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Instructions for CAT#3011 Water-soluble h-Coelenterazine (hCTZ-SOL) for *in vivo* use

Content:

500 µg h-CTZ per vial of sterile, water-soluble h-Coelenterazine for the *in vivo* use with *Renilla* Luciferase

Storage:

Please store unopened vials at -80°C, for shorter periods of time at -20°C. Keep dark and dry. Reconstituted h-Coelenterazine should be used within one day[§]; refreezing is possible but not recommended.

Usage:

1. Warm vial to room temperature.
2. Add 100 µl of **sterile water** to one vial (do not use PBS) to dissolve the h-CTZ-SOL.
3. After addition of water let the powder rehydrate for 15 min, vortex the vial until completely dissolved. Let the vial sit on the bench-top until air-bubbles disappear (approx. 5-10 min). The clear yellow liquid can be further diluted with sterile 0.9% NaCl solution if desired.

<u>Desired CTZ amount per injection</u>	<u>added water</u>	<u>volume per injection</u>
100 µg	100 µl	5 injections of 20 µl
250 µg	100 µl	2 injections of 50 µl
500 µg	100 µl	one injection of 100 µl

4. Draw up the desired volume (see table above) with Insulin syringe (e.g. BD cat. # 328430). Inspect for, and remove any air bubbles in the syringe and flush the needle. Inject via tail vein to ensure optimal distribution throughout the body. The advantage of using Insulin syringes is their very low (<2µl) holdup volume! **Inject slowly.**

[§]Loss of activity is approx. 10% after 24 hours if stored as reconstituted liquid at room temperature.