



LOS ANGELES FRESH WATER TESTING LAB



Background: Freshwater is placed in Los Angeles from precipitation. The water either goes into the ground (which percolates into groundwater reservoirs) or runs off into the Los Angeles River. If cement is placed on the ground, creating an impervious surface, the water cannot go into ground and recharge the underlying reservoir. Cement forces the water to runoff into the nearby rivers and lakes. Along the way, the freshwater picks up any chemicals, animal waste, and pollution on the ground. This pollutes the water with nitrates, phosphates, ammonium, heavy metals, and bacteria.

Purpose: In this activity, students will pick sources of freshwater in the Los Angeles River watershed. They will then test the pH, nitrate, phosphate, turbidity, dissolved oxygen, bacteria levels, and temperature of these waterways. With this information, students will assess, using evidence, the health of the waterway.

Hypothesis: With your group, construct a hypothesis on the health of your freshwater samples around Los Angeles. Which sample sites will be polluted the most or unhealthy for living organisms? Place in your lab notebook.

Materials:

- Snap Test Kits -Thermometers -Test Tubes -Test Tube Rack
- Colorimeter -Vial for Colorimeter -Petri Dish -Agar
- Q Tip Swab -Plastic Wrap -Refractometer

Procedure:

1. Collect water samples from a freshwater source around the Los Angeles watershed. In order to collect a water sample, take a clean container and place it under the water. Try to collect a sample from a moving part of the river/lake. Immediately cap the water sample and place it in the refrigerator. Note the weather of the location, time, animals, and trash observed on a separate sheet of paper.
2. Bring the sample to class for testing. Give to Mrs. Willis for refrigeration.
3. Follow the procedures on the Test Kits to perform the nitrate, phosphate, dissolved oxygen, and pH tests. For the bacteria test, follow the agar plate procedures. For turbidity, use the colorimeter and its instructions. Rotate to all stations until all tests have been performed. Give about 10-15 minutes per station.
4. After tests have been performed, record your data in your lab notebook. In the conclusion, assess the health of the waterway based upon your evidence (data collection).

Data:

Date Water Sampled: _____ Time: _____ Location (Detailed): _____

Weather (Description): _____

Animals Observed:

Trash Observed (Detailed):

Water Tests	Measurement (ppm/FTU/# of colonies)
Temperature of Water in Lab	
Dissolved Oxygen Level	
Nitrate Level	
Phosphate Level	
pH Level	
Turbidity	
# of Bacterial Colonies on Agar Plate	
Salinity	

STATION 1: NITRATES, NO₃⁻

Nitrate, NO₃⁻, is an essential nutrient for plants, but is often limiting in the environment. Plants take up nitrate from the soil. When plants receive nitrates, they can build proteins and make DNA (assimilation). This helps them grow. Fertilizers contain much nitrate because farmers want their plants to grow large and fast. Other sources of nitrate are the burning of fossil fuels (gasoline) and animal waste. When it rains in LA, the nitrates are washed away into the local waterway. When plants receive too many nitrates, eutrophication occurs. Plants can suffocate the ecosystem and kill all organisms in the waterway. A healthy waterway will have a level of 0-10 ppm of nitrates in the water. A level of 15-25 ppm would not be safe because an infant could get blue-baby syndrome as a result. And, this high level will lead to eutrophication in a waterway.

NITRATES: PROCEDURES FOR SNAP TEST

- 1) Fill the graduated cylinder to 25 mL with your water sample.
- 2) Empty the contents of one nitrate foil pack in the graduated cylinder.
- 3) Place a piece of plastic wrap over the cylinder top and shake vigorously for exactly three minutes.
- 4) Then, pour the sample into a test tube. Allow the sample to sit undisturbed for approximately 30 seconds. This allows any undissolved particles to settle.
- 5) Place the tapered tip of the test ampoule into the test tube. Snap the tip by squeezing the test ampoule toward the side of the tube. The sample will fill the ampoule and begin to mix with the reagent inside.
- 6) Remove the fluid filled ampoule from the test tube. Mix the contents of the ampoule by inverting it several times, allowing the air bubble to travel from end to end.
- 7) Wipe all of the liquid from the exterior of the ampoule and wait **10 minutes**.
- 8) Use the comparator to determine the level of nitrate nitrogen in the sample.
- 9) Convert the sample into ppm (parts per million) by **multiplying what you get by 4.4**. Place your results in the data section.

