

## ABP Renilla Luciferase Assay Kit

Catalog Number: FP306, FP307

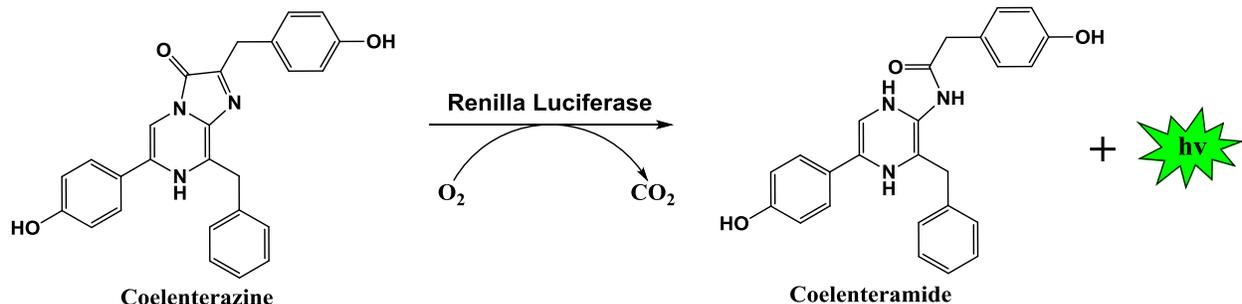
**Table 1. Kit Components and Storage**

Material	Amount	Concentration	Storage	Stability	
<b>ABP Renilla Luciferase Assay Kit (Cat. No. FP306)</b>					
Renilla Luciferase Substrate (Component A)	100 $\mu$ L	100X	-20 $^{\circ}$ C Protect from light	The product is stable for at least 6 months when stored as directed.	
Renilla Luciferase Buffer (Component B)	10 mL	1X			
<b>ABP Renilla Luciferase Assay Kit (Cat. No. FP307)</b>					
Renilla Luciferase Substrate (Component A)	500 $\mu$ L	100X	-20 $^{\circ}$ C Protect from light		
Renilla Luciferase Buffer (Component B)	50 mL	1X			

### Product Description

Transcriptional regulation using a reporter gene expression is routinely used to study a wide range of physiological events in the biotechnology and pharmaceutical industries. The traditional reporter of choice has been firefly luciferase, but recently the need for reporters that are smaller and do not require ATP has resulted in the increased use of other luciferases.

*Renilla* luciferase is a 36 kDa monomeric protein, derived from the sea pansy, *Renilla reniformis*. The enzyme does not require ATP (Figure 1), and the signal intensity is proportional to the amount of luciferase.



**Figure 1.** The Renilla luciferase reaction.

ABP Renilla Luciferase Assay Kit provides a simple and robust assay system for measuring Renilla luciferase activities from a single sample. The Renilla Luciferase Substrate and Renilla Luciferase Buffer provided with the kit, are combined to form the Renilla Luciferase Assay Reagent, which is added directly to cells in growth medium without washing or preconditioning. The assay is compatible with general used culture media with signal half-life of ~1 hr. The Renilla Luciferase Assay Reagent is used to lyse cells in culture medium and generate luminescent signal simultaneously with following features:

- **Simplicity:** Directly lyse cells in culture medium and measure luciferase activities without washing.
- **Signal stability:** The luminescent signal is highly stable with signal half-life of ~1 hr.
- **Versatility:** The assay is compatible with many eukaryotic cells (adherent or suspended).
- **High-throughput:** The assay system is adopted to high-throughput application.

### **General Considerations**

1. Culture cells in multi-well plates which are compatible with the type of luminometer for cell culture.
2. The luciferase assay is temperature sensitive, make sure the reagents be equilibrated to room temperature before measurement.
3. The luminescence signal is affected by assay conditions, results should be compared only between samples measured using the same media/serum combinations and at the same time.
4. To achieve linear assay performance at low light levels, the background luminescence must be subtracted from all readings.

### **General Protocol**

1. Place the frozen Renilla Luciferase Substrate (Component A) and Renilla Luciferase Buffer (Component B) in a water bath at room temperature. Mix well after thawing.
2. Prepare the Renilla Luciferase Assay Reagent by diluting Renilla Luciferase Substrate (Component A) at 1:100 with Renilla Luciferase Buffer (Component B). Mix well by inverting the tube several times.  
  
For example: If you are running 100 tests, add 100  $\mu\text{L}$  of Renilla Luciferase Substrate (Component A) to 10 mL of Renilla Luciferase Buffer (Component B).
3. Remove multiwell plates containing mammalian cells from the incubator, and equilibrate cultured cells to room temperature. The plates must be compatible with luminescence measurement in the luminometer being used.
4. Add a volume of reagent equal to that of culture medium in each well. Mix for optimal consistency. For 96-well plates, typically 80  $\mu\text{L}$  of reagent is added to cells grown in 80  $\mu\text{L}$  of medium. For 384-well plates, typically 30  $\mu\text{L}$  of reagent is added to cells grown in 30  $\mu\text{L}$  of medium.
5. Place the culture plates on a rocking platform or orbital shaker with gentle rocking/shaking for ~10 min.
6. Measure the luminescence signal in a luminometer (consult the instrument manual).