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Antibodies to targets that regulate Immune response

CD80 antigen (CD80) antibodies (Cat # CD80-101AP, P-CD80 and PC-CD80)

Alternate Nomenclature: B-lymphocyte activation antigen B7-1, B7, CD80.

The vertebrate immune system has an important self check mechanism that prevents it from destroying the host own immune system and regulatory T cells are part of this regulatory suppressive control by interacting with CD80 and CD86 (1). The cytotoxic T lymphocyte antigen 4 (CTLA4) is a protein that is expressed on regulatory T lymphocytes and is involved in restraining the immune response when needed and in tachyphylaxis and tolerance. The Fox3p-CD4+ (CD25+CD4+) regulatory T cells (Tregs) express transcription factor Foxp3 suppress aberrant immune responses as seen in autoimmune diseases and allergy (2). CD80 is a co-stimulatory ligand that interacts with CD28 and cytotoxic lymphocyte antigen 4 (CTLA4) in the regulation of T cell response. CD80 along with CD86 expression on antigen presenting cells (APCs) have the ability to augment T cell responses through interactions with CD28 (3) or inhibit T cell responses through interaction with CTLA-4 (4). The CD80/CD86 interaction with CD28/CTLA-4 occurs in the contact area between T cells and APC in parallel with T cell antigen receptor interaction with MHC-peptide complexes and LFA-1 interaction with ICAM-1 (4). The CTLA4 expressed in Fox3p positive tregs is critically required for their in vivo and in vitro suppression which is mediated at least in part by CTLA4 dependent down regulation of CD80 and CD86 in antigen presenting cells.

CD80 and CD86 are cell surface markers expressed on human dendritic cells and mature B-lymphocytes and they play a critical role in stimulating T lymphocyte response by their interaction with the ligand CTLA4 and CD28. CD80 and CD86 are up-regulated during the process of dendritic cell maturation and are over expressed in variety of lymphocytic neoplasm (6). CD80 expression is selectively increases in freshly prepared spleen B cells treated with LPS for 9-12 hours and when they are exposed to X-radiation, whereas CD86 is not affected and only slight increase in CD54, major histocompatibility complex class II, CD11a or surface immunoglobulin M was observed (5). CD80/CD86 co-stimulation is important for the immune response to vesicular stomatitis virus, herpes simplex virus and influenza virus. CD80/CD86 serve as a means of adenovirus entry on human dendritic cells and the interaction of CD80/CD86 with CTLA4 is also important in long-term control of murine gammaherpesvirus-68 virus. CD80 provides a regulatory signals for T lymphocytes as a consequence of binding to the CD28 and CTLA4 ligands of T cells. CD80 is a 321 amino acid (39kDa) protein expressed on Treg.

The CD80-selective antibodies were generated against peptide form unique region of the CD80 gene that is not present in CD antigen or other cellular proteins. The antigenic sequence was common in rat, mouse but distinct from human CD80 gene. FabGennix Inc. has generated epitope specific anti-CD80 mono-epitope-specific antibodies utilizing cyclic peptide methodology. The Anti-CD80 antibodies have been fully characterized for cross reactivity with other cellular proteins using Western blot analyses. Limited quantities of antigenic blocking peptide and Western blot positive controls in ready-to-use buffer for CD80 are also available. FabGennix International Inc., will conjugate this antibody to fluorophores and to secondary enzymes at a nominal charge. FabGennix also carry a wide selection of CD ant

Catalog #	Host	Description	Antigen/ control	Cross reactivity	Quantity
CD80-101AP	Rabbit	Affinity purified CD80 Antibody	Peptide antibody	M, R	100 ug
P-CD80	n/a	Antigenic blocking peptide for CD80-101AP antibody	Synthetic peptide	n/a	250 ug
*PC-CD80	n/a	Western blot positive control for CD80	Ready-to-use WB control for CD80	human	Inquire

R = rat; M = mouse; H = humans; R = rabbit * Actual volume is 103-110 µl; WB, Western Blot analyses; IMM, Immunoprecipitation; IHC, Immunohistochemistry, n.d, not determine.

