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ProductInformation

Thioredoxin reductase
from *E. coli*

Product Number **T7915**
Storage Temperature 2-8 °C

E.C. 1.8.1.9 (formerly 1.6.4.5)
CAS# 9074-14-0
Synonyms: Thioredoxin-disulfide reductase; NADPH:oxidized thioredoxin oxidoreductase

Product Description

Thioredoxin reductase from *E. coli* has a native molecular weight of 70 kDa (two 35 kDa monomers);[1] however, after hydroxylamine agarose chromatography, two bands are observed at 34 kDa and 46 kDa (SDS-PAGE).[1] The enzyme has a single peak when analyzed by gel filtration chromatography.[1] This product exhibits two bands at approximately 35 kDa and 42 kDa (SDS-PAGE) and is homogeneous by gel filtration.

Thioredoxin reductase is a FAD containing enzyme. Its mechanism involves the transfer of reducing equivalents from NADPH to a disulfide bond in the enzyme within the sequence Cys-Ala-Thr-Cys via the FAD moiety.[2] The thioredoxin reductase subunit contains binding domains for FAD and NADPH, which are different from those found in glutathione reductase. There appears to be a large conformational change in thioredoxin reductase during enzymatic reduction of thioredoxin (TRX).

The reduction of ribonucleotides, which is essential for DNA synthesis, in *E. coli* is carried out by the enzyme ribonucleotide diphosphate reductase (E.C. 1.17.4.1).[1] This reduction reaction is coupled *in vitro* to the oxidation of NADPH via the small protein thioredoxin and thioredoxin reductase. This reduction system is present in many procaryotic and eucaryotic cells, and is virtually universal.

The reduction reaction is shown below:

$$\text{TRX-S-S} + \text{NADPH} + \text{H}^+ \xleftrightarrow{\text{Thioredoxin reductase}} \text{TRX-(SH)}_2 + \text{NADP}^+$$

$$\text{TRX-(SH)}_2 + \text{protein} \xleftrightarrow{\text{Spontaneous}} \text{Trx-S-S} + \text{protein-(SH)}_2$$

The *E. coli* reductase is specific for TRX from *E. coli*. In contrast, the mammalian thioredoxin reductase is larger, approximately 116 kDa, and it will reduce other compounds such as selenite,[3] selenoglutathione, alloxan, and DTNB. Mammalian thioredoxins show a 25% sequence homology to the *E. coli* thioredoxin.

This product is supplied as a yellow suspension in 3.6 M $(NH_4)_2SO_4$ containing 30 mM potassium phosphate buffer, pH 7.5, and 2 mM EDTA.

Specific Activity: Minimum 25 units/mg protein (Bradford)

Unit definition: One unit will cause an increase in absorbance of 1.0 at 412 nm (when measured in a coupled assay with *E. coli* thioredoxin and DTNB) per minute per ml at pH 7.0 at 25 °C.

Precautions and Disclaimer

This product is for laboratory research use only. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

It is recommended to store the product at 2-8 °C. The suspension is stable for at least 2 years. This product retains 100% activity after 7 days at 37 °C.

References

1. Pigiet, V.P., and Conley, R.R., J. Biol. Chem., **252**, 6367-6372 (1977).
2. Holmgren, A., and Bjornstedt, M., Methods in Enzymol., **252**,199-208 (1995).
3. Bjornstedt, M., et al., Methods in Enzymol., **252** 209-219 (1995).

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