abcam

Product datasheet

Thioredoxin Reductase Assay Kit (Colorimetric) ab83463

28 References 5 图像

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说明

产品名称 Thioredoxin Reductase Assay试剂盒(Colorimetric)

检测方法 Colorimetric

样品类型 Urine, Cell culture extracts, Other biological fluids, Tissue Extracts

检测类型 Enzyme activity

检测时间 0h 40m

产品概述 Thioredoxin Reductase Assay Kit (ab83463) is a specific assay for detecting Thioredoxin

Reductase (TrxR) activity.

In the thioredoxin reductase assay protocol, TrxR catalyzes the reduction of DTNB to TNB^{2-} in the presence of NADPH, which generates a strong yellow color (ODmax = 412 nm).

Other enzymes present in crude biological samples such as glutathione reductase and glutathione peroxidase can also reduce DTNB. In order to measure TrxR-only activity, a TrxR specific inhibitor is used in a separate reaction to determine TrxR specific activity. The difference between total DTNB reduction in the sample and DTNM reduction in the sample in presence of TrxR inhibitor is the value of specific TrxR activity in the sample.

We have tested the GR positive control from <u>ab83461</u> in the conditions of ab83463, and we do not observe activity of the GR positive control.

Thioredoxin reductase assay protocol summary:

- add samples and standards to wells
- add reaction mix
- analyze with a microplate reader over 20 min

This product is manufactured by BioVision, an Abcam company and was previously called K763 Thioredoxin Reductase Activity Colorimetric Assay Kit. K763-100 is the same size as the 100

test size of ab83463.

Thioredoxin reductase (TrxR, EC 1.8.1.9) is a ubiquitous mammalian enzyme that catalyzes the NADPH-dependent reduction of the redox protein thioredoxin, as well as of other endogenous and exogenous compounds such as selenite, lipid hydroperoxides and hydrogen peroxide.

平台 Microplate reader

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存放说明

Store at -20°C. Please refer to protocols.

组 件	100 tests
DTNB	1 vial
NADPH I	1 vial
Thioredoxin Reductase Assay Buffer	1 x 25ml
Thioredoxin Reductase Inhibitor	1 vial
Thioredoxin Reductase Positive Control	1 vial
TNB Standard	1 vial

相关性

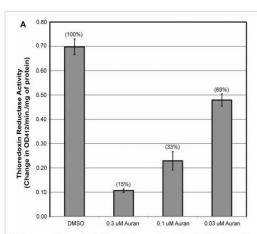
Thioredoxin reductase (TrxR) (EC 1.8.1.9) is a ubiquitous enzyme which is involved in many cellular processes such as cell growth, p53 activity, and protection against oxidation stress, etc. The mammalian TrxR reduces thioredoxins as well as non-disulfide substrates such as selenite, lipoic acids, lipid hydroperoxides, and hydrogen peroxide.

细胞定位

 $TXNRD1: Cytoplasmic.\ TXNRD2: Cytoplasmic.\ Nuclear.\ Microsome.\ Endoplasmic\ reticulum.$

TXNRD3: Mitochondrial.

图片



Bulman CA et al. PLoS One. 9:e0003534 (2015)

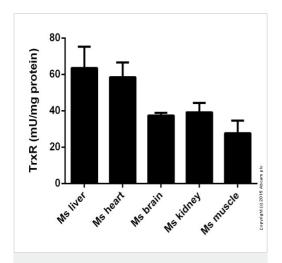
Functional Studies - beta Thioredoxin reductase Assay Kit (ab83463)

Image from Bulman CA et al., PLoS One. 2015;9(2):e0003534. Fig 4(A).; doi: 10.1371/journal.pntd.0003534. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

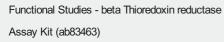
Activity of endogenous Brugia thioredoxin reductase from soluble worm lysates following incubation with 1% DMSO or 0.3 μ M, 0.1 μ M, or 0.03 μ M of auranofin *in vitro*. Percentages indicate the percent activity of TrxR compared to DMSO controls. Thioredoxin reductase activity was significantly reduced (p < 0.05) to 15%, 33% and 69% of endogenous activity, respectively, compared to the activity in DMSO-treated worms.

Thioredoxin reductase activity of worm lysates was assayed using female $B.\ malayi$ treated $in\ vitro$ with either 0.3 μ M, 0.1 μ M, or 0.03 μ M auranofin or 1% DMSO. After 5 hours of treatment, worm motility was measured using the Worminator, and then worms (24 in each group) were pooled, washed three times in PBS, and lysed by douncing in a glass homogenizer in assay buffer (ab83463) with 1 mM PMSF. The crude lysates were centrifuged at 10,000 rcf for 15 minutes at 4°C to pellet insoluble material. The total protein concentrations of soluble lysates were measured using the Bradford assay. The soluble lysates were incubated for 20 minutes in assay buffer or assay buffer with a proprietary thioredoxin reductase specific inhibitor before adding a specific substrate, DTNB (5, 5′-dithiobis (2-nitrobenzoic) acid), and measuring activity at 20 second

intervals for 40 minutes using the SpectraMax Plus Microplate Reader (Molecular Devices, Sunnyvale, CA) at λ = 412 nm. Lysates were tested in duplicate. TrxR activity was calculated based on the linear amount of TNB produced per minute per mg of total protein and adjusted for background activity from enzymes other than TrxR in the lysates.



Thioredoxin reductase measured in mouse tissue lysates showing activity (mU) per mg protein of sample tested

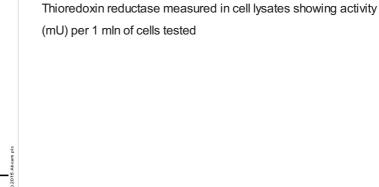


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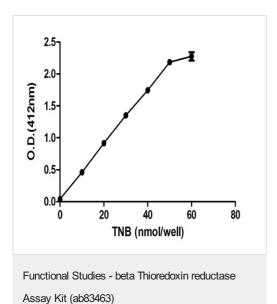
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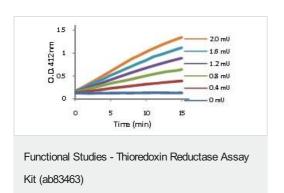
TrxR (mU/10⁶ cells)



Functional Studies - beta Thioredoxin reductase Assay Kit (ab83463)



Standard curve (colourimetric): mean of duplicates (+/-SD) with background readings substracted



Thioredoxin reductase Kinetic Data using ab83463.

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