Native E.coli dsbA

Cat. No. dsbA-8328E **Lot. No.** (See product label)

SPECIFICATION

Product Overview	Disulfide Oxidoreductase produced in E.coli is periplasmic protein isolated from E.coli, containing 208 amino acids having a molecular mass of 23,149 Dalton. The DsbA is purified by chromatographic techniques.
Species	Rat
Source	E.coli
Description	DsbA is a disulfide oxidoreductase containing a thioredoxin domain with an inserted helical domain of unknown function. Like other thioredoxin-based enzymes, DsbA"s catalytic site is a CXXC motif (CPHC in E. coli DsbA). The pair of cysteines may be oxidized (forming an internal disulfide) or reduced (as free thiols), and thus allows for oxidoreductase activity by serving as an electron pair donor or acceptor, depending on oxidation state. This reaction generally proceeds through a mixed-disulfide intermediate, in which a cysteine from the enzyme forms a bond to a cysteine on the substrate. DsbA is responsible for introducing disulfide bonds into nascent proteins. In equivalent terms, it catalyzes the oxidation of a pair of cysteine residues on the substrate protein. Most of the substrates for DsbA are eventually secreted, and include important toxins, virulence factors, adhesion machinery, and motility structures DsbA is localized in the periplasm, and is more common in Gram-negative bacteria than in Gram-positive bacteria. Within the thioredoxin family, DsbA is the most strongly oxidizing. Using glutathione oxidation as a metric, DsbA is ten times more oxidizing than protein disulfide-isomerase (the eukaryotic equivalent of DsbA). The extremely oxidizing nature of DsbA is due to an increase in stability upon

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	reduction of DsbA, thereby imparting a decrease in energy of the enzyme when it oxidizes substrate. This feature is incredibly rare among proteins, as nearly all proteins are stabilized by the formation of disulfide bonds. DsbA"s highly oxidizing nature is a result of hydrogen bond, electrostatic and helix-dipole interactions that favour the thiolate over the disulfide at the active site.
Form	The protein was lyophilized after from sterile solution containing 50mM sodium phosphate buffer and 100mM sodium chloride.
AA Sequence	The sequence of the first five N-terminal amino acidswas determined and was found to be Met-Ly-Lys-Ala-Trp.
Purity	Greater than 95.0% as determined by RP-HPLC and analysis by SDS-PAGE
Applications	Suitable for use in Western Blot. If this protein is to be used for Western Blot analysis, we recommend that the material be diluted in 1X SDS-PAGE sample buffer (1). On 15-well minigel system, 50ng of protein per lane should be sufficient when used in colorimetric Western Blot with D9625 at dilution of 1:10,000 as the primary antibody and an appropriate alkaline phosphatase conjugated secondary antibody for detection.
Notes	For research use only.
Storage	Lyophilized DsbA although stable at room temperature for weeks, should be stored desiccated below -18 ℃. Upon reconstitution DsbA should be stored at 4 ℃ between 2-7 days and for future use below -18 ℃. For long term storage it is recommended to add carrier protein (0. 1% HSA or BSA). Please prevent freeze/thaw cycles.
Reconstitution	It is recommended to reconstitute the lyophilized DsbA in sterile H_2O not less than

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	100 μg/ml, which can then be further diluted to other aqueous solutions.

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