

Anti-IDO (Indoleamine 2,3-Dioxygenase), clone 10.1

(mouse monoclonal IgG)

Catalog # 05-840

Lot # 32352

Immunogen: GST fusion protein corresponding to residues 78-184 of human Indoleamine 2,3-Dioxygenase (IDO). Clone 10.1.

Specificity: Recognizes IDO (Indoleamine 2,3-Dioxygenase), Mr 42kDa.

Species Cross-reactivity: Human and mouse.

Storage and Stability: Stable for 2 years at -20°C from date of shipment.

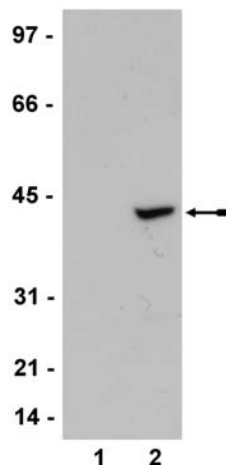
Formulation: 100µg of protein G purified mouse IgG in 185µl of 70% storage buffer (0.1M Tris-glycine, pH 7.4, 0.15M NaCl, 0.05% sodium azide) and 30% glycerol. Store at -20°C.

Handling Recommendations: Upon receipt, and prior to removing the cap, centrifuge the vial and gently mix the solution. Aliquot into microcentrifuge tubes and store at -20°C. **Avoid repeated freeze/thaw cycles, which may damage IgG and affect product performance.** Note: Variability in freezer temperatures below -20°C may cause glycerol-containing solutions to become frozen during storage.

**FOR RESEARCH USE ONLY
NOT FOR USE IN HUMANS**

Quality Control Testing

Immunoblot Analysis: 0.5-2µg/ml of this lot detected IDO (Indoleamine 2,3-Dioxygenase) in RIPA lysates from human IFN-gamma stimulated HeLa cells. **STIMULATE FOR 24 HOURS.**

**Immunoblot Analysis**

Representative blot from a previous lot. Unstimulated (lane 1) and human IFN-gamma 24-hour stimulated (lane 2) HeLa cell lysates were resolved by electro-phoresis, transferred to nitrocellulose and probed with anti-IDO (Indoleamine 2,3-Dioxygenase) (0.5µg/ml). Proteins were visualized using a goat anti-mouse secondary antibody conjugated to HRP and a chemiluminescence detection system. Arrow indicates IDO (Indoleamine 2,3-Dioxygenase) (~42kDa).

General References:

1. Munn, D.H., *et al.*, *Science* **297**: 1867-1870, 2002.
2. Takikawa, O., *et al.*, *J. Biol. Chem.* **263**: 2041-2048, 1988.
3. Taylor, M.W. and Feng, G.S., *FASEB J.* **5**: 2516-2522, 1991.
4. Moffett, J.R. and Namboodiri, M.A., *Immunol. Cell Biol.* **81**: 247-265, 2003.

Immunoblot Protocol

1. Perform SDS-polyacrylamide gel electrophoresis (SDS-PAGE) on a cell lysate sample (cell lysis buffer: 50mM Tris-HCl, pH 7.4; 1% NP-40; 0.25% sodium deoxycholate; 150mM NaCl; 1mM EDTA; 1mM PMSF; 1 μ g/ml each aprotinin, leupeptin, pepstatin; 1mM Na₃VO₄, 1mM NaF) and transfer the proteins to nitrocellulose. Wash the blotted nitrocellulose twice with water.
2. Block the blotted nitrocellulose in freshly prepared TBS containing 5% nonfat dry milk (Catalog # 20-200) and 0.05% Tween[®]-20 (TBST-MLK) for 30 minutes at room temperature with constant agitation.
3. Incubate the nitrocellulose with **0.5-2 μ g/ml of anti-IDO (Indoleamine 2,3-Dioxygenase)**, diluted in freshly prepared TBST-MLK overnight with agitation at 4°C.
4. Wash the nitrocellulose twice with water.
5. Incubate the nitrocellulose in the secondary reagent of choice (a goat anti-mouse HRP conjugated IgG, Catalog # 12-349, 1:3000 dilution was used) in TBST-MLK for 1.5 hours with agitation at room temperature.
6. Wash the nitrocellulose twice with water.
7. Wash the nitrocellulose in TBS-0.05% Tween[®]-20 for 3-5 minutes.
8. Rinse the nitrocellulose in 4-5 changes of water.
9. Use detection method of choice (enhanced chemiluminescence was used).

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