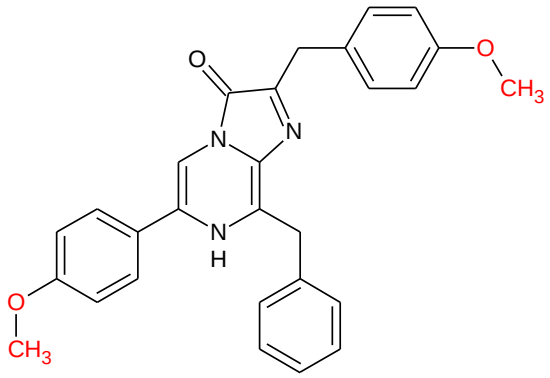
HOW TO USE "Prolume Purple II"

Cat. #367 Methoxy-COELENTERAZINE-Methoxy patent pending MW 451.52



General Notes: Methoxy-Coelenterazine-Methoxy (Me-O-CTZ-O-Me) called **"Prolume Purple II"** is a new synthetic analogue of Coelenterazine with two additional methoxy groups. This compound was developed by Nanolight™ Technology to work with **Green Renilla Luciferase (GreenRLuc)** and **Renilla Luciferase 8 (RLuc8)** to emit light at **~400 nm**.

Storage and Shelf Life: It is best stored as completely DRY powder under argon in air-tight O-ring plastic tubes at -20°C or for longer storage at -70°C, protected from light.

Usage: It is always best to make FRESH SOLUTIONS immediately before luminometer assays or experiments.

1. Dissolve lyophilized **Prolume Purple II** in **NanoFuel Solvent** as a 1 mg/ml solution (Ethanol won't dissolve Me-O-CTZ-O-Me).
2. Use this stock solution to make an aqueous solution in PBS or TBS (e.g. 50 µM for luminometer assays equal to 225.8 µl (1mg/ml) in 10 ml PBS).
3. Store dissolved Prolume Purple II at -80°C, do not store the aqueous working solution (it will oxidize over time). Prolume Purple II has the same stability in aqueous solutions like any other Coelenterazine analogue.

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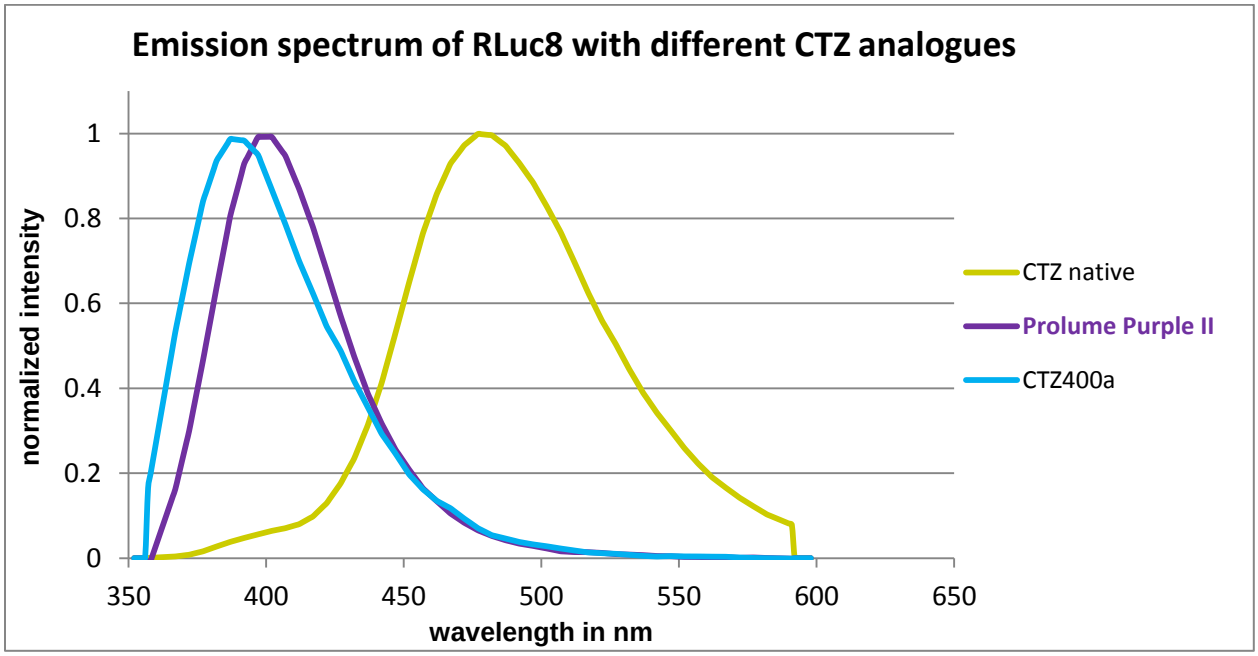
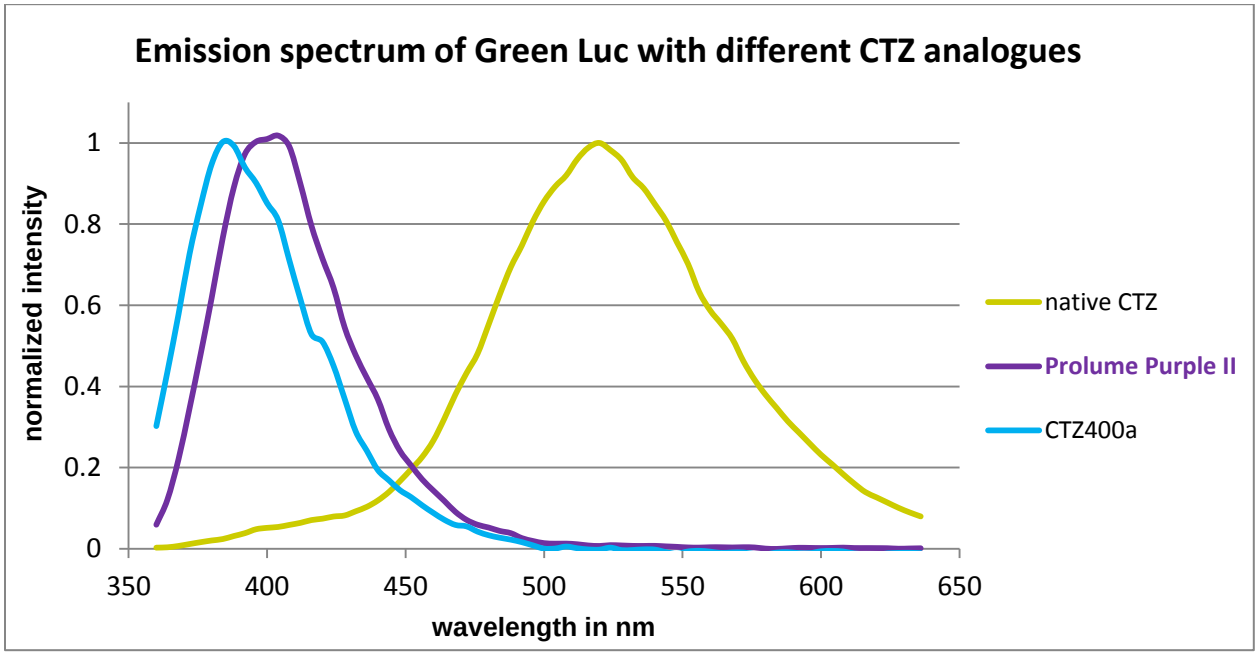
Properties of Nanolight™ Prolume Purple II Cat. #367

