

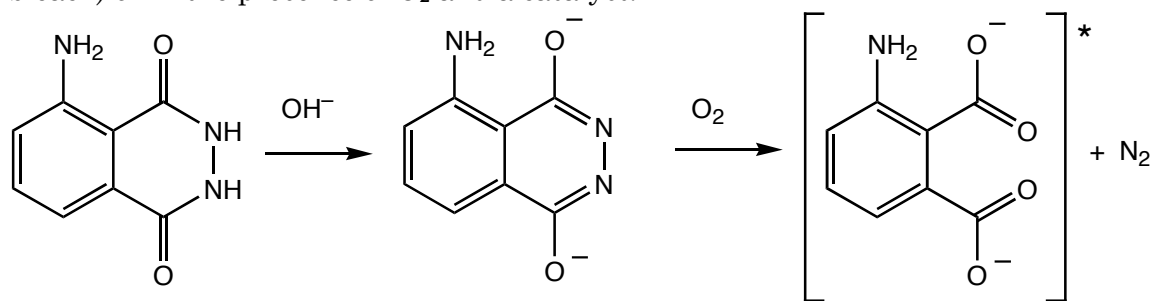
Luminol Synthesis

Objective

To develop organic lab techniques and synthesize luminol (5-amino-2,3-dihydro-1,4-phthalazinedione).

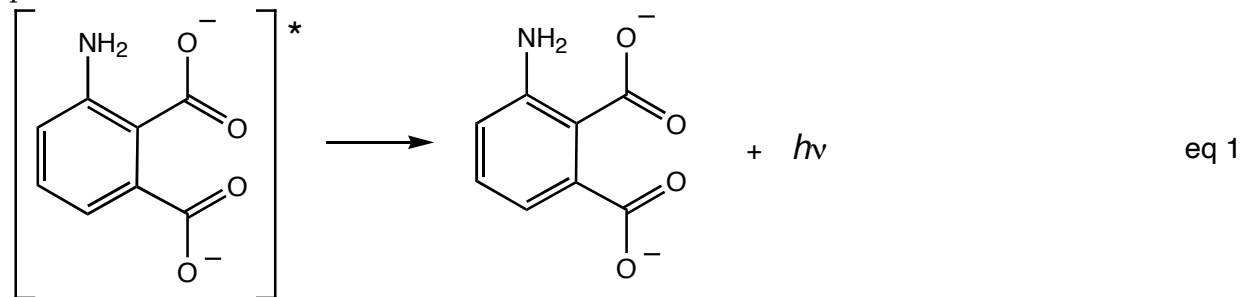
Background

Luminol is a popular star on a variety of crime scene investigation programs. Luminol luminesces when exposed to blood, and it is often used to find traces of blood at a crime scene. However, once you understand that luminol emits a photon after its oxidized, you realize that the heme in hemoglobin is simply catalyzing the oxidation of luminol. Thus, blood is not necessary; luminol will luminesce in the presence of any oxidizing agent (like bleach) or in the presence of O_2 and a catalyst.

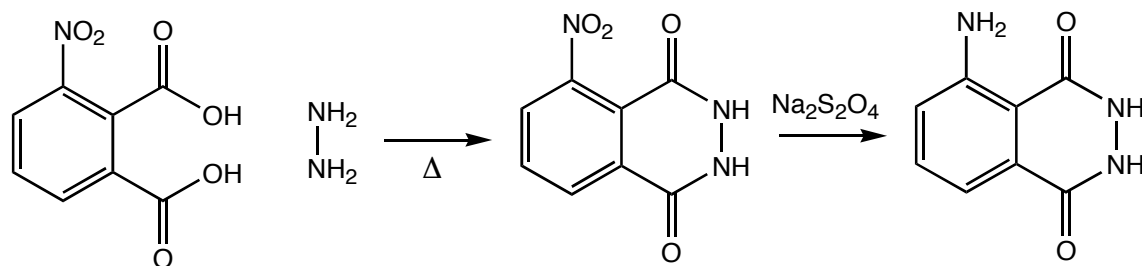


Scheme 1

The excited molecule produced in equation one relaxes to its ground state by releasing a photon.



To make luminol, we will follow the synthesis drawn in Scheme 2.



Scheme 2

Procedure¹

Synthesis of 5-nitrophthalhydrazide

Combine 0.300 g of 3-nitrophthalic acid and 0.4 mL of 10% aqueous hydrazine in a side arm test tube. Heat the test tube over a microburner until the solid dissolves. Add 0.8 mL of triethylene glycol to the test tube. Clamp the test tube to a ring stand, insert a thermometer into the test tube (use a two holed rubber stopper, one hole for the thermometer and the other to make certain that the system is **not** sealed), and connect the side arm to a vacuum source. Heat the solution to 200 °C and keep the solution at 210–220 °C for two minutes. Allow the test tube to cool to 100 °C and add 4 mL of boiling water to the test tube. Cool the suspension to room temperature by running tap water along the outside of the test tube. Collect the brown solid on a Hirsch funnel.

Synthesis of Luminol

Transfer the 5-nitrophthalhydrazide to a 18x150-mm test tube. Dissolve the solid in 1.3 mL of a 10% sodium hydroxide solution. Add 0.80 g of sodium dithionite (sodium hydrosulfite dihydrate $\text{Na}_2\text{S}_2\text{O}_4 \cdot 2 \text{H}_2\text{O}$). Rinse any solid that is clinging to the walls into the solution with 1–2 mL of water. Add a boiling stone and boil the solution for 5 minutes. Stop heating the solution and add 0.50 mL of glacial acetic acid. While agitating the test tube, cool it to room temperature by running tap water along the outside of the test tube. Collect the brown solid on a Hirsch funnel.

Luminescence

Cover the bottom of a 25-mL Erlenmeyer flask with potassium hydroxide pellets, and add enough dimethyl sulfoxide to just cover the pellets. To the mixture, add approximately 0.025 g of luminol. Cap the flask and shake the flask vigorously, but carefully. Vent the flask occasionally to release pressure and admit oxygen.

Report

Report the yield (actual and, assuming the 3-nitrophthalic acid is the limiting reagent, percent) for your product. Describe the chemiluminescence that was observed.

The first step of the synthesis of luminol was the uncatalyzed amidation of a carboxylic acid. Write a generic reaction for the reaction of an amine (RNH_2) with a carboxylic acid ($\text{R}'\text{CO}_2\text{H}$), and briefly explain why the reaction cannot be catalyzed by the addition of an acid.

¹ Adapted from Pavia, Lampman, Kiz, and Engel, "Luminol", *Introduction to Organic Laboratory Techniques: A Microscale Approach*. Saunders College Publishing, 1999.