

PD-L1 mAb (10F.9G2), InVivoPure

Endotoxin level ≤ 2 EU/mg

Description:

Programmed death-1 (PD-1) is a cell surface receptor that functions as a T cell checkpoint and plays a central role in regulating T cell exhaustion. Binding of PD-1 to its ligand, programmed death-ligand 1 (PD-L1), activates downstream signaling pathways and inhibits T cell activation. Moreover abnormally high PD-L1 expression on tumor cells and antigen-presenting cells in the tumor microenvironment mediates tumor immune escape, and the development of anti-PD-1/PD-L1 antibodies has recently become a hot topic in cancer immunotherapy.[1]

The 10F.9G2 antibody is a monoclonal antibody specific for the mouse protein PD-L1 (Programmed Death Ligand 1), also known as B7-H1 or CD274. PD-L1 is a 40 kDa type I transmembrane protein that belongs to the B7 family of the immunoglobulin superfamily. It is expressed on T cells, B cells, NK cells, dendritic cells, as well as on IFN- γ stimulated monocytes, epithelial cells and endothelial cells. The interaction of PD-L1 with its receptor PD-1 on T cells leads to the inhibition of T cell proliferation and cytokine production.

The anti-PD-L1 (10F.9G2) blocking antibody blocks interaction of PD-L1 with both of its binding partners, PD-1 and CD80 (B7-1). [2]

This antibody is produced exclusively under serum-free conditions from hybridoma and purified with Protein-A or Protein-G affinity chromatography.

Product-ID:	AK3612P
Clone:	10F.9G2
Immunogen:	murine PD-L1-cDNA + CHO-mPD-L1transfectants [3]
Host:	Rat
Clonality:	Monoclonal
Isotype:	Rat IgG2b κ
Formulation:	Clear Liquid, PBS, pH 7.4, 0.2 μ m sterile filtered
Concentration:	≥ 1.00 mg/mL
Purity:	≥ 90 % (CGE, reducing conditions) ≤ 10 % aggregates (analytical SEC)
Endotoxin:	≤ 2 EU/mg (LAL test)
Storage:	2 - 8 °C
Recommended Isotype Control:	Rat IgG2b Isotype Control (AK3616P)

The product is for research use only and not for use in diagnostic or therapeutic procedures.

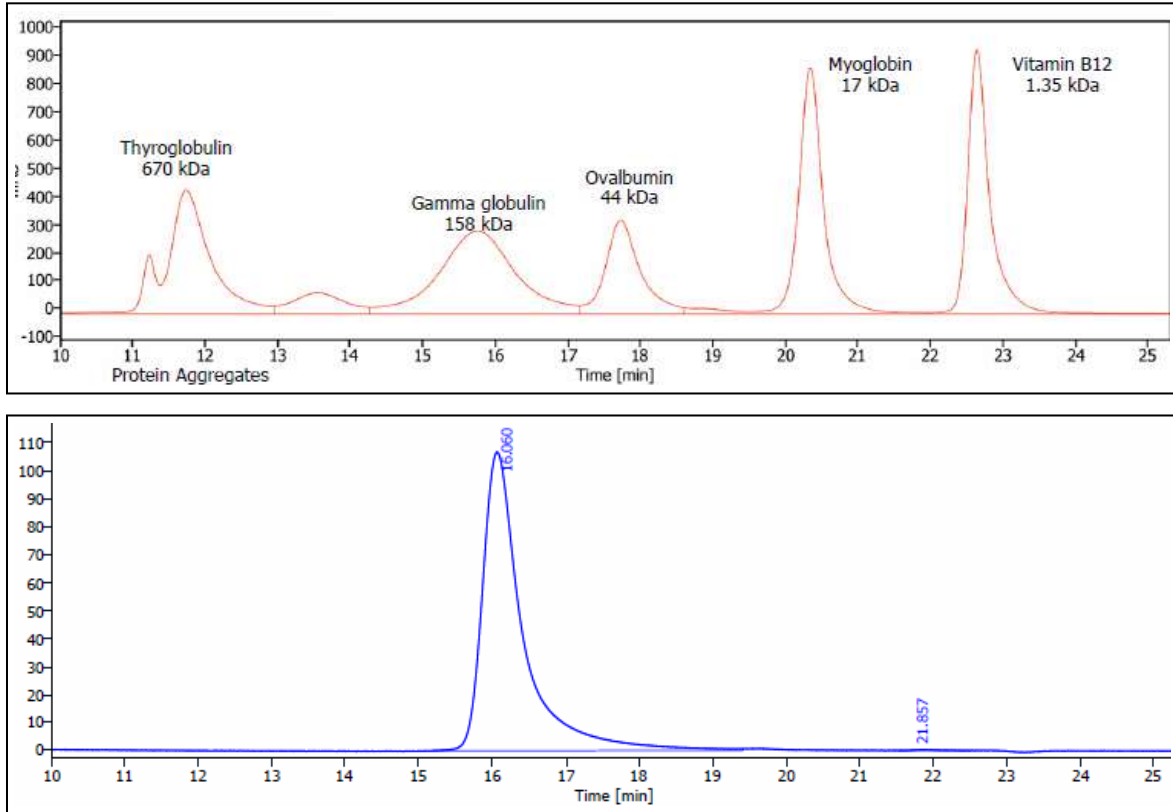
InVivo BioTech Services GmbH is certified to [ISO 9001](#) and [ISO 13485](#).

