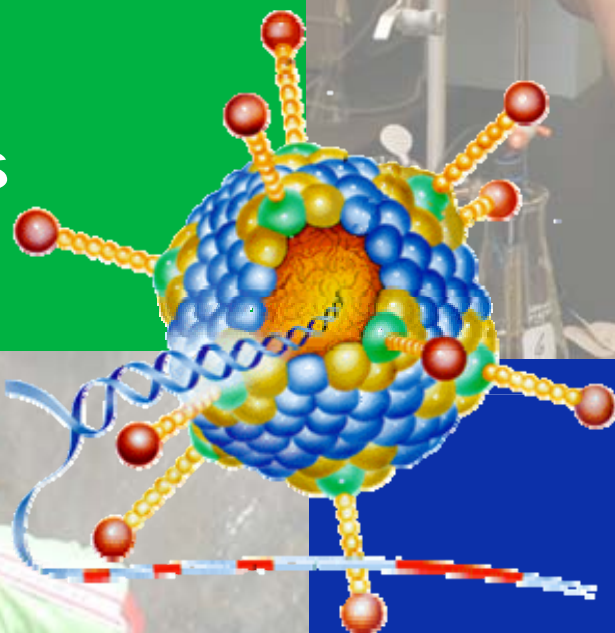


# Red Means Lead

## Teacher's Guide



*The waterCAMPWS*  
Center for Advanced Materials  
for Purification of Water with Systems



**Curriculum Connection:** Biology, Chemistry, or Environmental Sciences

**Bloom's Taxonomy:**

**Grade Level:** 11-12 AP

**Topic:** DNA enzymes, Lead detection

**Lesson Duration:** 30-60 minutes

### Purpose

The public at large probably sees little relationship between DNA and testing of lead. Yet, the two have been merged through a relatively simple color change test, red indicating lead and blue indicating no lead. Dr. Yi Lu and his graduate student Juewen Liu, at the University of Illinois, Urbana/Champaign, have developed a seemingly simple test reliably indicating the presence or absence of lead. Basing their test upon earlier research using an increase of fluorescence as a positive indicator for lead, Lu and Liu wanted a simpler test: color.

### Learning Goals and Objectives

1. Students will learn key vocabulary associated with DNA, including aggregate, Fluorescence, oligonucleotide(s), nanosphere(s), Catalytic DNA, micro-, nano- and substrate(s).
2. Students will use DNA solution to detect the presence of lead in water.
3. Students will understand the interdisciplinary qualities of science and of the study of genetics, physics, chemistry, and engineering.
4. Students will learn the qualities of DNA proteins that enable different enzymatic reactions.

### Benchmarks and Standards

#### Illinois State Learning Standards:

- *11.A.4a* Formulate hypotheses referencing prior research and knowledge.
- *11.A.4b* conduct controlled experiments or simulations to test hypotheses.
- *11.A.4c* Collect, organize and analyze data accurately and precisely.
- *12.C.4b* Analyze and explain the atomic and nuclear structure of matter.
- *13.A.4c* Describe how scientific knowledge, explanations and technological designs may change with new information over time.
- *13.A.4d* Explain how peer review helps to assure the accurate use of data and improves the scientific process.
- *13.B.4a* Compare and contrast scientific inquiry and technological design as pure and applied sciences.

#### NSES Standards: In grades 9-12, students will learn:

- *Content A* Abilities necessary to do scientific inquiry & understandings about scientific inquiry.
- *Content B* Structure and properties of matter, motions and forces, and interactions of energy and matter
- *Content C* Interdependence of organisms and behavior of organisms
- *Content E* Abilities of technical design and understandings about science and technology
- *Content F* Personal and Community health, population growth, natural resources, environmental quality, natural and human induced hazards, and science and technology in local, national and global challenges
- *Content G* Science as a human endeavor, nature of scientific knowledge
- *Teaching A* Plan an inquiry-based science program for their students
- *Teaching B* Guide and facilitate learning
- *Teaching D* Design and manage learning environments that provide students with the time, space and resources need for learning science.
- *Teaching E* Develop communities of science learners that reflect the intellectual rigor of scientific inquiry and the attitudes and social values conducive to science learning.

