



Luminol Test

Visitors mix a solution of luminol with fake blood (hydrogen peroxide) to produce a reaction that gives off blue light.

OBJECTIVES: Visitors learn that some chemical reactions release energy in the form of light, and that this process is called chemiluminescence.

SCIENCE TOPICS

PROCESS SKILLS

VOCABULARY

Chemical Reactions

Observing

Chemiluminescence

Chemical Bonds

Measuring

Electron

Properties of Electrons

Investigating

Energy

Light

Controlling Variables

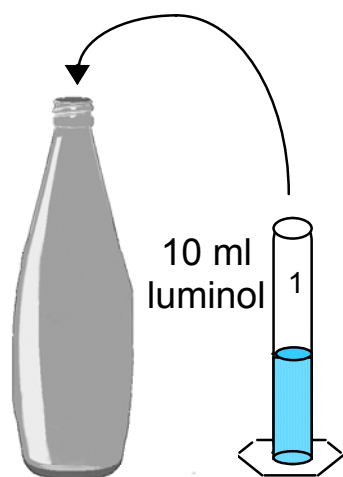
Solution

Chemiluminescence



Luminol Test

Procedure:



1. Always wear safety goggles.
2. Rinse the beaker, the two graduated cylinders, and the bottle in the sink.
3. Using graduated cylinder 1, measure 10 ml luminol solution. Carefully pour it into the dark bottle. Look into the bottle:

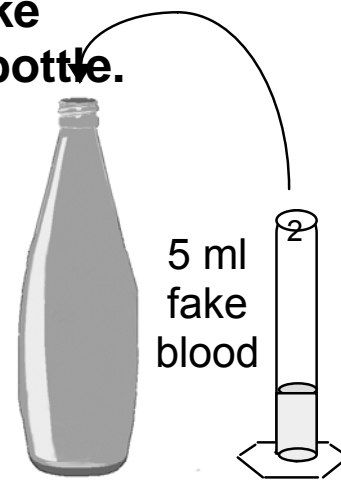
Does the luminol glow in the bottle?

4. Using graduated cylinder 2, measure 5 ml fake blood solution. Carefully pour it into the dark bottle. Look into the bottle:

Is there light in the bottle? What color is it?

5. Carefully pour the contents of the bottle into the beaker.

Is the solution the same color as the glow in the bottle?




6. Empty the beaker into the sink. Rinse the beaker, the two cylinders, and the bottle in the sink.



How do forensic scientists analyze invisible blood traces at crime scenes?

A Closer Look:



In this experiment, you observed the luminol reaction with a fake blood solution. Forensic scientists spray luminol onto surfaces at crime scenes to detect invisible blood stains. Wherever the luminol touches blood, it glows, and the glow is bright enough to be photographed. While the results can be revealing, forensic scientists usually use luminol as a last resort because it can damage DNA evidence in blood.

How does it work? Luminol solution reacts with blood to produce light. The luminol solution contains both luminol ($C_8H_7N_3O_2$) and hydrogen peroxide (H_2O_2). The hydrogen peroxide reacts with the iron in blood to produce oxygen. This oxygen then reacts with the luminol, changing the structure of the molecule and temporarily adding energy.

When energy is added to molecules, it is often absorbed by electrons (tiny charged particles). By absorbing the energy and becoming "excited," the electrons move to a higher energy level. Then, when the electrons return to their natural, "unexcited" level, they release the energy as visible light.

In this experiment the fake blood solution you used relies on copper, not iron, to help hydrogen peroxide supply oxygen to luminol.

MATERIALS

See *Materials Prep*
for more details

(with amounts to have on hand)

- One small dark opaque bottle
- Two 10-ml graduated cylinders
- Two 250-ml squeeze bottles
- NaHCO_3 (sodium bicarbonate) (keep 1000 g on hand)
- Na_2CO_3 (sodium carbonate) (keep 200 g on hand)
- $(\text{NH}_4)_2\text{CO}_3$ (ammonium carbonate) (keep 100 g on hand)
- $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ (copper sulfate) (keep 100 g on hand)
- $\text{C}_8\text{H}_7\text{N}_3\text{O}_2$ (luminol (3-aminophthalhydrazide)) (keep 25 g on hand)
- Six 500-ml brown plastic bottles
- One 1000-ml brown plastic bottle
- One 1-liter plastic bottle
- 3% H_2O_2 (hydrogen peroxide) (1 liter)
— **OR** — 30% H_2O_2 (100 ml)
- Black tape
- One 50-ml glass beaker
- red and green food coloring

Setup/Takedown Procedures

ORIGINAL SETUP

- Label the small bottle with “bottle” in white.
- Label the 10-ml graduated cylinders “1” in green and “2” in orange.
- Label cylinder 1 at the 10 ml mark, and cylinder 2 at the 5 ml mark
- Label the 250-ml squeeze bottles “ H_2O_2 (Hydrogen Peroxide)” in orange and “ $\text{C}_8\text{H}_7\text{N}_3\text{O}_2$ (Luminol)” in green.
- Label the 1000-ml brown bottle “Luminol” in green
- Label the 1 liter plastic bottle “0.15% H_2O_2 hydrogen peroxide” in orange.

