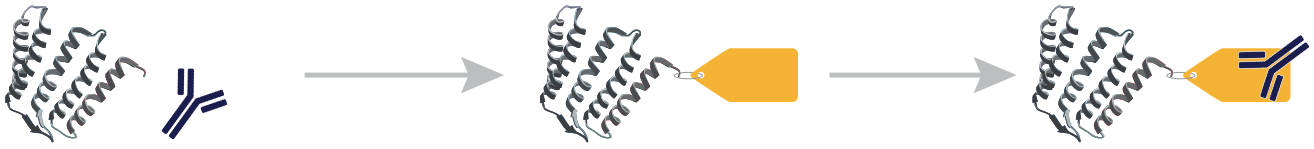
Epitope tags

Epitope tags are often genetically engineered into recombinant proteins during cloning, providing a method for the detection and/or purification of the expressed protein. They can be used to study the topology of proteins and protein complexes, identify associated proteins, and characterize newly identified, low-abundance proteins when specific antibodies are not available. Epitope tag antibodies are commonly used in structure/function studies where modified constructs are compared against native proteins.



Expression proteins against which no antibodies are available yet

A suitable tag genetically introduced into the protein of interest

Expressed proteins can now be readily detected through tag antibodies

Peptide tags

Short amino acid sequences. Some epitope tags, such as myc, are often used in tandem to increase their specific detection, or in combination with another tag.

Examples: Hemagglutinin (HA), His, myc, V5, FLAG™ tag

Uses

- Enable easy affinity purification of expressed proteins
- Enable localization and expression of proteins
- Used in pull-down immunoprecipitation (IP) experiments

Applications

- Western blot, ELISA, chromatin IP (ChIP), immunocytochemistry (ICC), and immunohistochemistry (IHC)

Detection

- By antibodies either directly conjugated to epitope tags or detected with secondary antibodies

Protein tags

Large-sized proteins when tagged with a protein of interest result in fusion protein.

Examples: GST

Uses

- Can aid in protein expression—enhances the solubility of expressed proteins
- GST can be easily purified using glutathione agarose

Applications

- IP, western blot, ELISA, ChIP, ICC, and IHC

Detection

- By antibodies either directly conjugated to epitope tags or detected with secondary antibodies

Fluorescent tags

Large-sized proteins when tagged with a protein of interest result in fusion protein and provide a fluorescent label to the protein of interest.

Examples: GFP, RFP, mCherry

Uses

- Useful to monitor protein–protein interactions by FRET, cell tracking, and cellular localization experiments

Applications

- Western blot, ELISA, IHC, and fluorescence measurement

Detection

- By innate fluorescence of the epitope tags or by antibodies when other detection methods are needed

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Most commonly used epitope tags

Name	Type of tag	Sequence/size	Description
HA	Peptide tag	YPYDVPDYA	A highly immunoreactive epitope and can be used to purify tagged target proteins; can be used for co-IP studies and western blots.
His	Peptide tag	6X-His HHHHHH tag	Widely used tag for purification of target proteins that can be used with almost all expression systems.
FLAG	Peptide tag	DYKDDDDK	The high hydrophilicity and small size of the FLAG tag tend to interfere less with protein expression, proteolytic maturation, antigenicity, and function.
Myc	Peptide tag	EQKLISEEDL	A reliable method for the detection and purification of tagged target proteins without a protein-specific antibody or probe.
V5	Peptide tag	GKPIPNPLLGLDST	Widely used in affinity purification in combination with His-tag.
GST	Peptide tag	27 kDa	Highly soluble protein, resulting in greater expression and solubility of recombinant proteins with the GST tag, and can be especially helpful when expressing proteins in bacteria; the GST protein also has a strong affinity for its substrate, glutathione.
GFP	Fluorescent tag	27 kDa	A versatile marker for monitoring physiological processes, visualizing protein localization, and detecting transgenic expression. GFP is also a powerful research tool for assessing gene expression and subcellular protein distribution in fixed or living cells.
mCherry	Fluorescent tag	28 kDa	Due to its improved brightness, superior photostability, and extremely rapid maturation rate, the mCherry monomeric red protein is one of the fluorescent proteins of choice for monitoring physiological processes and detecting transgenic expression.

Find out more at **thermofisher.com/epitopetags**

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