

Datasheet: TZC001

BATCH NUMBER 167145

Description:	SpyCatcher2
Name:	SPYCATCHER2
Format:	Catcher
Product Type:	Recombinant Protein
Quantity:	1 mg

Product Details

Applications

This product has been reported to work in the following applications. This information is derived from testing within our laboratories, peer-reviewed publications or personal communications from the originators. Please refer to references indicated for further information. For general protocol recommendations, please visit www.bio-rad-antibodies.com/protocols.

	Yes	No	Not Determined	Suggested Dilution
Immunoassay	■			

Where this product has not been tested for use in a particular technique this does not necessarily exclude its use in such procedures. Suggested working dilutions are given as a guide only. It is recommended that the user titrates the product for use in their own system using appropriate negative/positive controls.

Target Species	Protein/peptide tag
Product Form	SpyCatcher2 purified recombinant protein - liquid
Preparation	Recombinant protein expressed in <i>E. coli</i> and purified by affinity chromatography
Source	E.coli
Buffer Solution	Phosphate buffered saline
Preservative Stabilisers	0.0095% MIT
Approx. Protein Concentrations	250 µM (3.9 mg/ml)

Product Information **SpyCatcher2** (H-SpyC2) is a 15.7 kDa protein that forms a stable covalent isopeptide bond with a second protein that includes a SpyTag. The reaction occurs spontaneously on mixing, is rapid, irreversible, high-yielding and shows good specificity. SpyCatcher2 contains a His6-tag at the N-terminus. To avoid deamidation of asparagine, a N105D

mutation was introduced ([Hentrich *et al.* 2020](#)).

The reaction is robust at pH 5 to 8, at temperatures from +4°C to +37°C, in various buffer conditions (Ca²⁺/Mg²⁺ not needed) and in the presence of detergents. The reaction also occurs inside cells (*in vivo*). Since the speed of the coupling reaction is concentration dependent, it is recommended to use SpyCatcher2 undiluted.

All the different formats of SpyCatcher2 are compatible with Bio-Rad's recombinant HuCAL® antibodies with a SpyTag2 at the C-terminus of the heavy chain.

[Download SpyCatcher coupling protocol](#)

SpyCatcher2 (TZC001) is available conjugated to biotin (TZC001B). It is also available as a modified version, SpyCatcher2-CYS (TZC001CYS), which contains a cysteine residue for site-specific conjugation by the user.

SpyCatcher2-CYS can dimerize during storage by formation of a disulfide bond via the free cysteines. Before conjugation to these cysteines, the SpyCatcher2-CYS dimers should be mildly reduced e.g., by addition of 5mM DTT and incubation for 1 hr at room temperature, followed by a fast DTT removal step, e.g., by size exclusion chromatography using PD-10 desalting columns, as DTT can interfere with the subsequent conjugation chemistry. Dialysis is not recommended for DTT removal, as the cysteines can oxidize again during this slow process.

[View all available Catchers](#)

Protein Molecular Weight	Predicted 15687 Da
Purity	≥90% determined by SDS-PAGE under reducing conditions and visualized by coomassie blue staining
Instructions For Use	View SpyCatcher Coupling Protocol
References	<ol style="list-style-type: none">1. Keeble, A.H. <i>et al.</i> (2017) Evolving Accelerated Amidation by SpyTag/SpyCatcher to Analyze Membrane Dynamics. Angew Chem Int Ed Engl. 56 (52): 16521-5.2. Hentrich, C. <i>et al.</i> (2021) Periplasmic expression of SpyTagged antibody fragments enables rapid modular antibody assembly. Cell Chem Biol. 28 (6): 813-824.e6.
Storage	Store at -20°C only. Storage in frost-free freezers is not recommended. This product should be stored undiluted. Avoid repeated freezing and thawing as this may denature the protein. Should this product contain a precipitate we recommend microcentrifugation before use.
Guarantee	12 months from date of despatch
Acknowledgements	This product and/or its use is covered by claims of U.S. patents, and/or pending U.S. and

non-U.S. patent applications owned by or under license to Bio-Rad Laboratories, Inc. See [bio-rad.com/en-us/trademarks](https://www.bio-rad.com/en-us/trademarks) for details.

Health And Safety Information

Material Safety Datasheet documentation #20480 available at: <https://www.bio-rad-antibodies.com/SDS/TZC00120480>

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Regulatory

For research purposes only

Related Products

Recommended Useful Reagents

[SpyCatcher2:Biotin \(TZC001B\)](#)

[SpyCatcher2-CYS \(TZC001CYS\)](#)

[SpyTag3 PEPTIDE \(BLP086\)](#)

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