

Strep-tag® and One-STrEP-tag for Protein-protein Interaction Analysis

IBA GmbH

BioTechniques Protocol Guide 2009 (p. 55) doi 10.2144/000113007

Protein-protein interactions (PPI) govern almost all important processes in living organisms. Thus, their rapid and accurate determination and investigation is a major challenge in life sciences. With four different determination systems based on *Strep-tag*®II and One-STrEP-tag, we provide optimal solutions for in vivo protein-protein interaction analysis.

Materials

One-STrEP Set (Cloning and Purification Kit for Mammalia and *Escherichia coli*)

One-TAP Set (Cloning and Purification Kit for Mammalia and *E. coli*)

Two-TAP Set (Cloning and Purification Kit for Mammalia and *E. coli*)

Spine Set (Cloning and Purification Kit for *E. coli*)

Methods

The **One-STrEP** system is recommended for getting started. It needs one tag and one

purification step only. Due to its excellent performance, this method yields a favorable signal-to-noise ratio in most cases. Mild elution and fast washing allow the isolation of even weakly interacting preys.

In case the One-STrEP system provides sub-optimal data, the **One-TAP** system extends the options of the One-STrEP system since it adds a second independent purification step yet with the same tag. Two different purification steps may better discriminate specific from nonspecific binding but bear the risk of losing weakly interacting partners.

The **Two-TAP** system is recommended only as an option in case of unsatisfying data with the

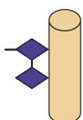
One-STrEP or One-TAP approach and not as first choice starting point.

In addition to these non-covalent capture methods of potential preys, **SPINE** adds the possibility to covalently link the preys to its bait by formaldehyde cross-linking. This linkage is achieved in the living organism enabling a time resolved snapshot of interacting proteins. SPINE is currently validated in prokaryotes only but its adaptation to mammalian system is under way.

StarGate for Bait Cloning

This novel cloning system is the perfect tool for efficient screening and fast identification of the optimal tag for PPI investigation with a given bait. Once the bait is cloned into a **Donor Vector**, a large selection of **Acceptor Vectors** for its expression with different tag arrangements in the desired host is available.

Address correspondence to IBA GmbH Headquarters, Rudolf-Wissell-Strasse 28, 37079 Göttingen, Germany; Tel.: Europe +49 551 50672-0, USA +1 877 422 4624; info@iba-go.com, www.iba-go.com.

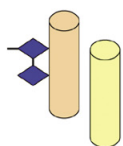


Recommended method to start

One-STrEP
one tag
one column

- only one tag
- even weakly interacting preys are isolated

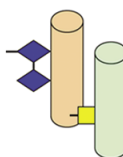
(Juntilla et al., Proteomics 2005)



Recommended in case of background with One-STrEP method

One-TAP
one tag
two columns

- two purification steps with only one tag increase signal:noise ratio
- recommended for high-affinity PPI

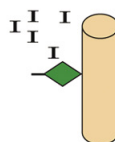


Alternative option only if One-TAP performs insufficiently

Two-TAP
two tags
two columns

- improved purification procedure for FLAG®-users with two tags
- improvement of original TAP procedure

(Gloeckner et al., Proteomics 2007)



Reversible cross-linking in vivo with formaldehyde

SPINE
one tag
one column
formaldehyde

- only one tag
- time-resolved map of interacting proteins possible

(Herzberg et al., Proteomics 2007)