

## Oxidized Protein Control (Carbonyl-BSA)

**CATALOG NUMBER:** STA-309

**STORAGE:** -20°C

**QUANTITY AND CONCENTRATION:** 200 µL of 50 µg/mL Carbonyl-BSA in 1X PBS

**SHELF LIFE:** 1 year from receipt under proper storage conditions; aliquot to avoid multiple freeze thaw cycles

### Background

Protein oxidation is defined as the covalent modification of a protein induced either directly by reactive oxygen species or indirectly by reaction with secondary by-products of oxidative stress. Oxidative modification of proteins can be induced in vitro by a wide array of pro-oxidant agents and occurs in vivo during aging and in certain disease conditions.

There are numerous types of protein oxidative modifications. The most common products of protein oxidation in biological samples are the protein carbonyl derivatives of Pro, Arg, Lys, and Thr. These derivatives are chemically stable and serve as markers of oxidative stress for most types of ROS. Many of the assays involve derivitization of the carbonyl group with dinitrophenylhydrazine (DNPH), which leads to formation of a stable dinitrophenyl hydrazone product, followed by immunoblotting with anti-DNP antibody.

### Methods

Dilute the Carbonyl-BSA control with reducing SDS-PAGE sample buffer to 5 µg/mL and heat for 5 min in boiling water. Load 20 µL per lane for western blot analysis of protein carbonyl using Cell Biolabs Protein Carbonyl Immunoblot Kit (STA-308) or anti-DNP antibody after DNPH derivitization.

### Example of Results

The following figures demonstrate typical results. One should use the data below for reference only. This data should not be used to interpret actual results.



**Figure 1: Immunoblotting of Oxidized BSA.** Carbonyl-BSA was first electroblotted onto nitrocellulose membrane. Following the electroblotting procedure, the membrane was incubated with (right strip) or without (left strip) DNPH solution. The derivatized Carbonyl-BSA is detected by immunoblotting with anti-DNP antibody as described in the Assay Protocol of Protein Carbonyl Immunoblot Kit (Cat. #STA-308).

## References

1. Cadenas, E., Boveris, A., Ragan, CI., and Stoppani, AO. (1977). *Archives of Biochemistry & Biophysics* 248–257.
2. Wakeyama, H., Takeshige, K., Takayanagi, R., and Minakami, S. (1982). *Biochem J* 593–601.
3. Talent, JM., Kong, Y., and Gracy, RW. (1998). *Anal.Biochem.* 31–38.

## Recent Product Citation

Biswas, C. et al. (2014). Nuclear heme oxygenase-1 (HO-1) modulates subcellular distribution and activation of Nrf2, impacting metabolic and anti-oxidant defenses. *J Biol Chem.* **289**:26882-26894.

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