abcam

Product datasheet

Glutathione Assay Kit (Fluorometric) ab65322

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Overview

Product name Glutathione Assay Kit (Fluorometric)

Detection method Fluorescent

Sample type Urine, Serum, Plasma, Other biological fluids, Tissue Extracts, Cell Lysate, Cell culture media

Assay type Quantitative
Assay time 2h 00m

Species reactivity Reacts with: Mammals, Other species

Product overview Glutathione Assay Kit (Fluorometric) (ab65322) provides a simple *in vitro* assay for detection of

total glutathione changes during cellular response to toxicity, apoptosis and other conditions. The assay uses the dye monochlorobimane (MCB), which forms an adduct with glutathione in a reaction catalyzed by glutathione-S-Transferase (GST). The unbound MCB is almost nonfluorescent, whereas it emits a fluorescent blue light (Ex/Em = 380nm/461nm) when bound to reduced or oxidized glutathione. Thus, the amount of glutathione can be easily detected using a

fluorometer or a 96-well fluorometric plate reader.

Visit our FAQs page for tips and troubleshooting.

Notes This product is manufactured by BioVision, an Abcam company and was previously called K251

Glutathione Fluorometric Assay Kit. K251-100 is the same size as the 100 test size of ab65322.

Glutathione (GSH) is a tripeptide that contains L-cysteine, L-glutamic acid and glycine. It is the smallest intracellular protein thiol molecule in the cells, which prevents cell damage caused by reactive oxygen species such as free radicals and peroxides. Glutathione exists in reduced (GSH) and oxidized (GSSG) states. Reduced glutathione (GSH) is a major tissue antioxidant that provides reducing equivalents for the glutathione peroxidase (GPx) catalyzed reduction of lipid hydroperoxides to their corresponding alcohols and hydrogen peroxide to water. In the GPx catalyzed reaction, the formation of a disulfide bond between two GSH molecules generates oxidized glutathione (GSSG). The enzyme glutathione reductase (GR) recycles GSSG to GSH with the simultaneous oxidation of β -nicotinamide adenine dinucleotide phosphate (β -NADPH2). In healthy cells, more than 90% of the total glutathione pool is in the reduced form (GSH). When cells are exposed to increased levels of oxidative stress, GSSG accumulates and the ratio of GSSG to GSH increases. An increased ratio of GSSG-to-GSH is an indication of oxidative stress. The monitoring of reduced and oxidized GSH in biological samples is essential for evaluating the redox and detoxification status of the cells and tissues against oxidative and free radicals mediated cell injury.

Platform Microplate reader

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Properties

Storage instructions

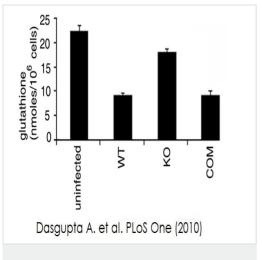
Store at -20°C. Please refer to protocols.

Components	100 tests
GSH Cell Lysis Buffer	1 x 25ml
GSH Standard	1 vial
GST Enzyme Mix	1 x 200µl
MCB Probe	1 x 200µl

Relevance

Glutathione is a small peptide composed of three amino acids: cysteine, glutamic acid, and glycine and is present in tissues in concentrations as high as one millimolar. Glutathione is the principal intracellular low-molecular-weight thiol that plays a critical role in the cellular defense against oxidative and nitrosative stress in mammalian cells. Diminished glutathione levels have been observed in the early stages of apoptosis.

Images

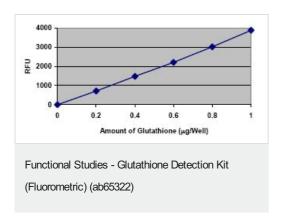


Glutathione Detection Kit (Fluorometric) (ab65322)

Dasgupta A et al., PLoS One, 5, , 2010 Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Glutathione pool measured in THP-1 macrophages: uninfected cells; WT: infected with M.tuberculosis wild type; KO: infected with M.tuberculosis OppD knock-out; COM: infected with M.tuberculosis OppD knock-out complemented with OppDA gene. 10^6 cells were infected and lysed by treating them with $100\mu l$ of ice cold lysis buffer. Cell lysate was diluted and mixed as described in the kit protocol. After 30 min incubation at 37C, fluorescence was measured at Ex=380nm/ Em=460nm. Results represent the means of \pm S.D. of three determinations.

Image obtained from Dasgupta A. et al; PLoS One; 2010 Aug 17; 5(8): e12225.



Glutathione assays were performed using various amounts of Glutathione as indicated. Results were analyzed according to the kit instructions.

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