A. Comparison of emission spectra of CTZ analogues with *Renilla muelleri* Luciferase 8 (RLuc8)



B. Comparison of emission spectra of CTZ analogues with Green *Renilla muelleri* Luciferase (GreenRLuc)



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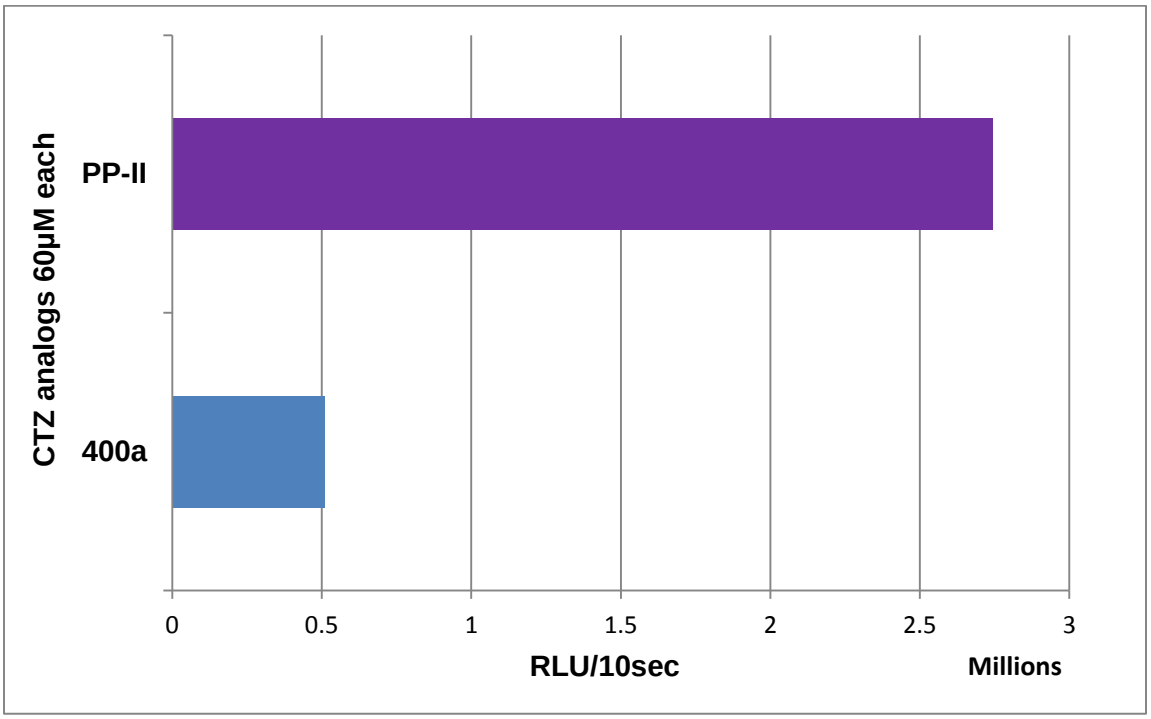
C. Comparison of luminescent intensity between "deep blue C" and "Prolume Purple II" with Green RLuc integrated over 10 sec.:



CTZ400a, also known as "deep blue C" and **"Prolume Purple II"** emit around 400 nm Green RLuc and RLuc8. The luminescent quantum yield of **"Prolume Purple II"** (PP-II) is around **5-fold higher** than CTZ400a.