

Datasheet: TZC040PE

BATCH NUMBER 64754329

Description:	SPYCATCHER2:RPE
Name:	SPYCATCHER2
Format:	Catcher-RPE
Product Type:	Recombinant Protein
Quantity:	0.25 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
Flow Cytometry	▪			

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	Purified recombinant protein conjugated to R. Phycoerythrin (RPE) - liquid		
Max Ex/Em	Fluorophore	Excitation Max (nm)	Emission Max (nm)
	RPE 488nm laser	496	578
	RPE 561nm laser	546	578
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	145µM (2 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 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](#)

Protein Molecular Weight	Predicted molecular weight of the purified Catcher is 15904 Da
Purity	≥90% determined by SDS-PAGE under reducing conditions and visualized by coomassie blue staining
Instructions For Use	View SpyCatcher Coupling Protocol
References	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 for details. His-tag is a registered trademark of EMD Biosciences.
Health And Safety Information	Material Safety Datasheet documentation #20480 available at: https://www.bio-rad-antibodies.com/SDS/TZC040PE
Licensed Use	For <i>in vitro</i> research purposes. Any re-sale in any form or any other commercial application needs a written agreement with Bio-Rad.
Regulatory	For research purposes only

Product inquiries: 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](https://www.bio-rad-antibodies.com/datasheets)
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