunit (human mitochondrial), mouse IgG ₁ , monoclonal 9H9, Alexa Fluor® 594 conjugate (anti-PDH E1 α subunit, Alexa Fluor® 594 conjugate) *1 mg/mL*	100 μ L
A-21324	Anti-pyruvate dehydrogenase E1 β subunit (human mitochondrial), mouse IgG ₁ , monoclonal 17A5 (anti-PDH E1 β subunit)	100 μ g
A-21325	Anti-pyruvate dehydrogenase E2 subunit (human mitochondrial), mouse IgG ₁ , monoclonal 15D3 (anti-PDH E2 subunit)	100 μ g
A-21326	Anti-pyruvate dehydrogenase E2/E3bp subunit (human mitochondrial), mouse IgG _{2a} , monoclonal 13G2 (anti-PDH E2/E3bp subunit)	100 μ g

Contact Information

Further information on Molecular Probes' products, including product bibliographies, is available from your local distributor or directly from Molecular Probes. Customers in Europe, Africa and the Middle East should contact our office in Leiden, the Netherlands. All others should contact our Technical Assistance Department in Eugene, Oregon.

Please visit our Web site — www.probes.com — for the most up-to-date information.

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