Literature:

- [1] Jiang Y, Chen M, Nie H, Yuan Y. PD-1 and PD-L1 in cancer immunotherapy: clinical implications and future considerations. *Hum Vaccin Immunother.* 2019;15(5):1111-1122. doi: 10.1080/21645515.2019.1571892. Epub 2019 Mar 19. PMID: 30888929; PMCID: PMC6605868.
- [2] Butte MJ, Keir ME, Phamduy TB, Sharpe AH, Freeman GJ. Programmed death-1 ligand 1 interacts specifically with the B7-1 costimulatory molecule to inhibit T cell responses. *Immunity.* 2007 Jul;27(1):111-22. doi: 10.1016/j.immuni.2007.05.016. Epub 2007 Jul 12. PMID: 17629517; PMCID: PMC2707944.
- [3] Eppihimer MJ, Gunn J, Freeman GJ, Greenfield EA, Chernova T, Erickson J, Leonard JP. Expression and regulation of the PD-L1 immunoinhibitory molecule on microvascular endothelial cells. *Microcirculation.* 2002 Apr;9(2):133-45. doi: 10.1038/sj/mn/7800123. PMID: 11932780; PMCID: PMC3740166.

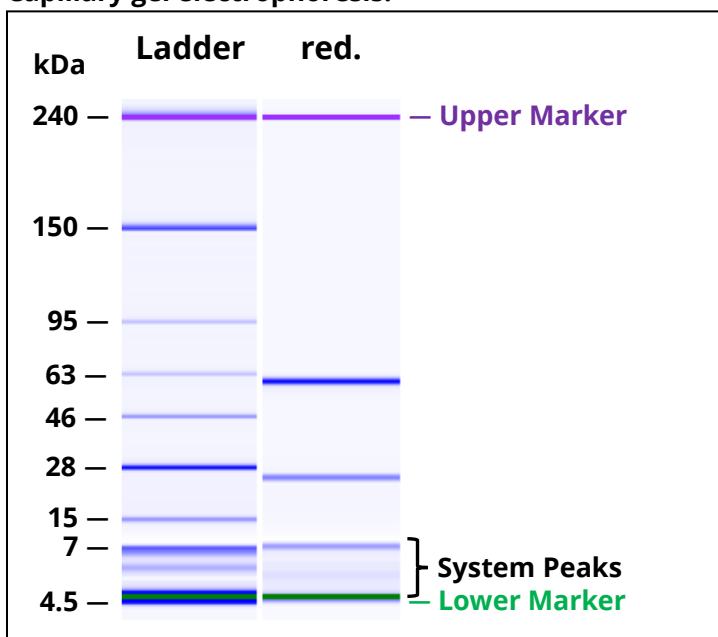
**PD-L1 mAb (10F.9G2), InVivoPure
— Supplementary Data**

Analytical SEC:



Analytical SEC of purified protein (blue) in comparison with gel filtration standard (red).

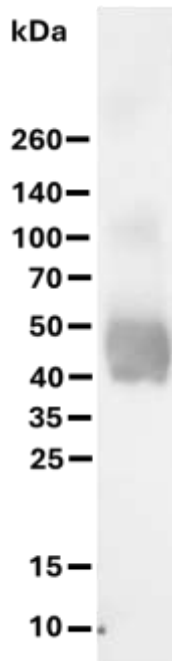
Capillary gel electrophoresis:



CGE of the purified protein under reducing (red.) conditions.