STATION 2: DISSOLVED OXYGEN

Animals in the water need dissolved oxygen to perform cellular respiration. With high levels of oxygen in the water, a wide range of organisms can live in the water. Oxygen is placed in the water by photosynthesis of plants and phytoplankton. With low levels of oxygen, animals cannot live and biodiversity will decrease in a waterway. Adding items like oil, heat, and dirt will decrease the dissolved oxygen in the water. Sediment, for example, blocks sunlight entering into a river/lake. Thus, photosynthesis is decreased, and oxygen content in the water is lowered. Sediments often come from construction sites or agricultural fields. Oil usually enters a waterway from runoff on streets. Heat, usually from power plant effluent, causes oxygen to vaporize, thus leaving a waterway. A healthy waterway will have a level of 8 ppm of dissolved oxygen in the water.

DISSOLVED OXYGEN: PROCEDURES FOR SNAP TEST

- 1) Fill a test tube with your water sample.
- 2) Place the tapered tip of the test ampoule into the test tube. Snap the tip by squeezing the test ampoule toward the side of the cup. The sample will fill the ampoule and begin to mix with the reagent inside.
- 3) Remove the fluid filled test ampoule from the test tube. Keeping the open end downward, cover the top of the ampoule with a finger.
- 4) Mix the contents of the ampoule by inverting it several times, allowing the bubble inside to travel end to end.
- 5) Wipe all the liquid from the exterior of the tube and **wait 2 minutes.**
- 6) After waiting 2 minutes, use the comparator to determine the level of dissolved oxygen in the sample.

STATION 3: PHOSPHATE, PO₄⁻

Phosphate, PO₄, is also an essential nutrient for plants, and again is often limiting in the environment. Plants take up phosphate from the soil. Plants build proteins and DNA with phosphate. Animal waste and the decay of dead animals or producers returns phosphate to the soil. Humans apply fertilizers to their fields to help plants grow. And, many detergents contain phosphate. These detergents include car wash and laundry soap. Animal waste from feedlots and dairy farms add phosphates into the water. A healthy waterway will have a level of 5-10 ppm of phosphate in the water. Any phosphate level over 10 ppm will result in too much algal growth (eutrophication). This will lead to decreased levels of oxygen in the waterway, and the death of many aerobic species.

PHOSPHATE: PROCEDURES FOR SNAP TEST

- 1) Fill a test tube with your freshwater sample.
- 2) Add 2 drops of the activator solution to the test tube. Stir briefly.
- 3) Place the tapered end of the test ampoule into the test tube. Snap the tip by squeezing the test ampoule toward the side of the tube. The sample will fill the ampoule and begin to mix with the reagent inside.
- 4) Remove the fluid filled ampoule from the test tube. Mix the contents of the ampoule by inverting it several times, allowing the bubble to travel from end to end inside.
- 5) Wipe all the liquid from the exterior of the ampoule and **wait 2 minutes**.
- 6) Use the comparator to determine the level of the phosphate in the sample.

STATION 4: PH

pH is a measure of how much hydrogen is in a solution. A solution with a pH of 0-6 is classified as an acid. 7 is considered neutral, and 8-14 is classified as a base. The pH of a waterway determines what types of organisms live there. Usually, organisms such as fish can only handle a limited pH range. If the pH of the river/lake changes, organisms may die, and thus biodiversity decreases. Humans can change the pH of water by burning fossil fuels and creating acid rain. This acid rain enters into a lake or river and causes it to become acidic. Also, adding detergents to water may make the water too basic. Such detergents could be car wash or laundry detergents. A healthy waterway will have a pH of 7-7.5, neutral.

PROCEDURES FOR PH

- 1) Fill a test tube $\frac{3}{4}$ full with your water sample.
- 2) Grab a pH meter and take the cap off the electrode.
- 3) Turn the pH meter on and place the electrode into your test tube.
- 4) Let the electrode sit in the water sample for 2 minutes.
- 5) After two minutes, record the pH of the water sample in your data section.
- 6) Rinse off the electrode with tap water and place the cap back on the electrode.

STATION 5: TURBIDITY

Turbidity is a measure of water clarity how much the material suspended in water decreases the passage of light through the water. Suspended materials include soil particles (clay, silt, and sand), algae, plankton, microbes, and other substances. These materials are typically in the size range of 0.004 mm (clay) to 1.0 mm (sand). Turbidity can affect the color of the water.

