



Firefly Luciferase 1-Step Assay Kit

Product Information

Name :	Firefly Luciferase 1-Step Assay Kit
Catalog Numbers:	FP-BX0320 50 ml (1000 tests in 96-well plate)
Contents:	• 50 ml <i>Firefly</i> Luciferase Assay buffer • One vial D-Luciferin as powder (please add to the buffer before use)

Storage: The FLuc Assay Kit is shipped with ice bricks. Upon receipt, please store the kit at -20°C. The separate components will be stable for at least one year.

Upon thawing (at room temperature) please add the provided D-Luciferin powder to the 50 ml of buffer. Please rinse the brown vial with buffer to remove residual D-Luciferin. Mix by inverting for one minute or add a small stir-bar to the bottle. Aliquot the 'ready-to-use' buffer and store at -80°C for up to 6 months.

Introduction

Firefly luciferase is widely used as a reporter gene for studying gene regulation and function, and for pharmaceutical screening. It is a very sensitive genetic reporter due to the lack of any endogenous activity in mammalian cells or tissues. The *Firefly* luciferase is a 62 000 Dalton protein, which is active as a monomer and does not require subsequent processing for its activity. The enzyme catalyzes ATP-dependent D-luciferin oxidation by oxygen into oxyluciferin with emission of light centered on 560 nm (figure 1).

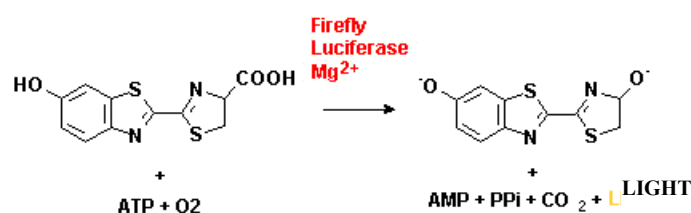


Figure 1: Bioluminescent reaction catalyzed by *Firefly* luciferase.

However, the light production resulting from the reaction leads to formation of suicidal adenyloxyluciferin at the enzyme surface. It results in very short half-life of the light emission with a flash-type kinetics. Several substances have been described to prolong light production by regenerating enzyme through removing inhibitory oxyluciferin from the enzyme surface. But the duration (10-15 min) is still too short for batch process screening.

FluoProbes' Luciferase 1-Step assay system is a proprietary mixture of substances that modify the enzymatic reaction to produce a long lasting signal (steady glow) by preventing the formation of adenyloxyluciferin at the enzyme surface. It is a homogeneous high sensitivity firefly luciferase reporter gene assay kit with a half-life of 2 hours for the quantification of firefly luciferase expression in mammalian cells. This kit is specially designed for batch processing systems using microplates such as 96-well plates. In addition, FluoProbes' Luciferase 1-Step assay kit offers higher sensitivity and wider dynamic range for detecting luciferase activity within

FT-BX032

mammalian cells (figure 2), consistent reproducibility and cost effectiveness along with the added convenience of a one step assay.

Reproducibility – CV less than 5%

Linear range- Assay linear over seven orders of magnitude

Limit of detection – less than 1 fg of luciferase per sample

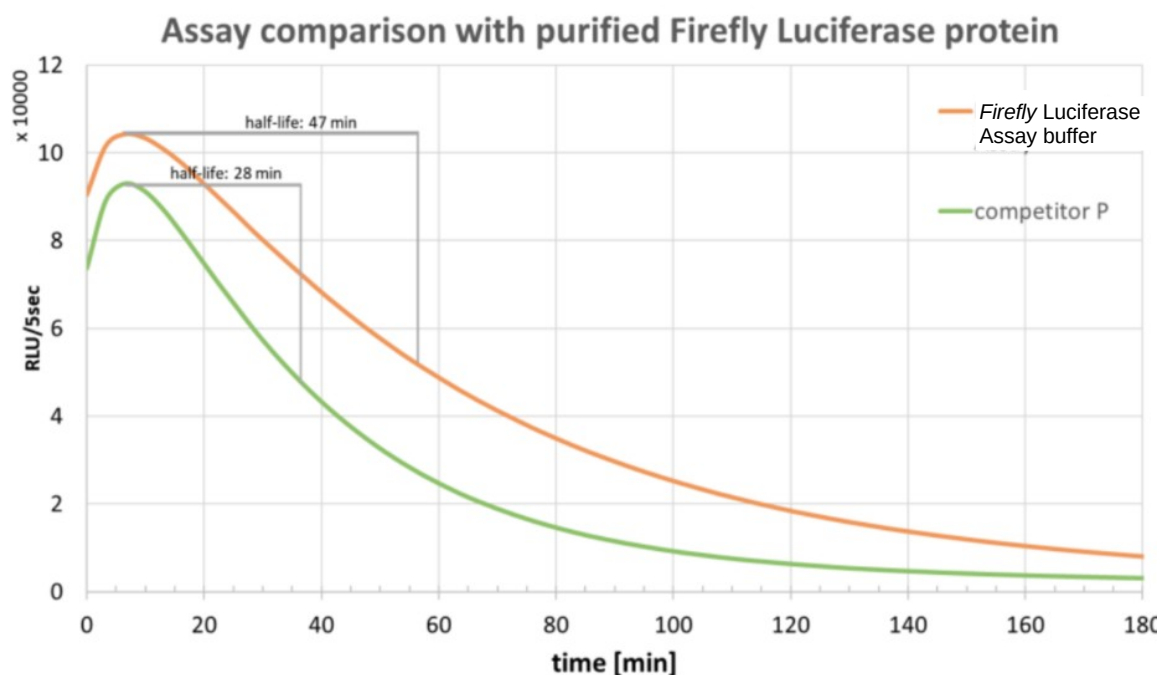
No disposal problems or hazards are associated with the use of these luciferase assay kits.

Directions for use

Protocol

1. Lysis (optional)
 - a. CAUTION! *Firefly* Luciferase Assay buffer does not contain any detergent.
 - b. If lysis is desired please add Triton-X 100 with a final concentration of 0.25% (v/v) to the buffer or use a lysis buffer of your choice in a low volume.
2. Assay preparation
 - a. Add *Firefly* Luciferase-Assay buffer directly to the cells or protein solution in a 96-well plate in a 1:1 ratio (e.g. 50 μ l FLuc-Assay buffer are added to 50 μ l HEK cells in DMEM)
3. Measurement
 - a. The signal maximum will be reached after approx. 10 min, depending on the lysis conditions and type of cells.

[A typical kinetic with *Firefly* Luciferase protein is displayed below]



Related products

- Growth plate 96x1ml, sterile [BS6200](#)
- Growth plate 96x2ml, sterile [BS6210](#)

References

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Ordering information

Catalog size quantities and prices may be found at <http://www.fluoprobes.com>

Please inquire for higher quantities (availability, shipment conditions).

For any information, please ask : FluoProbes® / Interchim; Hotline : +33(0)4 70 03 73 06

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