# abcam

#### Product datasheet

## TUNEL Assay Kit - Edu-Orange ab252888

#### 2 Images

#### Overview

Product name TUNEL Assay Kit - Edu-Orange

**Detection method** Flow cytometry-fluorescent

Sample type Tissue, Adherent cells, Suspension cells

**Assay duration** Multiple steps standard assay

Product overview TUNEL Assay Kit - Edu-Orange ab252888 uses an EdU click chemistry method to detect

fragemented DNA. The kit contains sufficient reagents to detect total/fragmented DNA in

apoptotic cells in a 1 X 96-well plate or on 50 cover slips.

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 uses modified EdUTP nucleotides. A click reaction is then used to attach an orange dye (Ex 490 / Em 580, FL2 channel) to the EdUTP for detection by either flow cytometry or fluoresence microscopy. A green DNA staining dye is used for contrast (Ex 440 / Em 540)

TUNEL assay protocol summary:

- fix cells for 15 mins at room temp
- wash cells
- incubate with permeabilization buffer for 10 mins and wash twice
- add TUNEL buffer and incubate for 10 mins
- spin down cells and remove buffer
- add TUNEL reaction cocktail and incubate for 1hr at 37°C
- spin down cells and remove cocktail, and wash
- add Click reaction cocktail, incubate for 30 mins and wash 3 times
- add DNA stain, incubate for 20 mins and wash
- analyze with flow cytometer or fluoresence microscope

This product is manufactured by BioVision, an Abcam company and was previously called K191 EZClick™ TUNEL – in situ DNA Fragmentation/Apoptosis Assay Kit. K191-100 is the same size

as the 100 test size of ab252888.

Tested applications Suitable for: Flow Cyt, FM

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Notes

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#### **Properties**

Storage instructions

Please refer to protocols.

Components	100 tests
1000X Total DNA Stain	1 x 10µl
100X Copper Reagent	1 x 100µl
10X Permeabilization Buffer	1 x 25ml
10X TUNEL Reaction Buffer	1 x 1ml
10X Wash Buffer IV	1 x 25ml
20X Reducing Agent	1 x 500µl
50X EdUTP DNA Label	1 x 100µl
Fixative Solution I	1 x 10ml
100X Fluorescent Azide I	1 x 100µl
TUNEL Enzyme	1 vial
TUNEL Enzyme Buffer	1 x 500µl

Relevance

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

#### **Applications**

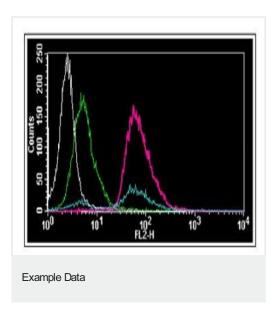
The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab252888 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

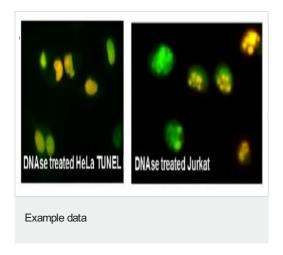
Application	Abreviews	Notes
Flow Cyt		Use at an assay dependent concentration.
FM		Use at an assay dependent concentration.

#### **Images**



Detection of TUNEL-positive apoptotic strand breaks. Jurkat (Human T cell leukemia cell line from peripheral blood) cells-DNAse treated (10<sup>6</sup> cells/ml) induced strand breaks.

Unstained cells w/vehicle (white), background control cells processed for click reaction (green), negative control (untreated cells, TUNEL and click reaction; blue), DNase-treated cells (pink).



**Left panel:** DNase treated HeLa (Human epithelial cell line from cervix adenocarcinoma) cells (10<sup>5</sup> cells/ ml). DNA staining and TUNEL and click reactions were performed.

Green: nuclear stain; Red (TUNEL positive); orange: apoptotic cells.

**Right panel:** DNase treated Jurkat (Human T cell leukemia cell line from peripheral blood) cells (10<sup>6</sup> cells/ ml). DNA staining and TUNEL and click reactions were performed.

Green: nuclear stain; Red (TUNEL positive); orange: apoptotic cells.

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