



Tagged Antigens for Antibody Development

www.dauids-bio.com (Custom Antibodies)

www.dauids-science.de (Lab Material)

- 1 - Introduction

Tags are used to purify recombinant proteins from a bacterial or eucaryotic cell culture expression. Different tags like HIS-, GST-, MBP- or Strep-tag can be used for a purification of the protein. In this way the purification of proteins is easier and usually leads to better results. When a tagged protein is used as antigen, the animal will produce antibodies against the protein and the tag.

For the immunizations it is helpful to know that the antigen contain a tag. Some tags like GST and MBP have a high antigenicity and it is important to know if the ELISA results represent the protein and not the tag. For this reason, please indicate at the start of the immunizations that the antigen contains a tag.

- 2 - Method

Anti-tag antibodies

In many cases, the anti-tag antibodies do not disturb applications. In this case the tagged proteins can be used directly as antigens.

Removal of tag

When the anti-tag antibodies disturb in the target application, a removal of the tag prior to immunization is recommended. If this is not possible there are other ways to get rid of antibodies that recognize the tag.

Immunization and Purification with different tags

When the antigen is available with two different tags, the immunization can be performed with Antigen-tag1 and the purification with Antigen-tag2. In this case the antiserum contains antibodies against the antigen and tag1. When the affinity purification is performed with Antigen-tag2, only antigen specific antibodies are eluted, because tag1 antibodies cannot bind the Antigen-tag2 matrix.

Removal of anti-tag antibodies

Especially, when the tag removal is expensive and time consuming, or when the tag is needed for solubility reasons, a removal of the tag might not be possible. In this case the anti-tag antibodies can be removed after the immunization from antisera or purified antibodies. The so called depletion removes unwanted antibodies with a tag-matrix.

Antigenicity of tags

Every tag is different, and some tags have a good antigenicity whereas others don't have. When the tag has a very good antigenicity and the protein only has a poor antigenicity, most of the final antibodies may recognize the tag.