



6042 Cornerstone Court W, Ste B  
San Diego, CA 92121  
Tel: 1.858.202.1401  
Fax: 1.858.481.8694  
Email: info@bpsbioscience.com

**Data sheet**  
**SARS-CoV-2 Spike S1-Biotin:**  
**ACE2 TR-FRET Assay Kit**  
Catalog #79949-1  
Size: 96 reactions

**DESCRIPTION:** The pandemic coronavirus disease 2019 (COVID-19) is caused by Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2). As a first step of the viral replication strategy, the virus attaches to the host cell surface before entering the cell. The Spike protein recognizes and attaches to the Angiotensin-Converting Enzyme 2 (ACE2) receptor found on the surface of type I and II pneumocytes, endothelial cells, and ciliated bronchial epithelial cells. Drugs targeting the interaction between the Spike protein of SARS-CoV-2 and ACE2 may offer some protection against the viral infection.

The SARS-CoV-2 Spike S1:ACE2 TR-FRET Assay is designed to measure the inhibition of the binding between SARS-CoV-2 Spike S1 and human ACE2 in a homogeneous 96 reaction format. This TR-FRET-based assay requires no time-consuming washing steps, making it especially suitable for high throughput screening applications. The assay procedure is straightforward and simple; the test inhibitor compound is incubated with biotinylated Spike S1, Eu-labeled ACE2, dye-labeled acceptor and an inhibitor for one hour. Then the TR-FRET signal is measured using a fluorescence reader.

**COMPONENTS:**

| Catalog # | Component   | Amount    | Storage   |                                    |
|-----------|---|-----------|-----------|------------------------------------|
| 100705    | ACE2, His-Tag, Eu-labeled                                 | 2 µg      | -80°C     | Avoid multiple freeze/thaw cycles! |
| 100679    | Spike S1, Fc fusion, Avi-tag, Biotin-Labeled (SARS-CoV-2) | 25 µg     | -80°C     |                                    |
|           | Dye-labeled acceptor                                      | 2 x 10 µl | -20°C     |                                    |
| 79953     | 3x ACE2-Spike TR-FRET Buffer                              | 4 ml      | -20°C     |                                    |
|           | 96-well white microplate                                  | 1         | Room Temp |                                    |

**APPLICATIONS:** This kit is useful for screening for inhibitors of the interaction between SARS-CoV-2 Spike S1 and human ACE2.

**STABILITY:** Up to 6 months from date of receipt, when stored as recommended.

**REFERENCES:**

Hoffmann, M. *et al.* 2020. *Cell*, **181**:1-10  
Yan, R. *et al.* 2020. *Science* **367(6485)**:1444-1448.

OUR PRODUCTS ARE FOR RESEARCH USE ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.

To place your order, please contact us by Phone **1.858.202.1401** Fax **1.858.481.8694**

Or you can Email us at: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

Please visit our website at: [www.bpsbioscience.com](http://www.bpsbioscience.com)



6042 Cornerstone Court W, Ste B  
San Diego, CA 92121  
Tel: 1.858.202.1401  
Fax: 1.858.481.8694  
Email: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

**MATERIALS OR INSTRUMENTS REQUIRED BUT NOT SUPPLIED:**

Fluorescence microplate reader capable of measuring Time Resolved-Fluorescence Resonance Energy Transfer (TR-FRET)  
Adjustable micropipettor and sterile tips

**ASSAY PROTOCOL:**

All samples and controls should be tested in duplicate.

