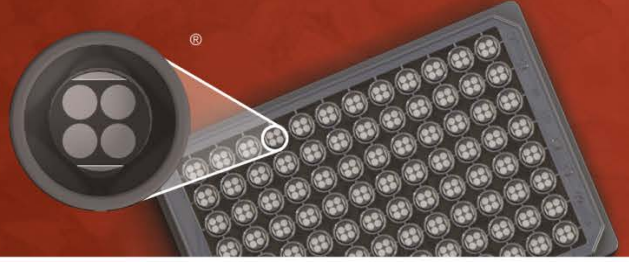
MSD® Total 4E-BP1 Kit

For quantitative determination in human whole cell lysate samples



Alzheimer's Disease
BioProcess
Cardiac
Cell Signaling
Clinical Immunology
Cytokines
Growth Factors
Hypoxia
Immunogenicity
Inflammation
Metabolic
Oncology
Toxicology
Vascular

Catalog Numbers

Total 4E-BP1 Kit	
Kit Size	**Catalog #**
1 plate	K151OLD-1
5 plates	K151OLD-2
25 plates	K151OLD-4

Ordering Information

MSD Customer Service
Phone: 1-301-947-2085
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Email: CustomerService@mesoscale.com

Scientific Support

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Company Address

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1. 4E-BP1
2. *BSA blocked*
3. *BSA blocked*
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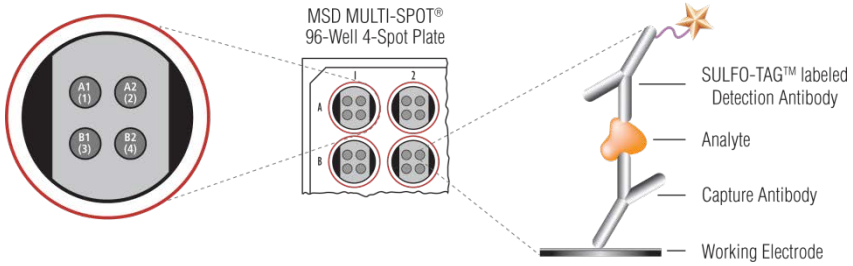
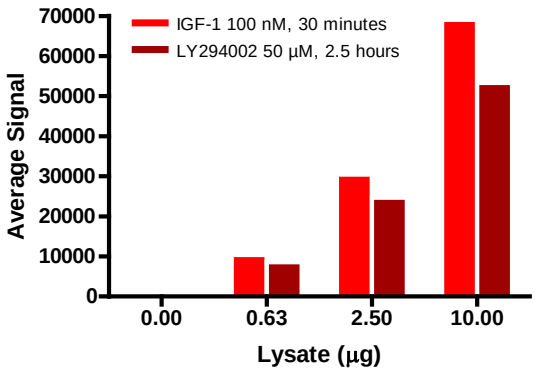


Eukaryotic translation initiation factor (eIF) 4E-binding protein-1 (4E-BP1) is a translational repressor protein that plays a critical role in the control of protein synthesis, survival, and cell growth.[1] During cap-dependent translation, eIF4E binds to the mRNA cap structure and promotes formation of the eIF4F initiation complex and ribosome binding. Non-phosphorylated 4E-BP1 binds eIF4E and impedes formation of the initiation complex, blocking translation and favoring apoptosis.[1,2] When 4E-BP1 is phosphorylated, however, its affinity for eIF4E is reduced, allowing eIF4E to interact with the cap complex and initiate translation. 4E-BP1 has multiple phosphorylation sites and is most often phosphorylated through the mammalian target of rapamycin (mTOR) signaling pathway, although several other kinases have also been shown to phosphorylate this key repressor (cyclin-dependent kinase 1, P13K-Akt, and ERK1/2).[1-3] Phosphorylated 4E-BP1 expression in breast, ovary, and prostate tumors has been shown to be associated with tumor growth and malignant progression.[1,4] Thus, phosphorylated 4E-BP1 may prove a highly relevant biomarker in oncogenesis, and a better understanding of the signaling pathways using this molecule may enhance the development of anti-cancer therapeutics and targets. The MSD Total 4E-BP-1 assay is available on 96-well, 4-spot plates. This datasheet outlines the performance of the assay.

Typical Data

Representative results for the Total 4E-BP1 Kit are illustrated below. The signal and ratio values provided are examples; individual results will vary depending upon the samples tested. Western blot analyses of each lysate type are shown for comparison.

Growing MCF-7 cells were treated with 50 nM IGF-1 for 30 minutes (positive) or with 50 µM LY294002 for 2.5 hours (negative). A dilution series of positive and negative lysates was assayed using the Total 4E-BP1 assay. Whole cell lysates were added to MSD MULTI-SPOT 4-spot plates coated with anti-total 4E-BP1 antibody on one of the four spatially distinct electrodes in each well. 4E-BP1 was detected with anti-total 4E-BP1 antibody conjugated with MSD SULFO-TAG.



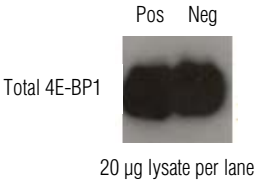


Figure 1: Sample data generated with Total 4E-BP1 assay. Increased signal is observed with the titration of positive and negative cell lysates. The Total 4E-BP1 assay provides a quantitative measure of the data obtained with the traditional Western blot.

Lysate Titration

Data for positive and negative cell lysates using the Total 4E-BP1 Kit are presented below.

Lysate (µg)/well	Positive			Negative			P/N
	Average Signal	StdDev	%CV	Average Signal	StdDev	%CV	
0	662	44	6.6	669	80	11.9	
0.63	10 359	186	1.8	8 533	478	5.6	1.2
2.5	30 436	913	3.0	24 645	1 183	4.8	1.2
10	69 054	898	1.3	53 312	4 475	8.9	1.3

For a complete list of products, please visit our website at www.mesoscale.com.

The MSD Advantage

- **Multiplexing:** Multiple analytes can be measured in one well using typical sample volumes of 25 µL or less without compromising speed or performance
- **Large dynamic range**: Linear range of up to five logs enables the measurement of native levels of biomarkers in normal and diseased samples without multiple dilutions
- **Minimal background:** The stimulation mechanism (electricity) is decoupled from the response (light signal), minimizing matrix interference
- **Simple protocols:** Only labels bound near the electrode surface are excited, enabling assays with fewer washes
- **Flexibility:** Labels are stable, non-radioactive, and conveniently conjugated to biological molecules

References

1. Armengol G, et al. 4E-binding protein 1: a key molecular "funnel factor" in human cancer with clinical implications. Cancer Res. 2007 Aug 15;67(16):7551-5.
2. Jackson RJ, Wickens M. Translational controls impinging on the 5'-untranslated region and initiation factor proteins. Curr Opin Genet Dev. 1997 Apr;7(2):233-41.
3. Asnaghi L, et al. mTOR: a protein kinase switching between life and death. Pharmacol Res. 2004 Dec;50(6):545-9.
4. Pons B, et al. The effect of p-4E-BP1 and p-eIF4E on cell proliferation in a breast cancer model. Int J Oncol. 2011 Nov;39(5):1337-45.