Datasheet: TZC002CYS3

BATCH NUMBER 173380

Description:	BiSpyCatcher2-CYS3
Name:	BISPYCATCHER2
Format:	Catcher-CYS3
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

BiSpyCatcher2 purified recombinant protein, with three engineered cysteine residues; use for site-specific conjugation to a label of choice - liquid

Preparation

Recombinant protein expressed in *E. coli* and purified by affinity chromatography

Source

E.coli

Buffer Solution

Phosphate buffered saline

Preservative Stabilisers

0.0095% MIT
5 mM DTT
5 mM EDTA

Approx. Protein Concentrations

238 µM (7 mg/ml)

Product Information

BiSpyCatcher2 (H-BiSpyC2) is a 29 kDa homodimer of SpyCatcher2 (TZC001) bridged with a flexible linker. Each SpyCatcher2 subunit can form 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. BiSpyCatcher2 contains a His6-tag at the N-terminus. To avoid deamidation of asparagine, a N105D mutation was introduced (Hentrich *et al.* 2020) for each of the two Catchers.

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 BiSpyCatcher2 undiluted.

All the different formats of BiSpyCatcher2 are compatible with Bio-Rad's recombinant HuCAL® antibodies with a SpyTag2 at the C-terminus of the heavy chain.

Download BiSpyCatcher coupling protocol

BiSpyCatcher2 (TZC002) is available in two modified versions, BiSpyCatcher2-CYS (TZC002CYS) and BiSpyCatcher-CYS3 (TZC002CYS3), which contain one or three cysteine residues, respectively, for site-specific conjugation by the user.
BiSpyCatcher2 (TZC002) is also available conjugated to biotin (TZC002B) and HRP (TZC002P).

BiSpyCatcher2-CYS can dimerize by formation of a disulfide bond via the free cysteines. Before conjugation to these cysteines, the BiSpyCatcher2-CYS must be 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, as DTT can interfere with the conjugation chemistry.

BiSpyCatcher2-CYS3 contains DTT to avoid oligomerization through disulfide bond formation. Immediately before conjugation, to avoid oxidation and disulfide bond formation, carry out a fast DTT removal step, e.g., by size exclusion chromatography.

View all available Catchers

Protein Molecular Weight	Predicted 29469 Da
Purity	≥90% determined by SDS-PAGE under reducing conditions and visualized by coomassie blue staining
Instructions For Use	View BiSpyCatcher Coupling Protocol

References

1. Keeble, A.H. *et al.* (2017) Evolving Accelerated Amidation by SpyTag/SpyCatcher to Analyze Membrane Dynamics. Angew Chem Int Ed Engl. 56 (52): 16521-16525.
2. Hentrich, C. *et al.* (2021) Periplasmic expression of SpyTagged antibody fragments enables rapid modular antibody assembly. Cell Chem Biol. 28 (6): 813-824.e6.
3. Dauben, H. & Matić, I. (2025) Versatile Modular Antibodies for Sensitive and Specific Detection of Poly-ADP-Ribose bio-Rxiv 10 Fep [Preprint].
4. Dauben, H. *et al.* (2026) Versatile and sensitive detection of mono- and poly(ADP-

	ribosyl)ation reveals XRCC1-dependent remodelling of PARP1 signalling. Nat Commun. 17 (1): 3216.
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.
Health And Safety Information	Material Safety Datasheet documentation #20483 available at: https://www.bio-rad-antibodies.com/SDS/TZC002CYS3
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.
Regulatory	For research purposes only

Related Products

Recommended Useful Reagents

BiSpyCatcher2 (TZC002)
BiSpyCatcher2:Biotin (TZC002B)
BiSpyCatcher2-CYS (TZC002CYS)
BiSpyCatcher2:HRP (TZC002P)
SpyTag3 PEPTIDE (BLP086)

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
'M442432:250528'

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