

Preparation of Luciferin for *In Vitro* and *In Vivo* Bioluminescent Assays

Caution: For Research Use. This product is intended for animal research only and not for use in humans. Not for human or animal therapeutic or diagnostic use.

Preparation of Luciferin for *In Vitro* Bioluminescent Assays

Materials

- D-Luciferin Firefly, potassium salt, 1.0 g /vial (PerkinElmer Part Number #122799)
- Sterile water
- Complete media

Procedure

1. Prepare a 200X Luciferin stock solution (30 mg/ml) in sterile water. **Note:** One can either reconstitute the entire 1.0 g of D-Luciferin in 33.3 mL of sterile water to make the 30 mg/mL (200x) stock solution, or reconstitute the quantity of D-Luciferin necessary for an individual experiment.
2. Mix gently by inversion until Luciferin is completely dissolved.*
3. Prepare a 150 µg/ml working solution of D-Luciferin in pre-warmed tissue culture medium. Quick thaw 200X stock solution of Luciferin and dilute 1:200 in complete media (150 µg/ml final concentration).
4. Aspirate media from cultured cells.
5. Add 1x Luciferin solution to cells just prior to imaging. **Note:** Incubating the cells for a short time at 37 °C before imaging can increase the signal.

Preparation of Luciferin for *In Vivo* Bioluminescent Assays

Materials

- D-Luciferin, Firefly, potassium salt, 1.0 g/vial (PerkinElmer Part Number #122799)
- DPBS, w/o Mg²⁺ and Ca²⁺
- Syringe filter, 0.2 µm

Procedure

1. Prepare a fresh stock solution of Luciferin at 15mg/ml in DPBS.*
2. Filter sterilize through a 0.2 µm filter.
3. Determine injection amount at 10 µL/g of body weight. Each mouse should receive 150 mg Luciferin/kg body weight. (e.g. For a 10 g mouse, inject 100 µL to deliver 1.5 mg of Luciferin.)
4. Inject the Luciferin intra-peritoneally (i.p.) 10-15 minutes before *in vivo* imaging, or as determined by kinetic curve.**

** A Luciferin kinetic curve should be performed for each new animal model to determine peak signal time. Please see our 'Determining the Luciferin Kinetic Curve for Your Model' instruction sheet available for download on our website.

* Immediate use of working solution after dilution is strongly recommended. If necessary, dissolved luciferin may be stored at 4°C for up to 3 weeks; however prolonged storage at either 4°C or -20°C may result in degradation of signal.

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