- 1) Thaw **ACE2-Eu** on ice. Upon first thaw, briefly spin tube containing the protein to recover the full contents of the tube. Aliquot into single use aliquots. Immediately store remaining undiluted protein in aliquots at  $-80^{\circ}\text{C}$ . Note: **ACE2-Eu** is very sensitive to freeze/thaw cycles. Avoid multiple freeze/thaw cycles.
- 2) Dilute one part **3x ACE2-Spike TR-FRET Buffer** with 2 parts of distilled water (3-fold dilution) to make **1x ACE2-Spike TR-FRET Buffer**. Prepare only a sufficient quantity needed for the assay; store remaining stock solution in aliquots at  $-20^{\circ}\text{C}$ .
- 3) Dilute **ACE2-Eu** in **1x ACE2-Spike TR-FRET Buffer** to 1 ng/ $\mu\text{l}$  (12 nM). Keep diluted protein on ice until ready to use. Discard any remaining unused diluted protein after use. Add 12.5  $\mu\text{l}$  of **ACE2-Eu** to all wells
- 4) Dilute **Dye-labeled Acceptor** 100-fold with **1x ACE2-Spike TR-FRET Buffer**. Add 12.5  $\mu\text{l}$  of **Dye-labeled Acceptor** to all wells
- 5) Prepare the **Test inhibitor** solution. If the inhibitor compound is water soluble (e.g. an antibody), make a solution of the compound 4-fold higher than the final concentration in **1x ACE2-Spike TR-FRET Buffer**. If the inhibitor compound is a small molecule soluble in DMSO, final DMSO concentration in the assay should be  $\leq 1\%$ , and Inhibitor buffer should contain the same concentration of DMSO as the test inhibitor.
- 6) Add 12.5  $\mu\text{l}$  of test inhibitor solution to each well designated "Test Inhibitor". For the "Positive Control" and "Blank", add 12.5  $\mu\text{l}$  of the same solution without inhibitor (**1x ACE2-Spike TR-FRET Buffer** with the same concentration of solvent as in the test inhibitor solution).

OUR PRODUCTS ARE FOR RESEARCH USE ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.

To place your order, please contact us by Phone **1.858.202.1401** Fax **1.858.481.8694**

Or you can Email us at: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

Please visit our website at: [www.bpsbioscience.com](http://www.bpsbioscience.com)



6042 Cornerstone Court W, Ste B  
 San Diego, CA 92121  
 Tel: 1.858.202.1401  
 Fax: 1.858.481.8694  
 Email: info@bpsbioscience.com

|                            | Positive Control | Blank        | Test Inhibitor |
|----------------------------|------------------|--------------|----------------|
| ACE2-Eu (1 ng/μl)          | 12.5 μl          | 12.5 μl      | 12.5 μl        |
| Dye-labeled acceptor       | 12.5 μl          | 12.5 μl      | 12.5 μl        |
| Test Inhibitor             | -                | -            | 12.5 μl        |
| Inhibitor buffer           | 12.5 μl          | 12.5 μl      | -              |
| 1x TR-FRET Buffer          | -                | 12.5 μl      | -              |
| Spike S1-Biotin (20 ng/μl) | 12.5 μl          | -            | 12.5 μl        |
| <b>Total</b>               | <b>50 μl</b>     | <b>50 μl</b> | <b>50 μl</b>   |

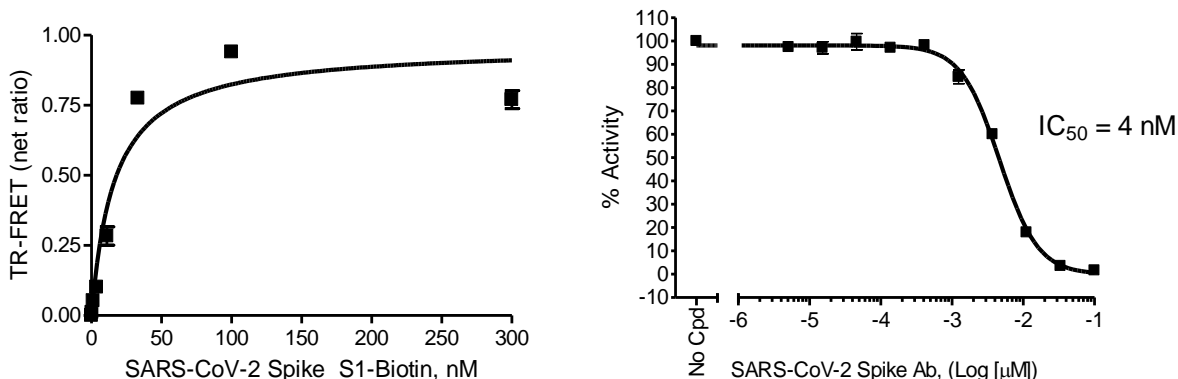
- 7) Thaw **Spike S1-Biotin** on ice. Upon first thaw, briefly spin tube containing the protein to recover the full contents of the tube. Aliquot into single use aliquots. Immediately store remaining undiluted protein in aliquots at -80°C. Note: **Spike S1-Biotin** is very sensitive to freeze/thaw cycles. Avoid multiple freeze/thaw cycles.
- 8) Dilute **Spike S1-Biotin** in **1x ACE2-Spike TR-FRET Buffer** to 20 ng/μl (200 nM). Keep diluted protein on ice until ready to use. Discard any remaining unused diluted protein after use. Add 12.5 μl of diluted **Spike S1-Biotin** to wells designated "Test Inhibitor" and "Positive Control". Add 12.5 μl of **1x ACE2-Spike TR-FRET Buffer** to wells designated "Blank". Incubate the plate at room temperature for 1 hour.
- 9) Read TR-FRET signal in a microtiter-plate reader under settings described below (settings may need optimization depending on the instrument). Blank value is subtracted from all other values.