Immunogen: Synthetic peptide from amino acids 45-63 corresponding to the following sequence (YDH FID CFI EYG DAH VS), was selected from the unique region of the CD80 protein, peptide was post-synthetically modified to achieve highest antigenicity before used for coupling to KLH using heterobifunctional cross linker for immunogen preparation.

Concentration: CD80-101AP antibody has Ig concentration ranging from 0.68-0.70mg/ml in antibody stabilization buffer.

Applications: For ELISA/Dot blot assay the antibody CD80-101AP can be diluted to 1:20,000 in diluObuffer (Cat # FGI-1963). Western blot applications, starting dilution of 1:500 is recommended. Other applications and species cross reactivity for this antibody is yet to be determined.

Reactivity: The antibody CD80-101AP labels a band of around 39kDa in PC-CD80 samples. The Western on the left is a 12% gel with antibody CD80-101AP diluted 1:500 in diluObuffer.

Protocols: Standard protocol for various applications (Western blot; immunoprecipitation and immunohistochemistry) of this antibody can be obtained by calling technical service hotline. The general information on this antibody is supplied with the product. FabGennix Inc. recommends investigators to optimize conditions for optimal usage of this product.

Form/Storage: The antiserum is supplied in antibody stabilization buffer with preservatives. For long-term storage of antibody, store at -20°C FabGennix Inc. does not recommend storage of very dilute antibody solutions unless they are prepared in specially formulated multi-use antibody dilution buffer (Cat # DiluOBuffer). Working solutions of antibodies in DiluOBuffer should be filtered

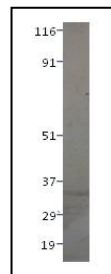
through 0.45um filter after every use for long-term storage.

Notes: Briefly centrifuge to collect liquid before opening the vial, heat the PC-CD80 tube in 90°C water bath for 1-2 minutes to dissolve any precipitate before use. This product is "ready-to-use" for electrophoresis. After thawing store at room temperature. Repeated freezing and thawing may result in appearance of higher molecular weight immunoreactive bands.

Note: Now you can recycle your western blots (nitrocellulose, supported membranes and PVDF membranes) by using our StripObuffer (Cat FGI-1989). Each stripping is guaranteed to give better signal (up to 8 stripping). No strong pungent smell of reducing agents or heating is required.

References:

1. Sakahiumiuchi S. et. al., Cell 133, 775, 2008.
2. Linsley, PS; Brady, W; Grosmaire, L; Aruffo, A; Damle, NK; Ledbetter, JA. Binding of the B cell activation antigen B7 to CD28 costimulates T cell proliferation and interleukin 2 mRNA accumulation. J Exp Med. 1991;173:721.
3. Thompson, CB; Allison, JP. The emerging role of CTLA-4 as an immune attenuator. Immunity. 1997;7:445.
4. Egen, JG; Allison, JP. Cytotoxic T lymphocyte antigen-4 accumulation in the immunological synapse is regulated by TCR signal strength. Immunity. 2002;16:23.
5. Hideko Torihata, Fumio Ishikawa, Yayoi Okada, Yuriko Tanaka, Tetsuya Uchida, Toru Suguro, and Terutaka Kakiuchi. Irradiation up-regulates CD80 expression through two different mechanisms in spleen B cells, B lymphoma cells, and dendritic cells. Immunology. 2004 June; 112(2): 219-227.
6. Maeda, T; Towatari, M; Kosugi, H; Saito, H. Up-regulation of costimulatory/adhesion molecules by histone deacetylase inhibitors in acute myeloid leukemia cells. Blood. 2000;96:3847-3856.



Western Blot of CD80. The CD80 antibody (Cat # CD80-101AP) at 1:500 in diluObuffer was probed with PC-CD80 sample. MW of CTLA4 is approximately 35kDa.

* For users who may require large amounts of CD80-101AP, please enquire about bulk material discounts. This Product is for Research Use Only and is NOT intended for use in humans or clinical diagnosis.

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