

Product datasheet

Anti-TNF alpha Affibody® Molecule (Agarose) ab31909

1 References 1 Image

Overview

<b>Product name</b>	Anti-TNF alpha Affibody® Molecule (Agarose)
<b>Conjugation</b>	Agarose
<b>Conjugation notes</b>	This molecule is immobilized on agarose at the unique C-terminal cysteine.
<b>Specificity</b>	ab31909 recognises TNF alpha.
<b>Tested applications</b>	<b>Suitable for:</b> IP
<b>Species reactivity</b>	<b>Reacts with:</b> Human
<b>Immunogen</b>	Other Immunogen Type corresponding to Human TNF alpha. Recombinant protein produced in E. coli.
<b>General notes</b>	ab31909 is a recombinant protein produced in E. coli.

**What are Affibody Molecules?**

*Affibody® affinity ligands are small, simple proteins composed of a three-helix bundle based on the scaffold of one of the IgG-binding domains of Protein A. Protein A is a surface protein from the bacterium Staphylococcus aureus. This scaffold has excellent features as an affinity ligand and can be designed to bind with high affinity to any given target protein. The domain consists of 58 amino acids, 13 of which are randomized to generate Affibody® libraries with a large number of ligand variants. Thus, the libraries consist of a multitude of protein ligands with an identical backbone and variable surface-binding properties. The current Affibody® libraries contains billions of variants. In function, Affibody® molecules mimic antibodies, nature's own binders to an infinite number of antigens. Compared to antibodies, the most striking dissimilarity of Affibody® molecules is the small size. Affibody® molecules have a molecular weight of 14 kDa, compared to the molecular weight of antibodies, which is 150 kDa. In spite of its small size, the binding site of Affibody® molecules is similar to that of an antibody. The advantages of Affibody® molecules over antibodies are · their small size · the simple structure of the molecules · its robust physical properties · its ability to fold correctly intracellularly · the fast and cost-efficient production in bacteria · the possibility to produce Affibody® molecules through chemical synthesis · the possibility to couple Affibody® molecules in multimeric constructs*

Properties

<b>Form</b>	Liquid
<b>Storage instructions</b>	Shipped at 4°C. Store at +4°C. Do Not Freeze.

<b>Storage buffer</b>	pH: 7.50 Preservative: 0.02% Sodium azide Constituents: 0.328% Sodium phosphate, 0.87% Sodium chloride
<b>Purification notes</b>	ab31909 is >98% as determined by SDS-PAGE and RP-HPLC analysis.
<b>Affibody® molecule notes</b>	<p><b>What are Affibody Molecules?</b></p> <p><i>Affibody® affinity ligands are small, simple proteins composed of a three-helix bundle based on the scaffold of one of the IgG-binding domains of Protein A. Protein A is a surface protein from the bacterium Staphylococcus aureus. This scaffold has excellent features as an affinity ligand and can be designed to bind with high affinity to any given target protein. The domain consists of 58 amino acids, 13 of which are randomized to generate Affibody® libraries with a large number of ligand variants. Thus, the libraries consist of a multitude of protein ligands with an identical backbone and variable surface-binding properties. The current Affibody® libraries contains billions of variants. In function, Affibody® molecules mimic antibodies, nature's own binders to an infinite number of antigens. Compared to antibodies, the most striking dissimilarity of Affibody® molecules is the small size. Affibody® molecules have a molecular weight of 14 kDa, compared to the molecular weight of antibodies, which is 150 kDa. In spite of its small size, the binding site of Affibody® molecules is similar to that of an antibody. The advantages of Affibody® molecules over antibodies are · their small size · the simple structure of the molecules · its robust physical properties · its ability to fold correctly intracellularly · the fast and cost-efficient production in bacteria · the possibility to produce Affibody® molecules through chemical synthesis · the possibility to couple Affibody® molecules in multimeric constructs</i></p>
<b>Function</b>	Cytokine that binds to TNFRSF1A/TNFR1 and TNFRSF1B/TNFR2. It is mainly secreted by macrophages and can induce cell death of certain tumor cell lines. It is potent pyrogen causing fever by direct action or by stimulation of interleukin-1 secretion and is implicated in the induction of cachexia, Under certain conditions it can stimulate cell proliferation and induce cell differentiation.
<b>Involvement in disease</b>	Genetic variations in TNF are a cause of susceptibility psoriatic arthritis (PSORAS) [MIM:607507]. PSORAS is an inflammatory, seronegative arthritis associated with psoriasis. It is a heterogeneous disorder ranging from a mild, non-destructive disease to a severe, progressive, erosive arthropathy. Five types of psoriatic arthritis have been defined: asymmetrical oligoarthritis characterized by primary involvement of the small joints of the fingers or toes; asymmetrical arthritis which involves the joints of the extremities; symmetrical polyarthritis characterized by a rheumatoidlike pattern that can involve hands, wrists, ankles, and feet; arthritis mutilans, which is a rare but deforming and destructive condition; arthritis of the sacroiliac joints and spine (psoriatic spondylitis).
<b>Sequence similarities</b>	Belongs to the tumor necrosis factor family.
<b>Post-translational modifications</b>	The soluble form derives from the membrane form by proteolytic processing. The membrane form, but not the soluble form, is phosphorylated on serine residues. Dephosphorylation of the membrane form occurs by binding to soluble TNFRSF1A/TNFR1. O-glycosylated; glycans contain galactose, N-acetylgalactosamine and N-acetylneuraminic acid.
<b>Cellular localization</b>	Secreted and Cell membrane.

