



SZABO SCANDIC

Part of Europa Biosite

Produktinformation



Forschungsprodukte & Biochemikalien



Zellkultur & Verbrauchsmaterial



Diagnostik & molekulare Diagnostik



Laborgeräte & Service

Weitere Information auf den folgenden Seiten!
See the following pages for more information!



Lieferung & Zahlungsart

siehe unsere [Liefer- und Versandbedingungen](#)

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 



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
FGL1:LAG3 TR-FRET Assay Kit
Catalog #79739-1
Size: 96 reactions

BACKGROUND: Lymphocyte-activation gene 3 (LAG3, also CD223) is a cell surface receptor that negatively regulates activation and proliferation of T cells. Fibrinogen-like protein 1 (FGL1), a liver-secreted protein, is a functional LAG3 ligand. Blockade of the FGL1-LAG3 interaction is implicated in promoting antitumor immunity.

DESCRIPTION: The FGL1:LAG3 TR-FRET Assay is designed to measure the inhibition of LAG3 binding to FGL1 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; a sample containing biotinylated LAG3, His-tagged FGL1 protein, and an inhibitor are incubated for one hour. Then, anti-His Tb donor and dye-labeled acceptor are added and fluorescence intensity is measured using a fluorescence reader

COMPONENTS:

Catalog #	Component	Amount	Storage	
100330	FGL1, His tag	5 µg	-80°C	Avoid multiple freeze/thaw cycles!
71147	LAG3 (CD223), Biotin-labeled (Human) HiP™	10 µg	-80°C	
30017	Anti-His Tb Donor	2 x 10 µl	-20°C	
	Dye-labeled Acceptor	2 x 10 µl	-20°C	
	3x FGL1 TR-FRET Buffer	4 ml	-20°C	
79696	White, 96-well microtiter plate	1	Room. temp	

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

APPLICATIONS: This kit is useful for screening for inhibitors of LAG3 binding to FGL1.

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

REFERENCES:

Wang, J., *et al. Cell* 2019, **176(1-2)**: 334-347
Visan. I., *et al. Nature Immunol.* 2019, **20(2)**: 111

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

Please visit our website at: 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

ASSAY PROTOCOL:

All samples and controls should be tested in duplicate.

Step 1:

- 1) Thaw **FGL1-His** 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: **FGL1-His** is very sensitive to freeze/thaw cycles. Avoid multiple freeze/thaw cycles.
- 2) Dilute one part **3x FGL1 TR-FRET Buffer** with 2 parts of distilled water (3-fold dilution) to make **1x FGL1 TR-FRET Buffer**. Prepare only a sufficient quantity needed for the assay; store remaining stock solution in aliquots at -20°C .
- 3) Dilute **FGL1-His** in **1x FGL1 TR-FRET Buffer** to $5\text{ ng}/\mu\text{l}$ (50 nM final assay concentration). Keep diluted protein on ice until ready to use. Discard any remaining unused diluted protein after use.
- 4) Dilute **LAG3-Biotin** in **1x FGL1 TR-FRET Buffer** to $10\text{ ng}/\mu\text{l}$ (50 nM final assay concentration). Keep diluted protein on ice until ready to use. Discard any remaining unused diluted protein after use.
- 5) Add $10\text{ }\mu\text{l}$ of diluted **FGL1-His** to all wells.
- 6) Dilute test inhibitor into **1x FGL1 TR-FRET Buffer**. Add $5\text{ }\mu\text{l}$ of test inhibitor solution to each well designated "Test Inhibitor". For the "Positive Control" and "Blank", add $5\text{ }\mu\text{l}$ of the same solution without inhibitor (**1x FGL1 TR-FRET Buffer** with the same concentration of DMSO as in the test inhibitor solution).
- 7) Add $10\text{ }\mu\text{l}$ of **1x FGL1 TR-FRET Buffer** to wells designated "Blank".
- 8) Add $10\text{ }\mu\text{l}$ of diluted **LAG3-Biotin** to wells designated "Test Inhibitor" and "Positive Control". Incubate the plate at room temperature for 1 hour.