## Materials and Equipment

Materials required for this lesson are (for each group of students):

- The Red Means Lead Class kit from The WaterCAMPWS
  - The DNA liquid compound
  - A solution of water and lead at levels .05% and higher.
- A WaterCAMPWS specialist (to assist with disposal of the materials)
- Beakers for the solutions

Please contact Sue Herricks at [sherrcks@uiuc.edu](mailto:sherrcks@uiuc.edu) to request this class kit.

## Introduction

According to the Environmental Protection Agency, over 900,000 children in the United States currently have elevated blood lead levels. Nancy Pavur became well aware of this fact after two weeks into a home renovation when her dog, Hero, died from lead poisoning. Three years later her daughter Elizabeth began to experience memory loss, one of the effects of lead poisoning. Her other two children are now showing other detrimental effects caused by lead poisoning. The lead present in her home in the form of paint and lead pipes was released into the air and water and subsequently taken in by the family.

In most cases, it is the younger children living in the inner city in homes built before 1978 who are affected by lead poisoning. Toddlers constantly test their environment through their senses, including taste, and tend to ingest the lead in the form of lead paint chips or dust on objects in the home or in the soil around the home. Current studies have discovered that trace amounts of lead, one microgram per deciliter of blood, can result in a three quarters of a point drop in IQ. Damage to the brain and nervous system may result in the form of behavior and learning problems, slowed growth, hearing problems and headaches. While small amounts of lead can cause brain damage, discovering even high blood lead levels is difficult because traditional lead tests are expensive and inexact, oftentimes giving false-negative or false-positive results. To make matters worse, symptoms associated with lead poisoning are not immediately visible. Apparently healthy children may have very high levels of lead that acts as a neurotoxin in their blood streams. Thus, there is a clear need for easier, less expensive and more exact methods of testing for lead.

The public at large probably sees little relationship between DNA and testing of lead. Yet, the two have been merged through a relatively simple color change test, red indicating lead and blue indicating no lead. Dr. Yi Lu and his graduate student Juewen Liu, at the University of Illinois, Urbana/Champaign, have developed a seemingly simple test reliably indicating the presence or absence of lead. Basing their test upon earlier research using an increase of fluorescence as a positive indicator for lead, Lu and Liu wanted a simpler test: color.

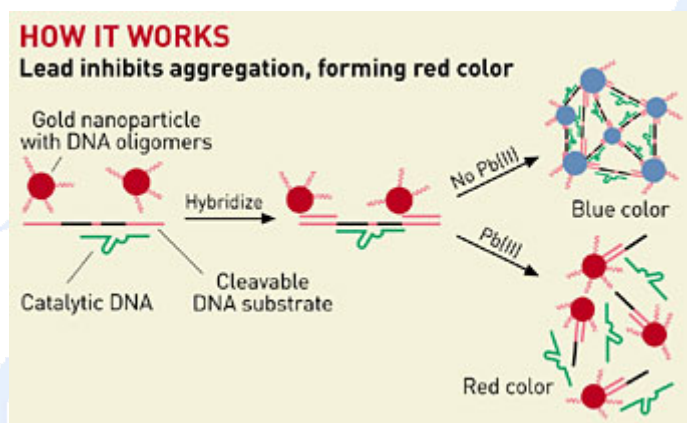
The ability to create a field test for lead coincided with the new technologies of the 1990's. The technological revolution and the resultant capability to create gold nanoparticles between 1 and 100 nanometers (nm) in diameter changed researchers' idea of "lead sensors". (View this web site for an idea of the size: <http://www.cellsalive.com/howbig.htm>). Gold nanoparticles can be adhered to a variety of substrates including silicon and bacteria cytoplasm. In addition when gold particles are linked with other substances, color changes occur. For example, the linking of gold nanoparticles to latex microspheres results in a color change from white to red; thus creating a reliable color change for that chemical reaction.

