



RIG-I Based dsRNA Assays Combined with dsRNA ELISA

Introduction:

Creative Diagnostics has historically provided high-quality dsRNA ELISA Kits which have been widely praised by our new and long-time clients. This time, our team presents the new RIG-I dsRNA Based Assay Kit, which can be used alone or in combination with the dsRNA ELISA Kit to obtain more comprehensive and reliable dsRNA residue detection results.

Highlights:

- Detection Range: 0.0195 - 2.5 ng/ μ L
- Better recognition of short sequence dsRNA, 30 bp~300 bp dsRNA
- Based on the receptor protein activation, more direct evaluation of the sample immune effect
- Complementary with ELISA method to more comprehensively evaluate the sample immune effect

Test Principle:

After RIG-I binds to dsRNA, ATPase activity is activated, hydrolyzing ATP into ADP + Pi. This kit measures the amount of dsRNA in the sample by detecting the amount of ADP produced during the reaction between RIG-I and the sample. The assay is performed in a multi-well plate and can be detected in a reaction volume as low as 5 μ L. The detection involves three steps:

1. RIG-I Reaction: Mix RIG-I enzyme, ATP, and the sample, followed by incubation.
2. ATP Depletion: Add an equal volume of Reagent 1 to deplete residual ATP.
3. ADP Detection: Add Reagent 2 to convert ADP back to ATP, which is then quantified using a luciferase/luciferin reaction.

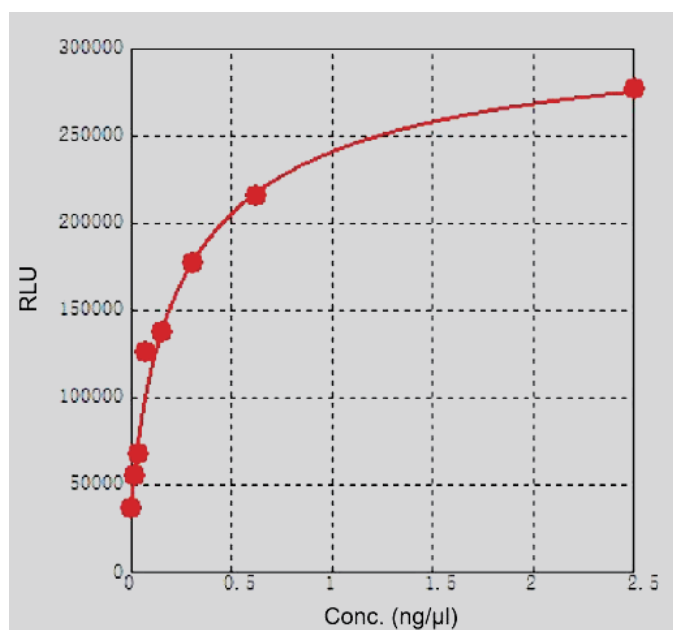
The luminescent signal is measured with a plate reader. dsRNA content is calculated based on a standard curve.

RIG-I Based dsRNA Assays Combined with dsRNA ELISA

Validation Data:

I. Standard curve of 300 bp dsRNA at 0.0195 - 2.5 ng/μL

300 bp N1-Me-pUTP dsRNA	
Conc. (ng/μL)	RLU
2.5	276765
0.625	214916
0.312	176600
0.156	137272
0.078	125769
0.039	68292
0.02	55058
0	36955



II. Recognizes short-chain dsRNA, with a length of 30 bp~300 bp having little effect

dsRNA Sample (RIG-I Identification)	30 bp	40 bp	50 bp	60 bp	120 bp	150 bp	163 bp	300 bp	300 bp N1-Me-pUTP
	RLU (0.1 μM RIG-I)								
2 ng/μL	618268	633005	669551	678494	708961	532731	776493	650178	474084
0.4 ng/μL	382455	388947	444730	458091	382082	206349	507608	343108	227784
No dsRNA	18636								
No RIG-I	27693								

RIG-I Based dsRNA Assays Combined with dsRNA ELISA

III. Verification of RIG-I activity

	ELISA Assay	IL-6: pg/ml	IFN- α : pg/ml	RIG-I dsRNA Assay
mEPO-WT-37°C	0.075%	211.82	85495.37	0.281%
mEPO-20-30-37°C	0.027%	24.36	1155.89	0.085%
CLDN6-WT-37°C	0.027%	150.82	23587.80	0.312%
CLDN6-20-30-50°C	0.003%	21.42	33.33	0.059%
Random Sequence 1000 nt-WT-37°C	0.047%	638.73	42480.43	0.110%
Random Sequence 1000 nt-20-30-37°C	0.006%	115.66	Lower than the detection limit	0.077%

** Results from the RIG-I dsRNA assay generally correlate with those obtained by ELISA. For individual samples (e.g., CLDN6-WT), the RIG-I dsRNA Assay results align more closely with the levels of immune effects.

We recommend this new product for enhanced dsRNA safety assessment in biopharmaceuticals.

Creative Diagnostics

RIG-I Based dsRNA Assays Combined with dsRNA ELISA



Contact Us:

Tel: 1-631-624-4882 (USA) / 44-161-818-6441 (Europe)

Fax: 1-631-938-8221

Email: info@creative-diagnostics.com

www.creative-diagnostics.com

CD Creative Diagnostics®