

Instructions for CAT#3551 Water-soluble e-Coelenterazine (e-CTZ-SOL) for *in vivo* use

Content:

500 µg e-CTZ per vial of sterile, water-soluble e-Coelenterazine for the *in vivo* use with *Renilla* Luciferase and its mutants or other luciferases that use e-CTZ as substrate. e-CTZ will give a higher initial activity and an overall higher light output if compared to native CTZ.

Storage:

Please store unopened vials at -80°C, for shorter periods of time at -20°C. Keep dark and dry. Reconstituted e-Coelenterazine should be used within a day[§]; do not refreeze.

Usage:

1. Warm vial to room temperature.
2. Depending on the desired amount of e-CTZ use following amounts of **sterile water** (do not use PBS) to dissolve the e-CTZ-SOL:

<u>Desired CTZ amount per injection</u>	<u>added volume</u>	<u>volume per injection</u>
50 µg	500 µl	10 injections of 50 µl
100 µg	250 µl	5 injections of 50 µl
250 µg	200 µl	2 injections of 100 µl
500 µg	100 µl	one injection of 100 µl

3. After addition of water let the powder rehydrate for 5 min and vortex the vial until completely dissolved (might take up to a minute using low volume). Let the vial sit on the bench-top until air-bubbles disappear (approx. 5-10 min). 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.**

e-Coelenterazine (as water soluble form or regular powder will not work with Gaussia Luciferase. For *Renilla* Luciferase or other e-CTZ utilizing luciferases, we recommend using 100-200 µg in a 25 gram mouse, more for higher signal.

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