In addition to the work with gold nanoparticles, initial work in the early 1990's used DNA and RNA to recognize and bind to analytes or targets of interest. Oligonucleotides, a short DNA fragment typically of 15 or more nucleotides, are generally a single stranded, synthetic DNA molecule used

as a probe or primer. Dr. Lu and Juewen Liu used these oligonucleotides and the color change associated with the gold nanospheres to create a new reliable simple test for lead detection. However, the research team of Lu and Liu took the known methods utilizing analytes to bind with DNA or RNA and changed the reaction process to an entirely different design to indicate the presence of lead. Their method was the first to use catalytic (cleavage or breaking of a substrate) events of DNA/RNA to design a lead sensor. The test itself involves a solution of catalytic DNA, a cleavable DNA substrate and gold nanoparticles with DNA oligomers. One might think of the reaction as the board game Clue in which there is a set number of characters with multiple outcomes. In the case of the DNA sensors, there are several players but only two outcomes, A and B. Under ideal conditions in which no lead is present the color blue is outcome A. If lead is present outcome B is the color red. While the test itself is seemingly simple, it is deceptively so, as anyone who has played Clue can tell you.

The set players in our game or “test” are:

- Gold nanoparticles with DNA oligomers
- DNA enzyme or DNAzyme (the catalytic DNA)
- Cleavable DNA substrate.
- Lead

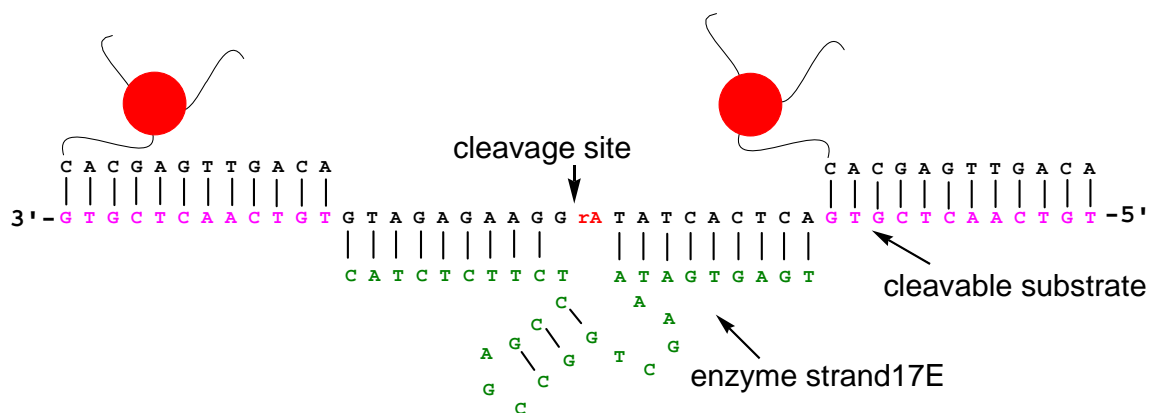


Dr. Lu and Juewen Liu chose gold nanoparticles because of previous work that demonstrated that different aggregate (crowded or massed into a dense cluster) states of the gold resulted in distinctive color changes.



Gold nanoparticles with oligonucleotides

The gold nanoparticles can be “functionalized” with DNA oligonucleotides, which are synthetically created to pair with the ends of the cleavable (breakable) DNA substrate. See illustration below.



The gold nanoparticles and associated DNA oligomers bond with the DNA substrate. The catalytic DNA acts just as a catalyst in chemistry, causing an increase reaction speed, but does not change themselves. If lead is not present in the solution, the gold nanoparticles, DNA oligomers and the cleavable DNA substrate aggregate causing the blue coloration. If lead is present, the reaction takes another turn. The lead causes the cleavable DNA substrate to break in two at the cleavage site shown above. Now, the gold nanoparticles, DNA oligomers and the cleavable DNA substrate cannot aggregate because the structures have been modified and the resultant solution color is red. Thus, the color change to either red or blue clearly indicates whether or not lead is present. This test can be easily conducted in the field by placing a solution containing the gold nanoparticles and DNA oligomers, the DNA substrate and the catalytic DNA in a test tube and adding the material to be tested. The test tube is held in the field testers hand at body temperature for about five minutes. The color change is then viewed for the presence of lead.

