Leinco Technologies, Inc.
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Anti-Mouse CD28 [Clone E18] — Purified *in vivo* PLATINUM™ Functional Grade

Hybridoma Monoclonal Antibody

Product Information

Product No.:	C2472
Clone:	E18
Isotype:	Mouse Mouse IgG2b κ

Product Description

Specificity:
E18 activity is directed against mouse CD28 at an epitope close to the B7 binding site.

Antigen Distribution:
CD28 is constitutively expressed on the surface of T cells.

Background:
CD28 is a 44 kD glycoprotein and member of the Ig superfamily that produces co-stimulatory signals necessary for T cell activation and survival as part of the B7-1/B7-2–CD28/CTLA-4 pathway[1]. The two B7 family proteins, B7-1 (CD80) and B7-2 (CD86), have dual specificity for the stimulatory receptor CD28 and the inhibitory receptor CTLA-4 (CD152). When B7-1 and B7-2 interact with CD28, an important co-stimulatory signal, transmitted via CD28, synergizes with the TCR signal to regulate the threshold for T cell activation and promote T cell survival, clonal expansion, and differentiation. CD28 also promotes interleukin-2 (IL-2) production. In contrast, when B7-1 and B7-2 engage with CTLA-4, a negative signal inhibits TCR- and CD28- mediated signaling as well as IL-2 synthesis, and the T-cell response is terminated.

E18 was generated by alternately immunizing $CD28^{-/-}$ mice with A20 cells expressing mCD28 and recombinant mCD28Ig[2]. Splenic cells were fused with X63Ag8.653 for hybridoma production. E18 completely blocks CD28 ligation by B7 molecules[3]. Additionally, E18 enhances anti-CD3-induced proliferation of peripheral T cells. In vivo, E18 acts as an inhibitor of CD28 signaling and causes a reversible reduction in Treg cell frequencies among $CD4^{+}$ cells.

Known Reactivity Species:
Mouse

Format:
Purified in vivo Functional Grade, in vivo PLATINUM™

Products are for research use only. Not for use in diagnostic or therapeutic procedures.

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Immunogen:
CD28 -/- mice were alternately immunized with A20 cells expressing mCD28 and recombinant mCD28Ig

Formulation
This monoclonal antibody is aseptically packaged and formulated in 0.01 M phosphate buffered saline (150 mM NaCl) PBS pH 7.2 - 7.4 with no carrier protein, potassium, calcium or preservatives added. Due to inherent biochemical properties of antibodies, certain products may be prone to precipitation over time. Precipitation may be removed by aseptic centrifugation and/or filtration.

Purity
≥98% monomer by analytical SEC, >95% by SDS Page

Endotoxin
<0.5 EU/mg as determined by the LAL method

Storage and Stability
Functional grade preclinical antibodies may be stored sterile as received at 2-8°C for up to one month. For longer term storage, aseptically aliquot in working volumes without diluting and store at ≤ -70°C. **Avoid Repeated Freeze Thaw Cycles.**

Product Preparation
Functional grade preclinical antibodies are manufactured in an animal free facility using *in vitro* cell culture techniques and are purified by a multi-step process including the use of protein A or G to assure extremely low levels of endotoxins, leachable protein A or aggregates.

Pathogen Testing
To protect mouse colonies from infection by pathogens and to assure that experimental preclinical data is not affected by such pathogens, all of Leinco's Purified Functional PLATINUM™ antibodies are tested and guaranteed to be negative for all pathogens in the IDEXX IMPACT I Mouse Profile.
Other Applications Reported in Literature:

Agonist,
B,
FA,
FC

Country of Origin
USA

References
1. Sharpe AH, Freeman GJ. Nat Rev Immunol. 2(2):116-126. 2002.
2. Dennehy KM, Elias F, Zeder-Lutz G, et al. J Immunol. 176(10):5725-5729. 2006.
3. Beyersdorf N, Ding X, Blank G, et al. Blood. 112(10):4328-4336. 2008.

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4. Langenhorst D, Haack S, Göb S, et al. Front Immunol. 9:1060. 2018.
5. Gladow N, Hollmann C, Ramos G, et al. PLoS One. 15(4):e0227734. 2020.

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