# MSD<sup>®</sup> Phospho-Histone H3 (Ser10) Assay Whole Cell Lysate Kit

For quantitative determination in human, mouse, and rat whole cell lysate samples



Alzheimer's Disease BioProcess Cardiac Cell Signaling

Clinical Immunology Cytokines Hypoxia

Immunogenicity Inflammation Metabolic Oncology Toxicology Vascular

## Catalog Numbers

Phospho-Histone H3 (Ser10) Assay: Whole Cell Lysate Kit					
Kit size					
1 plate	K150EWD-1				
5 plates	K150EWD-2				
20 plates	K150EWD-3				

	Phospho-Histone H3 (Ser10) Whole Cell Lysate Set					
200 μ <b>g</b>	C10EW-1					

#### Ordering information

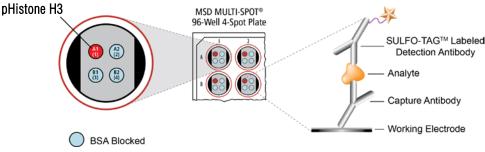
MSD Customer Service Phone: 1-301-947-2085 Fax: 1-301-990-2776 Email: CustomerService@ mesoscale.com

### **Company Address**

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**Histone H3**, along with H2a, H2b, H1/H5, and H4, is one of the five main classes of histones, which are found in the nucleus of eukaryotic cells, and are the main building blocks of chromatin. Histone H3 has an amino terminal tail and post-translational modification of this tail—an epigenetic modification—is involved in chromatin relaxation and transcriptional regulation.<sup>1</sup> Histones undergo many different types of post translational modifications, such as acetylation, methylation, phosphorylation, ubiquitylation, and sumoylation.<sup>2</sup> These modifications can change the chromatin structure and allow for binding of transcription factors to the DNA.<sup>3</sup>

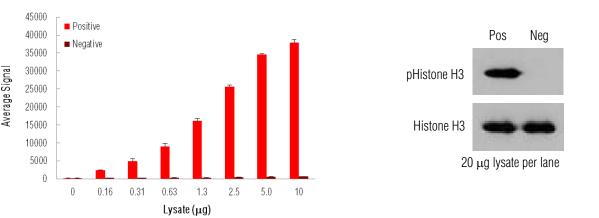
Acetylation of lysine residues on histones is frequently associated with actively transcribed regions of the genome, whereas methylation of lysine is seen both at transcribed and transcriptionally repressed areas of the genome.<sup>4</sup> Epigenetic modifications, of which histone post-translational modification is one example, have been shown to play a role in cancer, embryonic stem cell function, and mammalian development.<sup>5</sup>

The MSD Phospho-Histone H3 (Ser10) Assay is available on 96-well 4-Spot plates. This datasheet outlines the performance of the assay.

#### Typical Data

Representative results for the Phospho-Histone H3 (Ser10) Assay are illustrated below. The signal and ratio values provided below are example data; individual results may vary depending upon the samples tested. Western blot analyses of each lysate type were performed with phospho-Histone H3 (Ser10) and total Histone H3 antibodies and are shown below for comparison.

Growing HeLa cells (negative) were treated with nocodazole (1µg/mL; 19 hours) and calyculin A (50 nM; final 30 minutes) (positive). Whole cell lysates were added to MSD MULTI-SPOT 4-Spot plates coated with an anti-total Histone H3 antibody on one of the four spatially distinct electrodes per well. Phosphorylated Histone H3 was detected with anti-phospho-Histone H3 (Ser10) antibody conjugated with MSD SULFO-TAG<sup>™</sup> reagent.



**Fig. 1:** Sample data generated with the MULTI-ARRAY<sup>®</sup> Phospho-Histone H3 (Ser10) Assay. Increased signal is observed with the titration of pHistone H3 positive cell lysate. Signal for negative lysate remains low throughout the titration. The Phospho-Histone H3 (Ser10) Assay provides a quantitative measure of the data obtained with the traditional Western blot.





### Lysate Titration

Data for pHistone H3 positive and negative HeLa cell lysates using the MULTI-ARRAY Phospho-Histone H3 (Ser10) Assay are presented below.

Lysate	Positive			Negative			P/N
(µg)	Average Signal	StdDev	%CV	Average Signal	StdDev	%CV	F/N
0	228	9	3.7	231	15	6.3	
0.16	2445	115	4.7	269	15	5.5	9.1
0.31	4881	767	15.7	299	7	2.2	16
0.63	9006	855	9.5	332	7	2.1	27
1.3	16194	543	3.4	341	18	5.2	47
2.5	25663	399	1.6	451	42	9.3	57
5.0	34552	230	0.7	575	36	6.3	60
10	37864	903	2.4	676	31	4.6	56

#### MSD Advantage

- Multiplexing: Multiple analytes can be measured in one well using typical sample amounts of 25 µg/well 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 signal (light)
- Simple protocols: Only labels near the electrode surface are detected, enabling no-wash assays
- > Flexibility: Labels are stable, non-radioactive, and conveniently conjugated to biological molecules
- > High sensitivity and precision: Multiple excitation cycles of each label enhance light levels and improve sensitivity

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#### **References:**

- 1. Strahl B, Allis CD. The language of covalent histone modifications. Nature. 2000 Jan 6;403(6765):41-5.
- 2. Peterson CL, Laniel MA. Histones and histone modifications. Curr Biol. 2004 Jul 27;14(14):R546-51.
- 3. Taverna SD, Li H, Ruthenburg AJ, Allis CD, Patel DJ. How chromatin-binding modules interpret histone modifications: lessons from professional pocket pickers. Nat Struct Mol Biol. 2007 Nov;14(11):1025-40. Epub 2007 Nov 5.
- 4. Verdone L, Caserta M, Mauro E. Role of histone acetylation in the control of gene expression. Biochem Cell Biol. 2005 Jun;83(3):344-53.
- 5. Spivakov M, Fisher AG. Fisher Epigenetic signatures of stem-cell identity. Nat Rev Genet. 2007 Apr;8(4):263-71.

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