Fold ‘n’ Glow” Split GFP Detection Reagent

Product Number **#21004001**

PRODUCT INSERT

INTENDED USE: *FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES*

The split GFP system is a simple tagging and detection technique that can be used to quantify the expression level of an S11-tagged protein to determine the solubility of a protein, it’s domain or protein aggregation[1].

When incubated with a protein of interest (POI) terminally tagged with wild type GFP stand 11, the S1-10 detection reagent provides a time-dependent increase in fluorescence that allows for the determination of the amount of protein that is properly folded in a given sample as the folding reporter gives a signal directly proportional to the amount of correctly folded protein[2]. GFP fluorescence is readily detectable by fluorescence microscopy, providing direct visual evidence of complementation of GFP 1-10 and POI-linker-GFP S11 within 15 minutes after induction of the GFP 1-10 detector strand (*Fig. 1*).

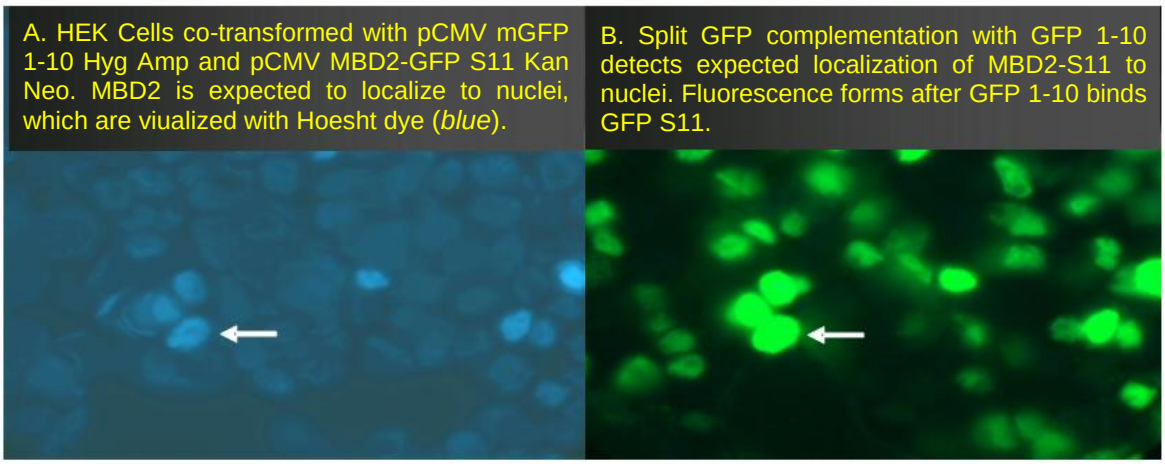

Figure 1. Using the split GFP mammalian system to follow nuclear localization. HEK cells were co-transfected with pCMV MBD2-GFP Cterm S11 Kan Neo and pCMV-mGFP 1-10 Hyg Amp. GFP S11 is the 16 amino acid strand 11 peptide of GFP that is detected by the GFP 1-10 by complementation to form fluorescent GFP. MBD2 is known to translocate to nuclei (*Fig 1A*). GFP 1-10 complements the GFP S11 tag, and the resulting GFP fluorescence is translocated to the nuclei (*Fig 1B)*. Since MBD2 is expressed with only the short GFP S11 tag, and subsequently complemented with GFP 1-10, there is minimal folding perturbation compared to expressing MBD2-GFP as a direct full-length GFP fusion. Nuclear fluorescence is bright and non-punctate as expected.

S1-10 Universal Detection Reagent is provided as a ready-to-use solution (ranging from 3.0-3.5 mg/mL) for use with either of the GFP “Fold ‘n’ Glow” Solubility Assay Kits provided by Sandia Biotech.

Note: *It is recommended to store this product at -20℃ or to re-aliquot reagents to smaller working volumes to avoid repeated freeze-thawing and store at -70℃. When stored properly, this reagent is stable until the date indicated either on the box or each component.*

DATA ANALYSIS

Data Analysis

Subtract the blank fluorescence values from the final fluorescence values of the sample(s) and the positive control. Estimate protein concentration by comparing fluorescence on the standard curve.

REFERENCES

1. Cabantous et al. "Protein tagging and detection with engineered self-assembling fragments of green fluorescent protein," *Nature Biotechnology* **23,** 102-107, December 2004.
2. Waldo et al. "Rapid protein-folding assay using green fluorescent protein," *Nature Biotechnology* 17, 691 - 695, July 1999.

Suggested Plate Configuration

	1	2	3	4	5	6	7	8	9	10	11	12
A	**Neat positive control**	**Neat positive control**	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown
B			Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown
C			Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown
D			Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown
E			Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown
F			Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown
G			Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	**Blank**	**Blank**
H	**.39nM positive control**	**.39nM positive control**	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	Unknown	**BSA negative control**	**BSA negative control**

For Technical Support and Customer Service: (505) 342-0224 / (866) 342-0224

Sandia BioTech, Inc. • 609 Broadway, NE • Albuquerque • New Mexico• 87102

www.SandiaBio.com

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