

DP3514**Polyclonal Antibody to His-tag (C-term) - Aff - Purified****Quantity:**

50 µg

Background:

In the last couple of years many peptide sequences/epitopes for the purification of recombinant proteins have been established. These so-called “tags” can be used e.g. to determine the cellular localization or to quantify proteins. The polyhistidine “tag” (His-tag) is the most used affinity epitope for the purification of recombinant proteins [1]. Proteins with a polyhistidine tag (e.g. 6xHis or 8xHis) can be purified in one step using a metal-chelate column (e.g. Ni²⁺, Zn²⁺, Cu²⁺ or Co²⁺) and imidazole as eluent. This method now is a very attractive system for the purification of larger amounts proteins for structural and functional studies. So far His-tagged proteins were successfully purified from different expression systems like E. coli, yeast, insect cells and plant cells [1,2]. An important requirement beside the efficient and robust purification method is the availability of a fast detection system for checking the purification steps of these His-tagged proteins if no specific antibody is available.

Host:

Rabbit

Immunogen:

Different highly purified 6x His-tagged proteins (C-terminal) produced in insect cells.

Format:**State:** Lyophilized purified IgG fraction.**Purification:** Antigen Affinity Chromatography using a His-peptide as matrix.**Buffer System:** PBS, pH 7.4, without preservatives.**Reconstitution:** Restore in sterile water to a concentration of > 0.5 mg/ml.**Applications:**

Western blot (0.5-1 µg/ml): The antibody was tested using several His-tagged proteins expressed in insect cells and E. coli.

Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.

Specificity:

Western analysis with several His-tagged proteins either expressed in insect cells or E.coli showed that the antibody recognizes all tested proteins fused to a C-terminal but not to a N-terminal His-tag.

The antibody might be a very good tool to test supernatants or cell lysates for expression of recombinant proteins.

The anti-His-Tag antibody is able to detect recombinant proteins in the conditioned media from insect cells and total lysate from E.coli.

Storage:

The lyophilized IgG is stable at 2-8°C for one month from despatch and for one year when kept at -20°C.

The reconstituted antibody can be stored at 2-8°C for one month or at -20°C for one year.

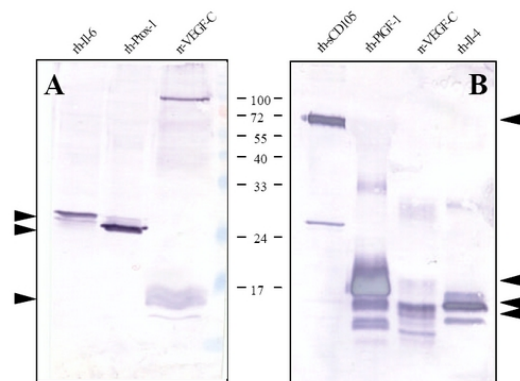
Avoid repeated freezing and thawing.

General Readings:

1. Terpe K, Appl. Microbiol. Biotechnol. 60:523, 2003.
2. Leelavathi et al, Molecular Breeding 11:49, 2003.
3. Bernhardt et al, Laborwelt 5:18, 2004 (German).

Pictures:

Western blot (AP) of several His-tagged proteins from supernatants (A) and after purification (B) by affinity chromatography using metal chelate columns (Ni²⁺, Co²⁺) as matrix. The anti-His-tag antibody concentration used was 0.1µg/ml.



Western blot (ECL) detection of different His-tagged proteins. For Western blot analysis each lane was loaded with 12,5-50 ng with the following His-tagged proteins: Lane 1: TbTX, Lane 2: IL-3, Lane 3: VEGF121, Lane 4: PlGF-1, Lane 5: Lac I repressor, Lane 6: soluble FLT-4.