| Channel | Variable                   | Recommended value |
|---------|----------------------------|-------------------|
| 1       | Excitation wavelength (nm) | 340 ± 20          |
|         | Emission wavelength (nm)   | 620 ± 10          |
|         | Lag time (μs)              | 60                |
|         | Integration time (μs)      | 500               |
| 2       | Excitation wavelength (nm) | 340 ± 20          |
|         | Emission wavelength (nm)   | 665 ± 10          |
|         | Lag time (μs)              | 60                |
|         | Integration time (μs)      | 500               |

OUR PRODUCTS ARE FOR RESEARCH USE ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.

To place your order, please contact us by Phone **1.858.202.1401** Fax **1.858.481.8694**  
 Or you can Email us at: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)  
 Please visit our website at: [www.bpsbioscience.com](http://www.bpsbioscience.com)

**Example of assay results:**



Titration of SARS-CoV-2 Spike S1 (BPS Bioscience, #100679) (left) and inhibition of SARS-CoV-2 Spike S1:ACE2 binding using human anti-SARS-CoV-2 Spike Antibody (BPS Bioscience, #100793) (right) in the *SARS-CoV-2 Spike S1:ACE2 TR-FRET Assay Kit* (BPS Bioscience, #79949-1). Data shown is lot-specific. For lot-specific information, please contact BPS Bioscience, Inc. at [info@bpsbioscience.com](mailto:info@bpsbioscience.com).

**RELATED PRODUCTS:**

| <u>Product Name</u>   | <u>Catalog#</u> | <u>Size</u>   |
|---|-----------------|---------------|
| Spike S1-Biotin (SARS-CoV-2): ACE2 TR-FRET Assay Kit          | 79949-2         | 384 reactions |
| SARS-CoV-2 Spike:ACE2 Inhibitor Screening Assay Kit           | 79931           | 96 reactions  |
| ACE2:SARS-CoV-2 Spike Inhibitor Screening Assay Kit           | 79936           | 96 reactions  |
| ACE2:SARS-CoV-2 Spike S1-Biotin Inhibitor Screening Assay Kit | 79945           | 96 reactions  |
| Spike S1, Fc Fusion, Avi-tag (SARS-CoV-2)                     | 100678          | 100 µg/1 mg   |
| Spike S1, Fc fusion, Avi-tag, Biotin-Labeled (SARS-CoV-2)     | 100679          | 25 µg/50 µg   |
| Spike S1 RBD, His-tag (SARS-CoV-2)                            | 100687          | 50 µg/100 µg  |
| Spike S1, Fc fusion (SARS-CoV-2)                              | 100688          | 20 µg/50 µg   |
| Spike S1 RBD, Fc fusion (SARS-CoV-2)                          | 100699          | 50 µg/100 µg  |
| ACE2 Inhibitor Screening Assay Kit                            | 79923           | 96 reactions  |
| ACE2, His-tag   | 11003           | 20 µg/100 µg  |
| ACE2, His-Avi-Tag, Biotin-labeled HiP™                        | 100665          | 20 µg/50 µg   |
| ACE2, Fc Fusion (Monkey)                                      | 100701          | 50 µg/1 mg    |
| ACE2, His-tag (Monkey)  | 100702          | 50 µg/1 mg    |

OUR PRODUCTS ARE FOR RESEARCH USE ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.

To place your order, please contact us by Phone **1.858.202.1401** Fax **1.858.481.8694**

Or you can Email us at: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

Please visit our website at: [www.bpsbioscience.com](http://www.bpsbioscience.com)