## CATALOG \& PRICING

| Cat. \# | Description | Size |
| :--- | :--- | :--- |
| $\mathbf{7 0 7 - 5 0}$ |  | 50 mL |
| $\mathbf{7 0 7 - 1 0 0}$ | Use with HRP in <br> chemiluminescent ELISA | 100 mL |
| $\mathbf{7 0 7 - 2 0 0}$ |  | 200 mL |

## BACKGROUND

Enhanced chemiluminescence (ECL) substrates are used with horseradish peroxidase (HRP)conjugated secondary antibodies to detect protein abundance in immunoassays. HRP catalyzes luminol oxidation in the presence of hydrogen peroxide, producing light that can be detected by an X-ray film, digital imager or chemiluminescence plate reader. ShQ ${ }^{\text {TM }}$ ECL Substrate Kit is proprietarily formulated for chemiluminescent ELISA test. Compared to ultrasensitive PiQ ${ }^{\text {TM }}$ ECL, ShQ ${ }^{\text {TM }}$ ECL Substrate Kit delivers superior performance by generating an extremely low background.

## Product Features

- Superior performance with an ultra-low background
- Suitable for detecting highly abundance analytes
- Ready-to-use kit with easy-to-follow instructions


## Storage

Stable at $4^{\circ} \mathrm{C}$ for at least 1 year.

## Kit Component

## Equal volume of Reagents A and B

## Experimental Protocol

1. Let the ECL reagents warm up at room temperature for approximately 30 min prior to use.
2. Combine Reagents A and B at a ratio of 1:1. Vortex to mix.
3. Apply the mixture to the microplate wells.
4. Immediately read the relative light unit (RLU) of the chemiluminescence signals using a chemiluminescence plate reader.

## Application Tested

Chemiluminescent ELISA

## Recommended Antibody Dilutions

Primary antibody: 1: 5,000-1:10,000
Secondary antibody: 1: 10,000-1:20,000

## Validation Data



Figure 1. Comparison of $\mathrm{ShQ}^{\mathrm{TM}} \mathrm{ECL}$ and $\mathrm{PiQ}^{\mathrm{TM}}$ ECL sensitivity in chemiluminescent ELISA. Different concentrations of gamma-irradiated SARS-CoV-2 viruses were lysed in the Viral Lysis Buffer (Cat\# VL101) and coated ( $150 \mu \mathrm{~L}$ ) on microplate wells. A rabbit monoclonal anti-N primary (Cat\# VYN7, 1:10,000) and an HRP-conjugated highly cross-adsorbed goat anti rabbit IgG secondary antibody (Cat\# 202, 1:20,000) were used to detect SARS-CoV-2 N protein. ShQ ${ }^{\text {TM }}$ ECL (Cat\# 707) and PiQ ${ }^{\text {TM }}$ ECL (Cat\# 636) Substrate Kits were used to generate chemiluminescent signals. RLU, relative light unit. $\Delta \mathrm{RLU}=\mathrm{RLU}$ of the well with the primary antibody - RLU of the corresponding well without the primary antibody, everything else being equal. Note: When the antigen is abundant, $\mathrm{ShQ}^{\mathrm{TM}} \mathrm{ECL}$ is a better choice based on linear regression data.

Table 1. Comparison of ShQ ${ }^{\text {TM }}$ ECL and $\mathrm{PiQ}^{\mathrm{TM}}$ ECL sensitivity and background noise.

|  | ShQ $^{\text {TM }}$ ECL |  |  | PiQ $^{\text {TM }}$ ECL |  |  |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- |
|  | w/o 1st Ab | $\mathrm{w} / 1$ st Ab | $\Delta$ RLU | w/o 1st <br> Ab | w/ 1st <br> Ab | $\Delta$ RLU |
| Lysis buffer | 3056 | 2927 | -129 | 130727 | 137531 | 6804 |
| Lysis buffer w/o 2nd Ab | 2860 | 2607 | -253 | 122382 | 127826 | 5444 |
| 105 TCID50 SARS-CoV-2 | 2781 | 73695 | 70914 | 128401 | 8817960 | 8689559 |
| 52.5 TCID50 SARS-CoV-2 | 2987 | 30316 | 27329 | 134264 | 5748869 | 5614605 |
| 42 TCID50 SARS-CoV-2 | 2726 | 23527 | 20801 | 128083 | 4198178 | 4070095 |
| 21 TCID50 SARS-CoV-2 | 2767 | 10342 | 7575 | 131388 | 1797070 | 1665682 |
| 8.4 TCID50 SARS-CoV-2 | 2901 | 4143 | 1242 | 129277 | 635622 | 506345 |

## Notes:

1. Different concentrations of gamma-irradiated SARS-CoV-2 viruses were lysed in the Viral Lysis Buffer (Cat\# VL101) and coated ( $150 \mu \mathrm{~L}$ ) on microplate wells.
2. A rabbit monoclonal anti-N antibody (Cat\# VYN7, $1: 10,000$ ) and an HRP-conjugated highly cross-adsorbed goat anti rabbit IgG secondary antibody (Cat\# 202, 1:20,000) were used to detect SARS-CoV-2 N protein.
3. $\mathrm{ShQ}^{\mathrm{TM}}$ ECL (Cat\#707) and PiQ ${ }^{\mathrm{TM}}$ ECL (Cat\# 636) Substrate Kits were used to generate chemiluminescent signals.
4. RLU, relative light unit. $\triangle R L U=$ RLU of the well with the primary antibody - RLU of the corresponding well without the primary antibody, everything else being equal.
