abcam

Product datasheet

TUNEL Assay Kit - BrdU-Red ab66110

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Overview

Product name TUNEL Assay Kit - BrdU-Red

Detection method Fluorescent

Sample type Tissue, Adherent cells, Suspension cells

Assay type Quantitative
Assay time 3h 00m

Species reactivity Reacts with: Mammals, Other species

Product overview TUNEL Assay Kit - BrdU-Red ab66110 uses a convenient and sensitive method to detect DNA

fragmentation by flow cytometry and fluorescence microscopy in live cells.

The TUNEL assay is used to detect DNA fragmentation, such as in apoptosis. It uses terminal deoxynucleotidyl transferase (TdT) to catalyze the incorporation of deoxynucleotides at the free 3'-hydroxyl ends of fragmented DNA. The deoxynucleotides are then labeled in a variety of ways for detection of the degree of DNA fragmentation.

This TUNEL assay protocol is based on Br-dUTP (bromolated deoxyuridine triphosphate nucleotide), which can be more readily incorporated into DNA strand breaks by the TdT enzyme than other dUTP labels such as FITC, biotin or dioxigenin. The greater incorporation rate produces a brighter signal when the Br-dUTP sites are detected with an anti-BrdU monoclonal antibody directly labeled with a red fluorochrome.

The BrdU-Red signal can be analyzed at Ex/Em 488/576 nm, with an optional 7-AAD counterstain at Ex/Em 488/655nm.

This TUNEL assay kit includes both negative and positive control cells. It is designed to be suitable for studying DNA fragmentation in GFP-transfected cells.

Tunel assay protocol summary:

- fix cells / tissues with formaldehyde, or deparaffinize and rehydrate if paraffin sections, and wash
- incubate cells in 70% ethanol for 30 min at 4°C, or if tissues incubate with proteinase K solution for 5 min at room temp and refix with formaldehyde
- wash
- incubate in DNA labeling solution for 60 min at 37°C
- wash

- incubate in antibody solution for 30 min at room temp
- add 7-AAD / RNase A solution and incubate for 30 min at room temp
- analyze with flow cytometry or fluorescent microscopy

Notes

This product is manufactured by BioVision, an Abcam company and was previously called K404 ApoBrdU Red DNA Fragmentation Kit. K404-60 is the same size as the 60 test size of ab66110.

This kit is BrdU-Red labeled (Ex/Em = 488/576 nm). It was previously called TUNEL Assay Kit - In situ BrdU-Red DNA Fragmentation.

To use FITC (Ex/Em = 495/519 nm) as a label, we recommend **TUNEL Assay Kit - FITC** (ab66108).

For chromogenic TUNEL staining, we recommend **TUNEL Assay Kit - HRP-DAB ab206386**.

Find out more about the TUNEL method in the **TUNEL staining / TUNEL assay guide**.

Abcam has not and does not intend to apply for the REACH Authorisation of customers' uses of products that contain European Authorisation list (Annex XIV) substances.

It is the responsibility of our customers to check the necessity of application of REACH Authorisation, and any other relevant authorisations, for their intended uses.

Platform

Flow cytometer, Fluorescence microscope

Properties

Storage instructions

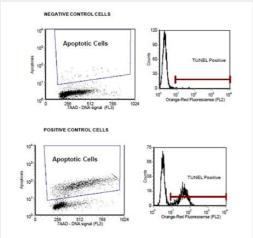
Please refer to protocols.

Components	60 tests	60 tests
7-AAD/RNase Staining Buffer	1 x 30ml	1 x 30ml
Anti-BrdU-Red Antibody	1 x 300µl	1 x 300µl
Br-dUTP	1 x 480µl	1 x 480µl
Negative Control Cells	1 x 5ml	1 x 5ml
Positive Control Cells	1 x 5ml	1 x 5ml
Reaction Buffer	1 x 600µl	1 x 600µl
Rinse Buffer	1 x 120ml	1 x 120ml
TdT Enzymes	1 x 45µl	1 x 45µl
Wash Buffer	1 x 120ml	1 x 120ml

Relevance

Internucleosomal DNA fragmentation is a hallmark of apoptosis in mammalian cells.

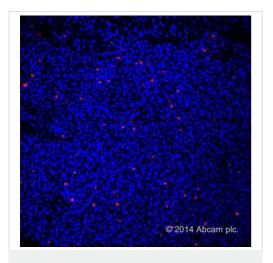
Images



In Situ BrdU-Red DNA Fragmentation (TUNEL)

Assay Kit (ab66110)

Detection of DNA fragmention (TUNEL staining) using the negative and positive control cells (HL-60 untreated and treated with camptothecin). Cells were stained following the assay protocol. The fluorescence signal was detected and analyzed using BD FACScan System (Becton Dickinson).

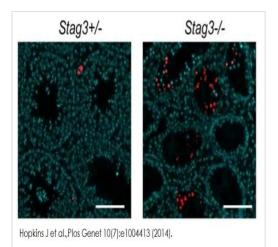


Fluorescence Microscopy - In situ BrdU-Red DNA Fragmentation (TUNEL) Assay Kit (ab66110)

This image is courtesy of an Abreview submitted by James Gahan

TUNEL staining in whole mount Hydractinia echinata using In situ BrdU-Red DNA Fragmentation (TUNEL) Assay Kit (ab66110).

Animals were fixed in 4% PFA in PBS for 1 hour and processed as per the protocol without proteinaseK treatment. In place of proteinaseK animals were permeabilised in 3% Triton in PBS for 15 minutes. Animals were counter-stained with DAPI.



Immunohistochemical analysis of paraffin embedded 5 micron thick testis tissues of 8 week old $Stag3^{+/-}$ and $Stag3^{-/-}$ (Stromal antigen) mice . Apoptotic cells were detected using In situ BrdU-Red DNA Fragmentation (TUNEL) Assay Kit (ab66110). DAPI was used as a counterstain.

Immunohistochemistry - ab66110

Image from Hopkins J et al., PLoS Genet 10(7), fig 1C. Doi: 10.1371/journal.pgen.1004413. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

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