

## Datasheet: ANNEX20B

|                      |                            |
|----------------------|----------------------------|
| <b>Description:</b>  | ANNEXIN V:Biotin ASSAY KIT |
| <b>Name:</b>         | ANNEXIN V KIT              |
| <b>Format:</b>       | Biotin                     |
| <b>Product Type:</b> | Kits                       |
| <b>Quantity:</b>     | 20 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](http://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<sub>3</sub>)  
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<sup>2+</sup>. 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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|----------------------------|----------------------------------------------------------------------------------------------------------------------------------|
| <b>Reagents In The Kit</b> | Annexin V:Biotin 1 x 0.1ml vial<br>Propidium Iodide 1 x 1.6ml vial at 20ug/ml<br>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).

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| <b>Instructions For Use</b> | <ol style="list-style-type: none"><li>1) Dilute the binding buffer 1:4 in distilled water (50ml binding buffer + 150ml distilled water).</li><li>2) Wash cells in PBS by gentle shaking or pipetting up and down.</li><li>3) Resuspend cells in 200ul pre-diluted binding buffer, adjusting to a cell density of 2-5 x 10<sup>5</sup> cells/ml.</li><li>4) Add 5ul Annexin V:Biotin to 195ul of the cell suspension prepared in step 3.</li><li>5) Mix and incubate for 15 minutes at room temperature.</li><li>6) Wash cells twice in 190ul of pre-diluted binding buffer.</li><li>7) Resuspend cells in 190ul pre-diluted binding buffer.</li><li>8) Add streptavidin:FITC conjugate.</li><li>9) Mix and incubate for 30 minutes in the dark, at room temperature.</li><li>10) Wash cells in 200ul pre-diluted binding buffer.</li><li>11) Resuspend cells in 190ul pre-diluted binding buffer.</li><li>12) Add 10ul of the Propidium Iodide solution.</li><li>13) Analyse by flow cytometry.</li></ol> |
|-----------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

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<sup>5</sup> cells/ml. Proceed with step 2 as described above.

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|-------------------|----------------------------------------------------------------------------------------------------------------|
| <b>References</b> | 1. Lu, K.H. <i>et al.</i> (2010) <i>In Vitro</i> and <i>In Vivo</i> 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-1 $\beta$  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.](#)

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**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.

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**Guarantee**

Guaranteed until date of expiry. Please see product label.

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**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>

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**Regulatory**

For research purposes only

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## Related Products

### Recommended Useful Reagents

[ANNEXIN V:Biotin ASSAY KIT \(ANNEX100B\)](#)

[ANNEXIN V:FITC ASSAY KIT \(ANNEX100F\)](#)

[ANNEXIN V:FITC ASSAY KIT \(ANNEX300F\)](#)

[ANNEXIN V:FITC ASSAY KIT \(ANNEX20F\)](#)

**North & South** Tel: +1 800 265 7376

**America** Fax: +1 919 878 3751

Email: [antibody\\_sales\\_us@bio-rad.com](mailto:antibody_sales_us@bio-rad.com)

**Worldwide**

Tel: +44 (0)1865 852 700

Fax: +44 (0)1865 852 739

Email: [antibody\\_sales\\_uk@bio-rad.com](mailto:antibody_sales_uk@bio-rad.com)

**Europe**

Tel: +49 (0) 89 8090 95 21

Fax: +49 (0) 89 8090 95 50

Email: [antibody\\_sales\\_de@bio-rad.com](mailto:antibody_sales_de@bio-rad.com)

To find a batch/lot specific datasheet for this product, please use our online search tool at: [bio-rad-antibodies.com/datasheets](https://www.bio-rad-antibodies.com/datasheets)

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