	Positive Control	Blank	Test Inhibitor
FGL1-His ($5\text{ ng}/\mu\text{l}$)	$10\text{ }\mu\text{l}$	$10\text{ }\mu\text{l}$	$10\text{ }\mu\text{l}$
1x FGL1 TR-FRET Buffer	-	$10\text{ }\mu\text{l}$	-
Test Inhibitor	-	-	$5\text{ }\mu\text{l}$
Inhibitor buffer	$5\text{ }\mu\text{l}$	$5\text{ }\mu\text{l}$	-
LAG3-Biotin ($10\text{ ng}/\mu\text{l}$)	$10\text{ }\mu\text{l}$	-	$10\text{ }\mu\text{l}$
Total	$25\text{ }\mu\text{l}$	$25\text{ }\mu\text{l}$	$25\text{ }\mu\text{l}$

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

Please visit our website at: 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

Step 2:

- 1) Dilute **Anti-His Tb Donor** 100-fold with **1x FGL1 TR-FRET Buffer**. Add 12.5 μ l per well. Shake plate briefly. Incubate at room temperature for 30 minutes.

Step 3:

- 1) Dilute **Dye-labeled Acceptor** 100-fold with **1x FGL1 TR-FRET Buffer**. Add 12.5 μ l per well. Incubate at room temperature for 60 minutes.
- 2) Read the fluorescent intensity in a microtiter-plate reader capable of TR-FRET.

Instrument Settings:

Reading Mode	Value
Excitation Wavelength	320 \pm 10 nm
Emission Wavelength	320 \pm 10 nm
Lag Time	60 μ s
Integration Time	500 μ s
Excitation Wavelength	320 \pm 10 nm
Emission Wavelength	665 \pm 10 nm
Lag Time	60 μ s
Integration Time	500 μ s

CALCULATING RESULTS:

Two sequential measurements should be conducted. Tb-donor emission should be measured at 620 nm followed by dye-acceptor emission at 665 nm. Data analysis is performed using the TRFRET ratio (665 nm emission/620 nm emission). If desired, data can be normalized to percent inhibition. Typically for inhibitor screens, the TR-FRET value from the positive control is set to zero percent inhibition and the TR-FRET value from the negative control is set to one hundred percent inhibition.

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

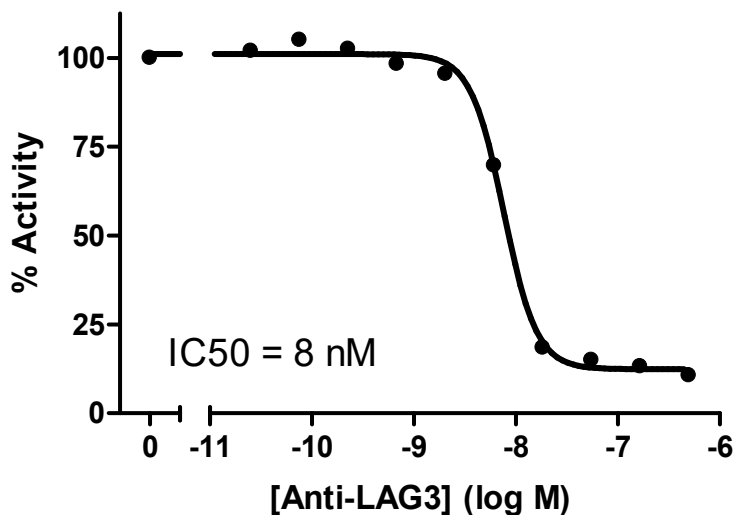
To place your order, please contact us by Phone **1.858.202.1401** Fax **1.858.481.8694**

Or you can Email us at: info@bpsbioscience.com

Please visit our website at: www.bpsbioscience.com

Example of assay results:

FGL1:LAG3-Biotin Binding Assay



Inhibition of FGL1:LAG3 binding using the LAG3 Neutralizing Antibody, BPS Bioscience #71219 and the *FGL1:LAG3 TR-FRET Assay Kit* (#79739-1). Data shown is lot-specific. For lot-specific information, please contact BPS Bioscience, Inc. at info@bpsbioscience.com.

RELATED PRODUCTS:

<u>Product Name</u>	<u>Catalog#</u>	<u>Size</u>
FGL1:LAG3 TR-FRET Assay Kit	79739	384 rxns
Anti-LAG3, Neutralizing Antibody	71219	100 µg
PE labeled anti-LAG3 antibody	71226-1	50 µg
PE labeled anti-LAG3 antibody	71226-2	100 µg
LAG3 / NFAT Reporter - Jurkat Recombinant Cell Line	71278	2 vials
LAG3 (CD223), Fc fusion (Human)	71146	100 µg
LAG3 (CD223), Biotin-labeled (Human) HiP™	71147	50 µg
LAG3 (CD223), Fc fusion (Mouse)	79050	100 µg

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

Please visit our website at: www.bpsbioscience.com