Datasheet: ANNEX100B

Description:	ANNEXIN V:Biotin ASSAY KIT
Name:	ANNEXIN V KIT
Format:	Biotin
Product Type:	Kits
Quantity:	100 TESTS

Product Details

Applications

This product has been reported to work in the following applications. This information is derived from testing within our laboratories, peer-reviewed publications or personal communications from the originators. Please refer to references indicated for further information. For general protocol recommendations, please visit www.bio-rad-antibodies.com/protocols.

	Yes	No	Not Determined	Suggested Dilution
Flow Cytometry	▪			Neat

Where this product has not been tested for use in a particular technique this does not necessarily exclude its use in such procedures. Suggested working dilutions are given as a guide only. It is recommended that the user titrates the product for use in their own system using appropriate negative/positive controls.

Preservative Stabilisers

0.02% Sodium Azide (NaN_3)
1% Bovine Serum Albumin

External Database Links

UniProt:
P08758 Related reagents

Entrez Gene:
308 ANXA5 Related reagents

Synonyms

ANX5, ENX2, PP4

Product Information

This test employs the property of Annexin V to bind to the membrane phospholipid phosphatidylserine (PS) in the presence of Ca^{2+}. PS is exposed at the cell surface during the early stages of apoptosis. Detection of PS is a very sensitive method for detecting cells entering apoptosis, at a time point considerably ahead of nuclear changes such as DNA degradation.

The conjugation protocol used to prepare this product has not changed the native phospholipid binding properties of Annexin V. This protocol is designed to measure apoptosis easily and quickly in a sample of suspended cells.

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Reagents In The Kit

Annexin V:Biotin	1 x 0.5ml vial
Propidium Iodide	1 x 1.6ml vial at 20ug/ml
Binding buffer	1 x 50ml vial at x 4 concentrate

Note: This assay also requires streptavidin:FITC conjugate for visualisation (not supplied - see recommended useful reagents section).

Instructions For Use

1) Dilute the binding buffer 1:4 in distilled water (50ml binding buffer + 150ml distilled water).

2) Wash cells in PBS by gentle shaking or pipetting up and down.

3) Resuspend cells in 200ul pre-diluted binding buffer, adjusting to a cell density of 2-5 x 10^5 cells/ml.

4) Add 5ul Annexin V:Biotin to 195ul of the cell suspension prepared in step 3.

5) Mix and incubate for 15 minutes at room temperature.

6) Wash cells twice in 190ul of pre-diluted binding buffer.

7) Resuspend cells in 190ul pre-diluted binding buffer.

8) Add streptavidin:FITC conjugate.

9) Mix and incubate for 30 minutes in the dark, at room temperature.

10) Wash cells in 200ul pre-diluted binding buffer.

11) Resuspend cells in 190ul pre-diluted binding buffer.

12) Add 10ul of the Propidium Iodide solution.

13) Analyse by flow cytometry.

The flow cytometer is preferably set such that the Mean Fluorescence Intensity of the Annexin V negative population is between 1 and 10. Optimal parameter settings can be found using a positive control. For a positive control, incubate the cells with 3% formaldehyde in buffer during 30 minutes on ice. Wash away the formaldehyde and suspend the cells in cold binding buffer at 2-5 x 10^5 cells/ml. Proceed with step 2 as described above.

References

1. Lu, K.H. *et al.* (2010) *In Vitro* and *In Vivo* Apoptosis-Inducing Antileukemic Effects of

Mucuna macrocarpa Stem Extract on HL-60 Human Leukemia Cells. Integr Cancer Ther. 9: 298-308.
2. Yen, J.H. *et al.* (2010) Glycine tomentella Hayata inhibits IL-1β and IL-6 production, inhibits MMP-9 activity, and enhances RAW264.7 macrophage clearance of apoptotic cells. J Biomed Sci. 17: 83.
3. Chen, C.W. *et al.* (2010) The signals of FGFs on the neurogenesis of embryonic stem cells. J Biomed Sci.17:33.
4. Smith, K. *et al.* (2011) Mono- and tri-cationic porphyrin-monoclonal antibody conjugates: photodynamic activity and mechanism of action. Immunology. 132: 256-65.
5. Lu, K.H. *et al.* (2012) Synergistic Apoptosis-Inducing Antileukemic Effects of Arsenic Trioxide and Mucuna macrocarpa Stem Extract in Human Leukemic Cells via a Reactive Oxygen Species-Dependent Mechanism. Evid Based Complement Alternat Med. 2012:921430.
6. Koutsogiannaki, S. *et al.* (2015) Effects of cadmium and 17β-estradiol on *Mytilus galloprovincialis* redox status. Prooxidant-antioxidant balance (PAB) as a novel approach in biomonitoring of marine environments. Mar Environ Res. 103: 80-8.
7. Iulia, A.I. *et al.* (2015) Inhibition of tumor necrosis factor alpha using RNA interference in oral squamous cell carcinoma. J BUON. 20 (4): 1107-14.
8. Hounkong, K. *et al.* (2015) Mechanisms of 1-hydroxy-2-hydroxymethylanthraquinone from *Coptosapelta flavescens* as an anti-giardial activity. Acta Trop. 146: 11-6.

Storage	Store at +4°C. DO NOT FREEZE. This product should be stored undiluted. Should this product contain a precipitate we recommend microcentrifugation before use.
Guarantee	Guaranteed until date of expiry. Please see product label.
Health And Safety Information	Material Safety Datasheet documentation #10229 #10230 #10181 available at: Recombinant Annexin V (10229): https://www.bio-rad-antibodies.com/uploads/MSDS/10229.pdf Binding Buffer (10230): https://www.bio-rad-antibodies.com/uploads/MSDS/10230.pdf Propidium Iodide (10181): https://www.bio-rad-antibodies.com/uploads/MSDS/10181.pdf
Regulatory	For research purposes only

Related Products

Recommended Useful Reagents

ANNEXIN V:FITC ASSAY KIT (ANNEX100F)
ANNEXIN V:FITC ASSAY KIT (ANNEX300F)
STREPTAVIDIN:FITC (STAR2B)

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To find a batch/lot specific datasheet for this product, please use our online search tool at: bio-rad-antibodies.com/datasheets
'M393929:220120'

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