The following methods were used for the additional characterization of one exemplary batch:

Western blot analysis:



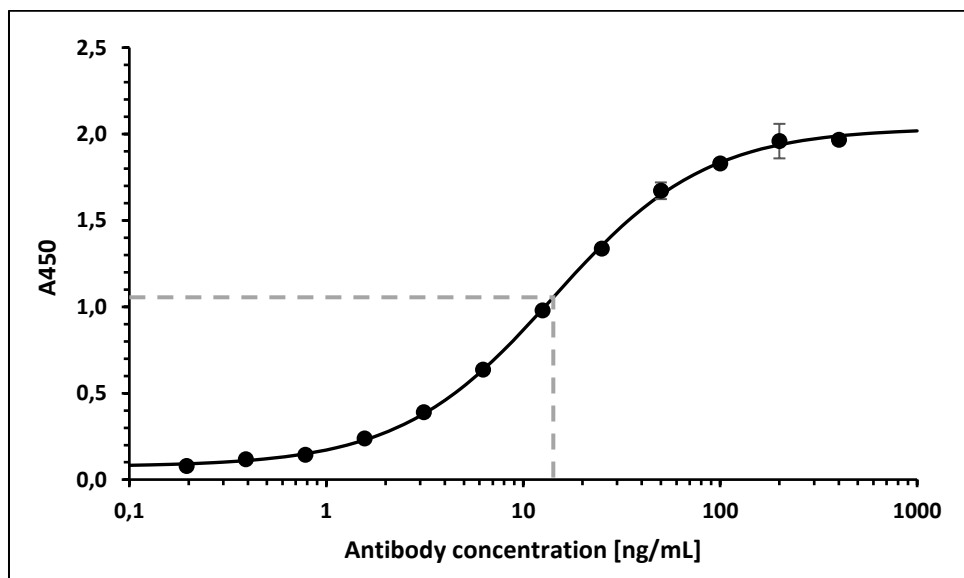
Lane 1: 1 µg reduced purified mouse PD-L1 with C-terminal his-tag

Primary antibody: PD-L1 mAb (10F.9G2), InVivoPure+ (AK3612P/01.1), 1:145 dilution

Secondary antibody: HRP labelled goat anti-rat IgG (H+L), 1:5.000 dilution

Predicted band size: ~40-50 kDa

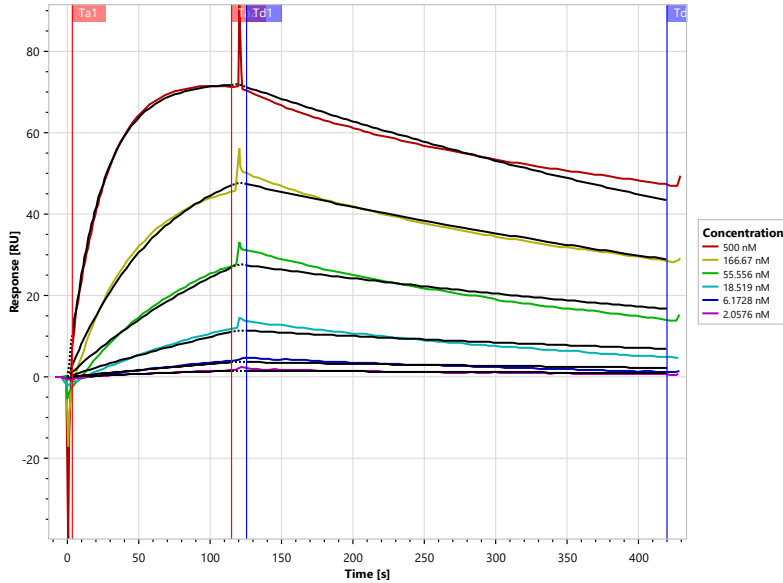
ELISA analysis:



ELISA analysis of PD-L1 mAb (10F.9G2) (Product-ID: AK3612P/01.1). Coating antigen: Mouse PD-L1 (B7-H1) His-tag, at 1 µg/mL. The EC50 of the antibody is 14,22 ng/mL.

SPR analysis:

Target	Captured Target	Analyte	K_D [M]	k_{on} [$M^{-1}s^{-1}$]	k_{off} [s^{-1}]
ProtA/G	PD-L1 mAb (10F.9G2), AK3612P/01.1	PDL1	2.1E-08	8.2E+04	1.7E-03



A high capacity amine chip (Bruker Part No:1862614) was immobilized with Protein A/G. The antibody was captured at a concentration of 1.5 $\mu\text{g/mL}$. For the analyte, a concentration range of 2.06 to 500 nM is used in a multi-injection cycle kinetics assay on a Bruker SPR-32 to determine the K_D , k_{on} and k_{off} of the antibody-antigen-binding.