

## Datasheet: TZC040B

<b>Description:</b>	SPYCATCHER2:Biotin
<b>Name:</b>	SPYCATCHER2
<b>Format:</b>	Catcher-Biotin
<b>Product Type:</b>	Recombinant Protein
<b>Quantity:</b>	0.5 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](http://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.

<b>Target Species</b>	Protein/peptide tag
<b>Product Form</b>	Purified recombinant protein conjugated to Biotin - liquid
<b>Preparation</b>	Recombinant protein expressed in <i>E. coli</i> and purified by affinity chromatography
<b>Source</b>	E.coli
<b>Buffer Solution</b>	Phosphate buffered saline
<b>Preservative Stabilisers</b>	0.0095% MIT
<b>Approx. Protein Concentrations</b>	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 (Ca<sup>2+</sup>/Mg<sup>2+</sup> 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 is available conjugated to biotin. It is also available as a modified version, SpyCatcher2-CYS, 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](#)

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<b>Protein Molecular Weight</b>	Predicted molecular weight of the purified Catcher is 15904 Da The molecular weight specified excludes conjugation and will vary upon conjugation depending on the molecular weight of the label
<b>Purity</b>	≥90% determined by SDS-PAGE under reducing conditions and visualized by coomassie blue staining
<b>Instructions For Use</b>	<a href="#">View SpyCatcher Coupling Protocol</a>
<b>References</b>	1. Keeble, A.H. <i>et al.</i> (2017) Evolving Accelerated Amidation by SpyTag/SpyCatcher to Analyze Membrane Dynamics. <a href="#">Angew Chem Int Ed Engl. 56 (52): 16521-5.</a> 2. Hentrich, C. <i>et al.</i> (2021) Periplasmic expression of SpyTagged antibody fragments enables rapid modular antibody assembly. <a href="#">Cell Chem Biol. 28 (6): 813-824.e6.</a>
<b>Storage</b>	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.
<b>Guarantee</b>	12 months from date of despatch
<b>Acknowledgements</b>	This product and/or its use is covered by claims of U.S. patents, and/or pending U.S. and

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His-tag is a registered trademark of EMD Biosciences.

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**Health And Safety Information**      Material Safety Datasheet documentation #20480 available at: <https://www.bio-rad-antibodies.com/SDS/TZC040B>

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**Licensed Use**      For *in vitro* research purposes. Any re-sale in any form or any other commercial application needs a written agreement with Bio-Rad.

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**Regulatory**      For research purposes only

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## Related Products

### Recommended Useful Reagents

[ANTI SpyCatcher \(HCA379\)](#)

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

**Product inquiries:** [www.bio-rad-antibodies.com/technical-support](http://www.bio-rad-antibodies.com/technical-support)

To find a batch/lot specific datasheet for this product, please use our online search tool at: [bio-rad-antibodies.com/datasheets](http://bio-rad-antibodies.com/datasheets)

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