Further, the intensity of the red color in solution indicates the amount of lead present. The color range is likened to litmus paper in that it gives a definite color identifying the quantity of lead in solution. See figure below.



Currently the EPA standard for lead paint is 0.05%. Paint containing less than 0.05% lead is not considered to be leaded paint. According to the figure above the color change is discernable at the 0.05% level.

The test developed by Dr. Lu and Juewen Liu is a dependable, reliable and simple field test for lead. Hopefully it can be used by people such as Nancy Pavur and those living in pre 1978 buildings to eliminate the potential harmful effects of lead.

## Activities

1. Present the introduction to this lab. If possible, show the video available at <http://www.watercampws.uiuc.edu/waterclear/webquests/red-lead-webquest/RedSignalsLeadAnimation.html>
  - a. **Adaptation:** You can also request a graduate student from *The WaterCAMPWS* to help present this lecture and serve as a guest speaker.
2. Divide students into groups. Distribute to each group four beakers with four solutions of water and lead.
3. Have students make observations about the water. Allow them to smell, NOT taste, the water. Consider letting them examine the water under a microscope. Have them make a prediction about which beakers have a higher percentage of lead if possible. Random guesses are not acceptable.
4. Give each person in the group a small vial of the DNA compound. Have each student break their capsule and pour the compound into a different beaker of water.
5. Wait 5-10 minutes to see if the color changes.
6. Have the students organize the beakers by color in order of lowest to highest percentage of lead.
7. Have students complete the comprehension and analysis questions in their lab books.
8. Conclude the lab with a discussion of their results.

## Conclusion

Have the students assist The WaterCAMPWS representative clean up the lab. With remaining time, have some of the groups share their results. Collect the lab books at the end.

## Assessment

The lab book should provide an assessment tool for this lesson.

**Biology/Chemistry Activity:**

- 1) The structure labeled **(a)** acts to increase the speed of the reaction, though there is no change in the reaction pathway. \_\_\_\_\_ (Catalytic DNA)
- 2) \_\_\_\_\_, indicated by **(b)**, become functional as colorimetric indicators of lead when paired with single stranded DNA fragments that act as probes. These fragments are known as \_\_\_\_\_. ©.  
(gold nanoparticles, oligonucleotide)
- 3) DNAzymes act on the \_\_\_\_\_, labeled **(d)**, which can be cleaved in the presence of lead (II). (substrate)
- 4) A \_\_\_\_\_ colored product results in the presence of lead (II). Which figure accurately illustrates the product of this reaction? \_\_\_\_\_ (red, f)
- 5)  $10^{-9}$ m, or one billionth of a meter is equal to one \_\_\_\_\_ meter. (nano)

**Chemistry Activity:**

First, you will need to determine how much blood this individual carries. Blood represents approximately 7% of body mass. (Note that at for individuals living in areas of higher altitudes, blood represents a greater percentage of body mass. Assume 7% for this problem.) How much blood, by mass, does this individual carry?

Answer:

$$180 \text{ lbs} \times (0.4536 \text{ kg} / 1 \text{ lb}) = 81.6 \text{ kg}$$

$$\text{Male} = 81.6 \text{ kg} \approx 82 \text{ kg}$$

$$82 \text{ kg} \times (.07) = 5.7 \text{ kg}$$

What volume of blood is present in this man's body? (Blood is slightly denser than water, but assume equal densities for this problem.)

Answer:

$$5.7 \text{ kg} \times (1 \text{ L} / 1 \text{ kg}) = 5.7 \text{ L}$$

How many grams of lead can this person "safely" consume according to Canadian standards?

$$\text{Answer: } 5.7 \text{ L} \times (10 \text{ dL} / 1\text{L}) = 57 \text{ dL}$$

$$57 \text{ dL blood} \times (10 \mu\text{g Pb} / \text{dL blood}) = 57 \mu\text{g Pb}$$

$$57 \mu\text{g Pb} (10^{-6} \text{ g} / 1 \mu\text{g}) = 5.7 \times 10^{-4} \text{ g Pb}$$

$$\therefore < 5.7 \times 10^{-4} \text{ g Pb}$$

