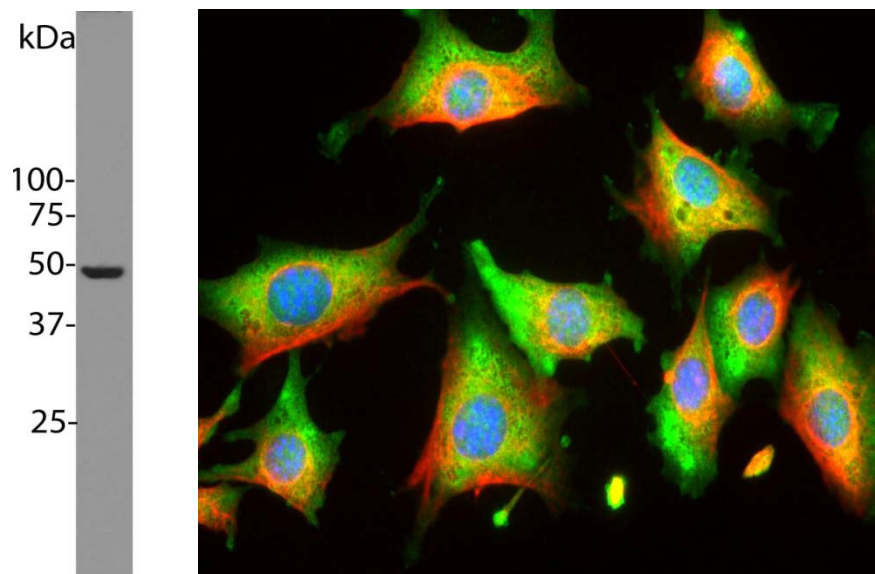


Catalog # MCA-253: Mouse Monoclonal Antibody to α -enolase, non-neuronal

The Immunogen: Enolases are enzymes which catalyze the conversion of 2-phosphoglycerate to phosphoenolpyruvate in the [glycolytic pathway](#), and also the reverse reaction in [gluconeogenesis](#). The [Enzyme Commission](#) classification number for this class of enzyme is [EC4.2.1.11](#), and they can also be referred to as 2-phospho-D-glycerate hydrolases.

There are three enolase proteins each coded for by a distinct gene. They are known as enolase 1, 2 and 3 or alternately as α , β and γ -enolase. Confusingly, although enolase 1 is α -enolase, enolase 2 corresponds to γ -enolase while enolase 3 is also known as β -enolase. All three enzymes exist as dimers *in vivo* and the three molecules are very similar in primary amino acids sequence, as can be seen from a sequence alignment downloadable from [here](#). This similarity allows not only homodimers but also most possible heterodimers to form. The expression pattern of the three enzymes differs in a tissue specific manner, so that antibodies to them are useful cell type specific markers, especially as all three molecules are very abundant. Enolase 1 or α is also known as non-neuronal enolase (NNE) and is expressed in most kinds of tissue, but is absent from neurons. Abnormal expression of NNE is associated with tumor progression in some breast and head and neck cancer (1, 2). Enolase 2 or γ is also known as neuron specific enolase (NSE), and we also market antibodies directed against this protein, specifically [RPCA-NSE](#). A switch from NNE to NSE occurs in the development of neurons (3). Enolase 3 or β is expressed primarily in muscle cells.

Monoclonal antibody MCA-253 was raised against the N-terminal 12 amino acids of bovine enolase 1 which was synthesized on a 8-amine lysine core using the multiple antigen presentation method (5). This produces a [dendrimer](#) presenting 8 peptides to the immune system. The characterization of the antibody has been published in peer reviewed form and in this publication is there referred to as Enol2-53 (4). The antibody was tested for binding to expressed bovine enolase 1, 2 and 3 and shown to be specific for only enolase 1. The [HGNC](#) name for this protein is [ENO1](#).



Left : Blot of HeLa cell lysates probed with monoclonal antibody MCA-253. The antibody stains a single sharp band corresponding to enolase-1/ α -enolase/NNE at about 47 kDa **Right:** Rat 3T3 cells stained with MCA-253 (green) and counterstained with EnCor's chicken polyclonal antibody to vimentin [CPCA-Vim](#) (red) and DNA (blue). The MCA-253 antibody reveals strong cytoplasmic staining, while the vimentin antibody reveals cytoplasmic intermediate filaments.

Antibody characteristics: MCA-253 is a mouse IgG1 class antibody. The antibody solution is purified from tissue culture supernatant and is at a concentration of 1 mg/mL in phosphate buffered saline. The antibody recognizes non-neuronal enolase specifically both in western blots and in immunofluorescence experiments. On blots, M253 4E4 reveals a band at 47 kDa, and on cells in tissue culture the antibody cytoplasmic enolase. Since tubulin is highly conserved, it is likely that the antibody is effective on other species also.

Suggestions for use: The antibody solution can be used at dilutions of at least 1:2,000-1:5,000 in immunofluorescence experiments. In western blotting using chemiluminescence it can be used at dilutions of 1:5,000-1:10,000. Antibody preparation contains 10 mM sodium azide preservative (Link to <http://www.encorbio.com/MSDS/azide.htm> for Material Safety Data Sheet). Avoid repeated freezing and thawing, store at 4°C or -20°C.

References:

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3. Marangos PJ, Schmechel DE, Parma AM, Goodwin FK. Developmental profile of neuron-specific (NSE) and non-neuronal (NNE) enolase. [Brain Res. 190:185-93 \(1980\)](#).
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5. Tam JP, Zavala F.J. Multiple antigen peptide. A novel approach to increase detection sensitivity of synthetic peptides in solid-phase immunoassays. [J. Immunol Methods. 124:53-61 \(1989\)](#).

Limitations: This product is for research use only and is not approved for use in humans or in clinical diagnosis.

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