abcam

Product datasheet

Annexin V-iFluor 488 Apoptosis Detection Kit ab219916

1 References 1 Image

Overview

Product name	Annexin V-iFluor 488 Apoptosis Detection Kit
Detection method	Fluorescent
Sample type	Adherent cells, Suspension cells
Assay type	Quantitative
Species reactivity	**Reacts with:** Mammals, Other species
Product overview	Annexin V-iFluor 488 Apoptosis Detection Kit (ab219916) contains Annexin V labeled with our proprietary green fluorescent dye iFluor 488, which allows the identification and quantitation of apoptotic cells on a single-cell basis by flow cytometry. Optionally, it can be used for simultaneous staining of cells with Annexin V-iFluor488 (green fluorescence) and the nonvital dye propidium iodide (PI) (orange fluorescence) which allows the discrimination of intact cells (Annexin V-iFluor 488 negative, PI Staining Solution negative), early apoptotic (Annexin V-iFluor 488 positive, PI Staining Solution negative) and late apoptotic or necrotic cells (Annexin V-iFluor 488 positive, PI Staining Solution positive). The iFluor 488 dye (Ex/Em = 490/525 nm) has spectral properties almost identical to those of FITC or Alexa Fluor® 488, making it convenient to be used with the common fluorescence instruments equipped with the light sources and filters for FITC, the most common fluorophore.
Notes	Apoptosis is a regulated process of cell death that occurs during embryonic development as well as maintenance of tissue homeostasis. Inappropriately regulated apoptosis is implicated in different disease states, such as neurodegeneration disease and cancer. The apoptosis program is characterized by morphologic features, including loss of plasma membrane asymmetry and attachment, condensation off the cytoplasm and nucleus, and compaction and fragmentation of the nuclear chromatin. Exposure of phosphatidylserine (PS) on the external surface of the cell membrane has been reported to occur in the early phases of apoptotic cell death, during which the cell membrane remains intact. In leukocyte apoptosis, PS on the outer surface of the cell marks the cell for recognition and phagocytosis by macrophages. The human vascular anticoagulant, annexin V, is a 35-36 kDa Ca^{2+} dependent phospholipid binding protein that has a high affinity for PS, and shows minimal binding to phosphatidylcholine and sphingomyelin. Changes in PS asymmetry, which can be analyzed by measuring annexin V binding to the cell membrane, are generally observed before morphological changes associated with apoptosis occurred and before membrane integrity is lost. *Alexa Fluor® 488 is the trademark of Invitrogen.*

Platform	Flow cytometer, Fluorescence microscope

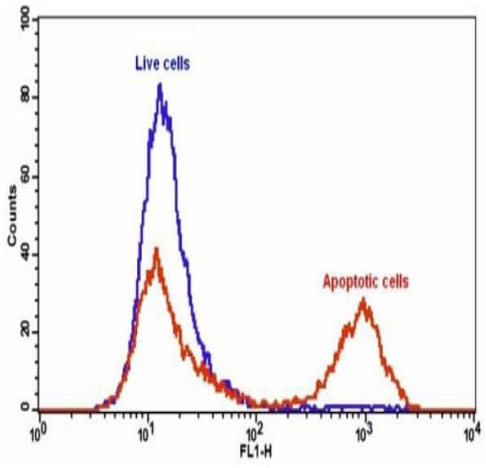
Properties

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

Components	100 tests
100X Propidium Iodide	1 x 100µl
Annexin 488 (100X stock solution)	1 x 200µl
Assay Buffer	1 x 50ml

Function	This protein is an anticoagulant protein that acts as an indirect inhibitor of the thromboplastin-specific complex, which is involved in the blood coagulation cascade.
Involvement in disease	Pregnancy loss, recurrent, 3
Sequence similarities	Belongs to the annexin family. Contains 4 annexin repeats.
Domain	The [IL]-x-C-x-x-[DE] motif is a proposed target motif for cysteine S-nitrosylation mediated by the iNOS-S100A8/A9 transnitrosylase complex. A pair of annexin repeats may form one binding site for calcium and phospholipid.
Post-translational modifications	S-nitrosylation is induced by interferon-gamma and oxidatively-modified low-densitity lipoprotein (LDL(ox)) possibly implicating the iNOS-S100A8/9 transnitrosylase complex.

Images



The detection of binding activity of Annexin V-iFluor 488 to phosphatidylserine in Jurkat cells

Annexin V-iFluor 488 Apoptosis Detection Kit (ab219916). Detection of phosphatidylserine (PS) exposure in Jurkat cells. Jurkat cells were left untreated (blue) or treated with 20 µM camptothecin (red) in a 37°C, 5% CO_2 incubator for 4-5 hours.

Cells were then incubated with Annexin V-iFluor reagent and PI for 30 minutes. The fluorescence intensitiy of Annexin V-iFluor 488 was measured with a FACSCalibur (BD Systems) flow cytometer using the FL1channel.

In live non-apoptotic cells, Annexin V-iFluor 488 conjugate detects innate apoptosis in non-induced cells, which is typically 2-6% of all cells. In apoptotic cells Annexin V-iFluor 488 conjugate binds to phosphatidylserine, which is located on the outer leaflet of the cell membrane, resulted in increased staining intensity.

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