

CAT. & Size A2001001N (0.1 g)

A2001002N (1 g)

Storage at -20°C or below

VKEYBIO-01-2024

For Research Use Only

Not For Diagnostic Or Therapeutic Use

KeyTec® D-LUCIFERIN POTASSIUM SALT

Instruction Manual

1. Introduction

KeyTec® D-LUCIFERIN POTASSIUM SALT is designed for imaging living Firefly luciferase reporter gene cells in vitro or in vivo. It is a highly efficient, stable and reliable substrate for Firefly luciferase, producing a sensitive and robust luminescence signal. Its excellent water and fat solubility properties allow it to easily cross cell membranes.

The detection principle is based on Luminescent technology. D-Luciferin-K reacts with Firefly luciferase in cells, generating a sensitive and robust luminescence signal. The process provides a highly sensitive, robust, and homogeneous assay for the imaging living Firefly luciferase reporter gene cells, whether in vitro or in vivo. (Figure 1)

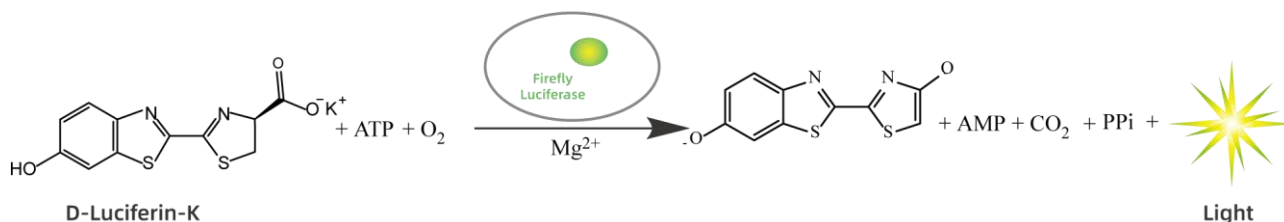


Figure 1. The Firefly luciferase reaction of KeyTec® D-LUCIFERIN POTASSIUM SALT

2. Components

CAT.	Description	Size
A2001001N	KeyTec® D-LUCIFERIN POTASSIUM SALT	0.1 g

The kit contains the following components:

- 1 × 0.1 g KeyTec® D-LUCIFERIN POTASSIUM SALT

CAT.	Description	Size
A2001002N	KeyTec® D-LUCIFERIN POTASSIUM SALT	1 g

The kit contains the following components:

- 1 × 1 g KeyTec® D-LUCIFERIN POTASSIUM SALT

3. Storage Conditions

- ◆ Upon receipt, store the kit below -20 °C in a dark place.
- ◆ Up to 1 years from date of receipt, when stored and handled as recommended.
- ◆ Once reconstituted, the stock solution must be aliquoted and stored below -20 °C in a dark place to avoid multiple freeze-thaw cycles. Use within 3 months.
- ◆ We recommend preparing the working solution immediately before use.

4. Assay Procedure

(Follow the assay protocol corresponding to the reference for the particular assay application.)

4.1 In vitro living cell

1) Reagents Preparation

- ◆ Prepare a 30 mg/mL stock solution by dissolving 1 g of KeyTec® D-LUCIFERIN POTASSIUM SALT in 33.3 mL of sterile water, or dissolve the quantity of KeyTec® D-LUCIFERIN POTASSIUM SALT necessary for an individual experiment.
- ◆ Gently tap or invert the vial to ensure thorough dissolution of the lyophilized powder, ensuring complete dissolution

2) Detection or Imaging

- ◆ Seed the Firefly luciferase reporter gene cells in a plate
- ◆ Dilute the prepared KeyTec® D-LUCIFERIN POTASSIUM SALT stock solution with cell culture medium to obtain a working solution of 150 µg/ml.
- ◆ Remove the cell culture medium and add the KeyTec® D-LUCIFERIN POTASSIUM SALT working solution. Incubate at 37 °C for 10 minutes before using for cell imaging.

4.2 In vivo living cell

1) Reagents Preparation

- ◆ Prepare a 15 mg/mL stock solution by dissolving 1 g of KeyTec® D-LUCIFERIN POTASSIUM SALT in 66.7 mL of DPBS, or dissolve the quantity of KeyTec® D-LUCIFERIN POTASSIUM SALT necessary for an individual experiment.
- ◆ Gently tap or invert the vial to ensure thorough dissolution of the lyophilized powder, ensuring complete dissolution.
- ◆ Filter sterilize using a 0.2 µm filter.

2) Imaging

- ◆ Determine injection volume at 10 µL/g of body weight. Each mouse should receive 150 mg/kg of body weight (e.g., for a 10 g mouse, inject 100 µL to provide 1.5 mg of KeyTec® D-LUCIFERIN POTASSIUM SALT).
- ◆ Inject the KeyTec® D-LUCIFERIN POTASSIUM SALT working solution intra-peritoneally 10-15 minutes before in vivo imaging, or as determined by Luciferin kinetic curve

3) Determining the Luciferin Kinetic Curve for Your Model

- ◆ A Luciferin kinetic curve should be performed for each new animal model to determine peak signal time.
- ◆ Inject the KeyTec® D-LUCIFERIN POTASSIUM SALT working solution, dissolved in DPBS at a concentration of 15 mg/mL, intraperitoneally into animals at a dose of 150 mg/kg of body weight
- ◆ Wait for three minutes, then sedate the animal using your preferred method of anesthesia, either gas or injectable anesthesia.
- ◆ Place the sedated animal in the imaging chamber and capture the first image, which will be approximately five minutes after the KeyTec® D-LUCIFERIN POTASSIUM SALT injection.
- ◆ Capture images every 5-10 minutes for up to approximately 40 minutes. This will establish the Luciferin kinetic curve for your model.
- ◆ Determine the optimal imaging time based on the Luciferin kinetic curve