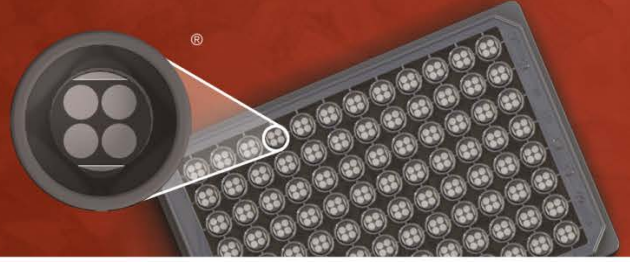
MSD® CHOP Kit

For quantitative determination in human, mouse, and rat 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

CHOP Kit	
Kit Size	**Catalog #**
1 plate	K150QJD-1
5 plates	K150QJD-2
25 plates	K150QJD-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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Email: ScientificSupport@mesoscale.com

Company Address

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Rockville, MD 20850-3173 USA
www.mesoscale.com®

For Research Use Only. Not for use in diagnostic procedures.

1. CHOP
2. *BSA blocked*
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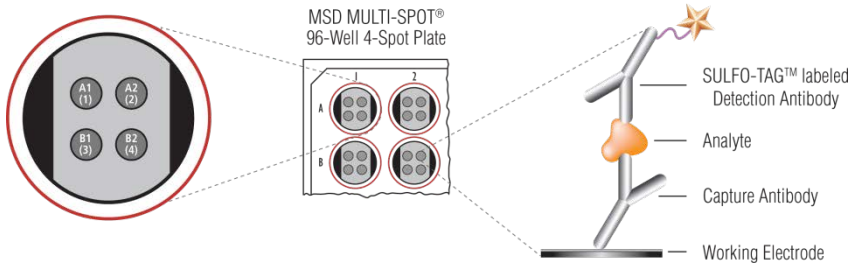

C/EBP Homology Protein (CHOP), also known as Growth Arrest and DNA Damage Inducible Protein 153 (GADD153), DNA–damage-inducible transcript 3 (DDIT3) and C/EBPz, is a transcription factor that mediates one of the three arms of unfolded protein response (UPR) adaptation to endoplasmic reticulum (ER) stressors. The accumulation of unfolded or misfolded proteins in the ER is a threat to cell survival and results in a condition known as ER-stress.[1,2] To overcome this stress, the ER initiates specific signaling pathways encompassed by the ER stress response.[3] Among these are translational attenuation, upregulation of ER chaperone proteins and proteins that facilitate folding, activation of NFkB signaling, and, as a last resort, the induction of apoptosis via CHOP.[4-7] CHOP is a 29 kDa protein that serves as a dominant negative inhibitor of C/EBPs.[8] CHOP is composed of an amino-terminal transcription activation domain and a carboxy-terminal basic amino-acid-rich DNA-binding domain and a leucine zipper dimerization domain. CHOP is able to act as a dominant negative inhibitor of C/EBP transcriptional activation by forming a heterodimer with C/EBPs that are normally active homodimers. The heterodimer has reduced DNA binding activity due to several key proline and glycine substitutions in the basic amino-acid-rich DNA-binding domain of CHOP. CHOP is present at low levels in the cytosol under non-stressed conditions. During ER stress, the ER stress-activated kinase PERK phosphorylates eIF2α causing a decrease in its activity that increases the translation of ATF4 mRNA. ATF4 binds to and activates the CHOP promoter thereby increasing its levels during ER stress.[9] While the precise role of CHOP in ER stress-induced apoptosis is not completely understood, one of the most widely cited mechanisms is via the suppression of the pro-survival protein Bcl-2.[10] CHOP is involved in the response of many diseases (ischemia, neurodegeneration, cancer, diabetes) which makes this an attractive marker for many therapeutic areas. The MSD CHOP assay is available on 96-well, 4-spot plates. This datasheet outlines the performance of the assay.

Typical Data

Representative results for the CHOP 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.

This assay was developed using recombinant human CHOP protein (data shown below) as well as with the nuclear fraction of rat hepatoma cells treated with ER stress-inducing agents, thapsigargin and tunicamycin (data not shown). Recombinant protein or nuclear cell lysate fractions were added to MSD MULTI-SPOT 4-spot plates coated with anti-CHOP antibody on one of the four spatially distinct electrodes in each well. CHOP was detected with anti-CHOP antibody conjugated with MSD SULFO-TAG reagent.

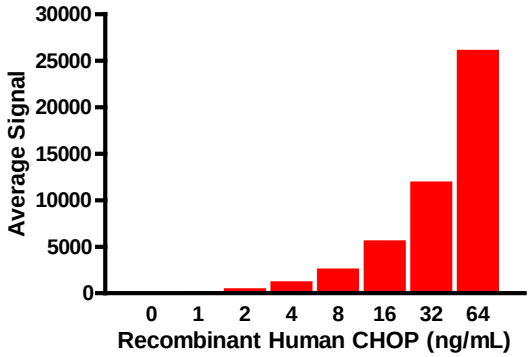

Figure 1: Sample data generated with CHOP assay. Increased signal is observed with the titration of recombinant human CHOP protein.

Sample Titration

Data for recombinant CHOP protein using the CHOP Kit are presented below.

Sample (ng/well)	CHOP recombinant protein		
	Average Signal	StdDev	%CV
0	52	2	4.1
1.0	399	13	3.2
2.0	754	21	2.8
4.0	1509	37	2.5
8.0	2888	150	5.2
16	5900	122	2.1
32	12244	332	2.7
64	26383	268	1.0

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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

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