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## Applications

Our [Abpromise guarantee](#) covers the use of **ab31909** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

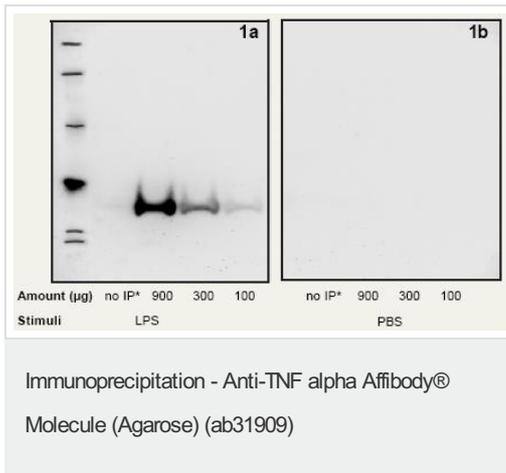
Application	Abreviews	Notes
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IP

Use at an assay dependent concentration.  
 The molecule is immobilized on agarose at the unique C-terminal cysteine.

This agarose immobilized Anti-TNF alpha Affibody® molecule is excellent for immunoprecipitation studies of TNF alpha in cell extracts or other solutions that contain TNF alpha proteins. Can also be used for affinity chromatography.

## Images



The agarose immobilized Anti-TNF alpha Affibody® molecule precipitates TNF-alpha protein from cell extracts derived from LPS stimulated but not from unstimulated THP-1 cells.

### RESULTS

#### IMMUNOPRECIPITATION

The human pro-monocytic cell line THP-1 was primed with PMA (200 nM) over night and stimulated with LPS (200 ng/ml) or with PBS (control) for 2 hours prior to termination of culture. Cell extracts prepared from LPS stimulated cultures and from control cells were incubated with agarose immobilized Anti-TNF-alpha Affibody® molecule for 2 hours. After incubation, the unbound proteins were washed away and the bound protein was eluted with SDS-PAGE separation and blotted onto a PVDF filter. The filter was stained with an antibody against full length TNF-alpha protein (25 kD).

Figure 1a and 1b show that TNF-alpha was precipitated from LPS stimulated, but not from PBS stimulated THP-1 cell extract, using agarose immobilized Anti-TNF alpha Affibody® molecule. With increased amount of total protein, the TNF-alpha band became more intense. TNF-alpha was not detected in cell extracts prior to immunoprecipitation proving that an immunoprecipitation step is needed for TNF-alpha detection in this model system.

These results show that the Anti-TNF alpha Affibody® molecule efficiently precipitates TNF-alpha proteins from a complex mix of proteins. When performing immunoprecipitation experiments with antibodies, there is often a problem with cross reactions between the enzyme conjugated second step reagent and the precipitating antibody. This type of cross reaction is avoided with an Affibody® molecule as the precipitating reagent.

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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