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In Vivo Luciferin FAQs

How much luciferin should I use per mouse?

Our <u>luciferin in vivo handbook</u> suggests using 10 μ l of a 15 mg/ml luciferin stock solution per gram of body weight (10 μ l/g). In a mouse weighing 20 grams (the average mouse weighs between 17-25 grams), that would be roughly 200 μ l for a standard 150 mg/kg injection.

How many mice will one aliquotted tube of luciferin give me?

This answer depends on how you aliquot your luciferin. Referring back to question one, we determined that the average injection of stock luciferin would be 200 μ l per mouse. Therefore, you could aliquot volumes of 200 μ l, one tube per mouse, or you could aliquot 1-2 ml so that one aliquotted tube works for a group of mice.

How many mice does one luciferin stock solution provide for?

Our stock solution preparation protocol makes a 15 mg/ml solution using 1 gram of luciferin and 66.6 ml of DPBS. Total volume of stock solution shouldn't change dramatically, so we'll maintain the 66.6 ml in our math.

- In a 20 g mouse, use 200 μl luciferin stock solution.
- 66.6 ml yields a total volume of 66, 600 μl.
- 66, 600 μl / 200 μl = 333 mice

What are the injection methods for *in vivo* work?

There are 3 types of injection methods: intraperitoneal, intravenous and subcutaneous.

Intraperitoneal (IP): Here you are injecting the luciferin stock solution into the body cavity.

Intravenous (IV): This is injection directly into the tail vein.

Subcutaneous (SQ): This is injection under the skin, specifically the subcutis layer which is below the dermis and epidermis.

These methods and how to apply them are given in detail in our <u>in vivo luciferin handbook</u>. Not only will it describe how to perform the injections, but it states how soon you can do imaging after each injection type.



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How much luciferin should I buy if this is my first time doing in vivo imaging?

If this is your first time working with luciferin or working with luciferin in an *in vivo* setting, buying the smallest pack sizes (100 mg) is one of the best ways to go. This allows you to see first-hand, the quality of the product and perform a trial on a smaller scale.

Another way to look at this is to determine what the needs of your experiment are and how long the product will last. If you're comfortable with buying everything up front, then consider these factors when determining the amount to purchase:

- Storage and stability: D-luciferin potassium and sodium powder can be stored desiccated at -20°C for at least one year. Make sure to protect the powder product from light, and desiccation will really help the longevity of this product. Something else to keep in mind when it comes to storage and stability is that D-luciferin stock solutions are stable to at least 5 freeze-thaw cycles.
- Amount of luciferin per mouse and number of mice: Remember from section 1 how much luciferin stock solution would be needed for one mouse. Section 3 mentions the approximate number of mice 1 gram of d-luciferin can provide.

If you still are not sure of the best route to take, GoldBio's technical team is here to help. Call us at (800) 248-7609 or email at techsupport@goldbio.com

How long will imaging take?

Rather than thinking about the experiment in time, let's think about it in phases.

- The preparation of fresh solution or the thawing of a fresh aliquot.
- Animal injection.
- Kinetic curves for luciferase activity in each model to determine peak signal time for imaging. Continuous images are taken every 5-10 minutes for up to 40-60 minutes for IP and SQ models. IV injected models should be imaged every 1-5 minutes for up to 20-30 minutes.
- Determine the best point for imaging based on the kinetic curve. Most models reach peak signal time between 10-520 minutes post IP or SQ luciferin injection and only 2-5 minutes post IV luciferin injection.
- Once the kinetic curve and imaging point is determined, begin these phases for your actual experiment.

I've heard of AquaSparkTM, can that work with luciferin?

AquaSparkTM is a new, highly sensitive chemiluminescent substrate distributed by GoldBio. Currently available substrates are the AquaSparkTM Broad Range Phosphatase Substrate and the



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AquaSparkTM Alkaline Phosphatase Substrate. These products are separate from luciferin and are better employed for applications such as ELISA and Western blot.

However, there is future potential to develop other versions for a variety of substrates. We encourage our researchers who might be considering using AquaSparkTM to share their experiences and the potential they see for new technology.

Is luciferin made by crushing up fireflies?

Our luciferin is recombinant, so no fireflies have been used or injured in its production.

What cell lines are compatible with your luciferin?

All cell lines expressing firefly luciferase should be compatible with GoldBio's D-Luciferin potassium or sodium.

Which luciferin is better for *in vivo* use, potassium or sodium?

The only differences between potassium and sodium luciferin is small, physical characteristic differences. For example, sodium luciferin is more granular and more soluble than potassium luciferin. Most researchers prefer to use d-luciferin potassium in *in vivo* research; however, either will work equally well.

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