WEEKLY SETUP

- Prepare fresh luminol solution; STORE IN REFRIGERATOR (see Materials Prep).



- Prepare fresh 0.15% H_2O_2 (hydrogen peroxide) solution; STORE IN REFRIGERATOR (see Materials Prep).

DAILY SETUP

- Set out the visitor instructions in a Plexiglas stand.
- On a tray, set out the following:
 - One small bottle wrapped in black tape
 - Two labeled graduated cylinders
 - Labeled $C_8H_7N_3O_2$ (luminol) squirt bottle (from refrigerator)
 - Labeled 0.15% H_2O_2 (hydrogen peroxide) squirt bottle (from refrigerator)
 - One 50-ml glass beaker
- Test the chemicals by following the experiment instructions once. If the light reaction is absent, is too brief, or takes too long to occur, the hydrogen peroxide probably needs to be replenished from stock. If it still does not work, prepare fresh 0.15% H_2O_2 (hydrogen peroxide) (see Materials Prep).
- Refill the luminol and hydrogen peroxide squirt bottles with solutions from stock bottles.

DAILY TAKEDOWN

- Rinse graduated cylinders, beaker, and bottle.
- Return all equipment to tub.
- RETURN LUMINOL AND HYDROGEN PEROXIDE TO REFRIGERATOR.

**RUNNING SUGGESTIONS**

- ◇ $C_8H_7N_3O_2$ (luminol) and H_2O_2 (hydrogen peroxide) solutions react with daylight and air; therefore:
 - Store them in opaque bottles,
 - REFRIGERATE them, and
 - Set out small amounts at a time.
- ◇ Luminol solution should be a medium blue color; if not, discard it and refill with fresher stock.
- ◇ When H_2O_2 (Hydrogen peroxide) is added to luminol, it should produce a bright blue glow lasting several seconds; if not, discard it and refill with fresher stock.
- ◇ Visitors should look in the bottle right after the second chemical is added.

- ◇ The chemicals may react too quickly if the opaque bottle is not clean.



EXTENSIONS

Examples of similar chemiluminescent reactions:

- Glow sticks
- Fireflies and some fish

SAFETY & DISPOSAL



Copper sulfate, ammonium carbonate, luminol, and hydrogen peroxide are hazardous substances; follow handling and disposal instructions in Materials Prep.

Consult Material Safety Data Sheets (MSDS) for additional information.

MATERIALS PREP

To prepare luminol solution:

- Dilute 500 ml stock solution to 1 liter with H₂O
- To prepare luminol **stock** solution:
 - CAUTION: Avoid breathing copper sulfate, ammonium carbonate, or luminol dust.**
 - To a 3 L beaker, add 1 L of water.
 - To beaker slowly add:
 - 144g baking soda
 - 28.1 g Na₂CO₃ · H₂O
 - 3 g (NH₄)CO₃
 - 2.4 g CuSO₄ · 5H₂O
 - 1.2 g luminol (add luminol last after other ingredients have (mostly) dissolved)
 - dilute to 3 liters
- Use a stir bar and stir plate to mix the items well. The luminol may take a while to dissolve.
- Fill 500-ml brown bottle(s); label the bottle “Stock Luminol Solution” in green with your initials and date.
- Note: you will need to dilute 500 ml stock solution with 500 ml H₂O o make 1000 ml luminol solution for the experiment.

To prepare 0.15% H_2O_2 (hydrogen peroxide):

- Add 50 ml of 3% H_2O_2 to 950 ml H_2O (water) (to prepare 3% H_2O_2 see instructions below).

To prepare 3% H_2O_2 (hydrogen peroxide):



CAUTION: 30% hydrogen peroxide is a strong oxidizer. Handle with care. Wear protective eyewear, gloves, and apron. Use only in the fume hood. Avoid contact with skin and clothing. If contact occurs, wash affected area with copious amounts of water.

- Wear protective eyewear, chemical safety gloves, and apron or lab jacket.
- In the fume hood, measure 25 ml 30% H_2O_2 .
- Add H_2O (water) to a final volume of 250ml.
- Store in a labeled/dated, 250-ml plastic squeeze bottle
IN REFRIGERATOR.

