Rapid Confirmation of Expressed Proteins in Gel with the TC-FIAsH[™] Expression Analysis Detection Kits

The TC- FlAsH™ Expression Analysis Detection Kits provide a rapid and easy method of detecting TC-tagged expression proteins. By directly staining the cell lysate with FlAsH dye and orange or red total protein stain, you can visualize the TC-tagged protein as well as total protein in contrasting colors without gel post-staining. Westerns are not needed to visualize your TC-tagged protein. However, you can perform western blotting, if desired. Rapid confirmation saves time, reagent, and money. TC-tagged proteins are easily visualized in the gel cassette (without removing the gel from the glass or plastic cassette) or even after removal of gel from the cassette, using a fluorescent imager or UV transilluminator with appropriate filter sets.

A linear relationship exists between relative fluorescence units and concentration of the detected protein, allowing you to perform quantitative determinations, if necessary. The TC-tag detection kits from Invitrogen are available for use in detecting TC-tagged proteins in cell or on a gel.

To learn more, visit www.invitrogen.com/TCFlashGelDetection

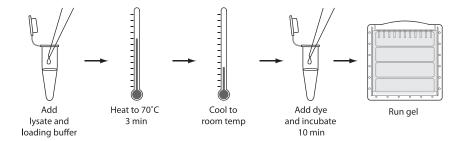


Figure 1. Experimental Protocol.

Advantages of the TC-Tag

The 6-amino acid tetracysteine (TC) tag is small so there is less protein structure and function disruption—which is important in studying your protein of interest. The TC-tag is readily fused to the N-or C-terminus, or an internal site of your protein and is easily detected using the biarsenical stains, FlAsH (488 nm: green) and ReAsH (593 nm: red).

Applications of the TC-Tag

The versatile TC-tag is useful for:

- Protein localization, turnover, and trafficking
- Receptor signaling and internalization
- Correlation of light microscopy with electron microscopy
- Pulse-chase and double-labeling experiments
- Enzyme activity studies

- SDS-PAGE detection of tagged proteins
- Affinity purification

Features of TC-FIAsH[™] Detection Kits

Tag size:

FlAsH dye detection: 488 nm / 520 nm or mid-UV

Total protein detection: Yes

Staining time: 13 minutes

Visualization: Within the gel cassette or after

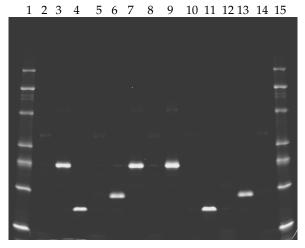
removal of gel from the cassette

Table 1. Detection sensitivity chart.

Fluorescent Dye	Wavelength	Protein
TC-FlAsH [™] (in the cassette)	488 nm	4 ng*
	UV	60 ng*
TC-FlAsH [™] (out of the cassette)	488 nm	250 pg*
	UV	1 ng*
Total protein (in the cassette)	Orange (532 nm)	100 ng**
	Red (620 nm)	100 ng**
Total protein (out of the cassette)	Orange (532 nm)	12 ng**
	Red (620 nm)	50 ng**

^{*}Indicates amount of TC-tagged protein

^{**}Indicates total amount of protein loaded



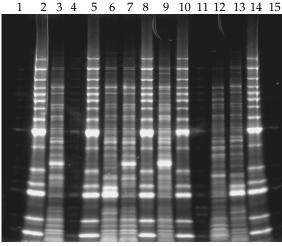


Figure 3. Purified TC-tagged proteins, crude lysates containing TC-tagged protein, and BenchMark[™] Protein Ladder were subjected to protein detection using the TC-FlAsH[™] Expression Analysis Detection Kit - Red (Cat. no. A10068). Samples were analyzed on a 4-20% Tris-Glycine SDS-PAGE gel and imaged using a Fuji FLA3000 laser scanner. With FIAsH detection (Panel A), only TC-tagged proteins appear with the Benchmark Fluorescent Protein Ladder while with total protein detection (Panel B), all proteins, including TC-tagged proteins are detected.

Panel A: TC-tagged proteins and Benchmark Fluorescent Protein Ladder visualized with 473 nm excitation with 520 nm long pass emission filter. **Panel B**: Total protein detection obtained with 633 nm excitation with 675 nm long pass emission filter. Lanes 1, 15: BenchMark Fluorescent Protein Ladder (4 µL); Lanes 2, 5, 8 14: BenchMark Protein Ladder (200 ng/band); Lanes 3, 7: Lysate expressing TC-tagged CFP protein (4 µL); Lane 4: Purified TC-tagged ACP protein (200 ng); Lanes 6, 13: Lysate expressing TC-tagged calmodulin protein (4 µL); Lane 9: Lysate expressing TC-tagged GFP protein (4 µL); Lane 10: BenchMark™ Protein Ladder (100 ng/band); Lane 11: Purified TC-tagged ACP protein (100 ng); Lane 12: E. coli lysate $(4 \mu L)$.

Product List Current prices are available from www.invitrogen.com or from our Customer Service Department

Catalog no.	Product Name	Unit Size
A10067	TC-FlAsH™ Expression Analysis Detection Kit - Orange *orange fluorescent in-gel detection of TC-tagged and total protein*	1 kit
A10068	TC-FlAsH™ Expression Analysis Detection Kit - Red *red fluorescent in-gel detection of TC-tagged and total protein*	1 kit

Contact Information

Molecular Probes, Inc.

29851 Willow Creek Road Eugene, OR 97402 Phone: (541) 465-8300 Fax: (541) 335-0504

Customer Service:

6:00 am to 4:30 pm (Pacific Time) Phone: (541) 335-0338 Fax: (541) 335-0305 probesorder@invitrogen.com

Toll-Free Ordering for USA:

Order Phone: (800) 438-2209 Order Fax: (800) 438-0228

Technical Service:

8:00 am to 4:00 pm (Pacific Time) Phone: (541) 335-0353 Toll-Free (800) 438-2209 Fax: (541) 335-0238 probestech@invitrogen.com

Invitrogen European Headquarters

Invitrogen, Ltd. 3 Fountain Drive Inchinnan Business Park Paisley PA4 9RF, UK Phone: +44 (0) 141 814 6100 Fax: +44 (0) 141 814 6260 Email: euroinfo@invitrogen.com Support: eurotech@invitrogen.com

For country-specific contact information, visit www.invitrogen.com

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