Higher turbidity increases water temperatures because suspended particles absorb more heat. This, in turn, reduces the concentration of dissolved oxygen (DO) because warm water holds less DO than cold. Higher turbidity also reduces the amount of light penetrating the water, which reduces photosynthesis and the production of DO. Suspended materials can clog fish gills, reducing resistance to disease in fish, lowering growth rates, and affecting egg and larval development. As the particles settle, they can blanket the stream bottom, especially in slower waters, and smother fish eggs and benthic macroinvertebrates. Sources of turbidity include:

- Soil erosion
- Waste discharge
- Urban runoff
- Eroding stream banks
- Large numbers of bottom feeders (such as carp), which stir up bottom sediments
- Excessive algal growth.

PROCEDURES FOR TURBIDITY

- 1) Obtain a empty vial from the colorimeter.
- 2) Invert your water sample 3 times and then pour your water sample into the vial.
- 3) Wipe off the vial with a Kimwipe or tissue. Then, hold the vial from the cap (no fingerprints on the glass wanted).
- 4) Open the colorimeter and place the “blank” vial in the colorimeter (this is the one that is already filled with clear distilled water).
- 5) Press enter to read the clear blank sample.
- 6) Then, when done, open the colorimeter. Take the blank sample out and place your water sample vial in.
- 7) Press the enter button to read the turbidity of your sample. Record the answer in FTU in your data table.

STATION 6: TEMPERATURE

Temperature impacts both the chemical and biological characteristics of surface water. It affects the dissolved oxygen level in the water, photosynthesis of aquatic plants, metabolic rates of aquatic organisms, and the sensitivity of these organisms to pollution, parasites, and disease.

Thermal pollution is the introduction of water that is warmer than the body of water into which it flows. It generally occurs near power plants. These industries discharge hot water that has been used to cool equipment directly into streams. Another source of thermal pollution is urban runoff. This is water that has been heated as it flowed over parking lots, streets, and sidewalks. Plowing near streams or the removal of the forest canopy during construction also contributes to thermal pollution by decreasing shade, thereby increasing solar heating of the water's surface. In addition to increasing the amount of solar radiation reaching the water's surface, removal of vegetation near streams often results in increased erosion and increased amounts of sediments in the water. The sediments absorb heat from sunlight rather than reflect it. This heats the water further.

Warm water is less capable of holding dissolved oxygen. For this reason, temperature should be measured at the same place within the stream at which dissolved oxygen is measured. This allows the correlation between the two parameters to be observed.

The problem of low dissolved oxygen levels is magnified by the fact that the metabolic rates of aquatic plants increase as water temperature rises, thus increasing their biochemical oxygen demand. Low dissolved oxygen levels leave aquatic organisms in a weakened physical state and more susceptible to disease, parasites, and other pollutants.

PROCEDURES FOR TEMPERATURE

- 1) Fill a test tube $\frac{3}{4}$ full with your water sample.
- 2) Obtain a thermometer and place the thermometer in the test tube.
- 3) Let the thermometer sit for 5 minutes in the water sample.
- 4) After 5 minutes, record the temperature of the water sample (in Celcius). Place this information in your data table.

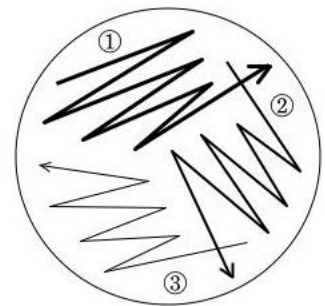
STATION 7: BACTERIA COLIFORM

Total coliform bacteria are a collection of relatively harmless microorganisms that live in large numbers in the intestines of man and warm- and cold-blooded animals. They aid in the digestion of food. A specific subgroup of this collection is the fecal coliform bacteria, the most common member being *Escherichia coli*. These organisms may be separated from the total coliform group by their ability to grow at elevated temperatures and are associated only with the fecal material of warm-blooded animals.

The presence of fecal coliform bacteria in aquatic environments indicates that the water has been contaminated with the fecal material of man or other animals. At the time this occurred, the source water may have been contaminated by pathogens or disease producing bacteria or viruses which can also exist in fecal material. Some waterborne pathogenic diseases include typhoid fever, viral and bacterial gastroenteritis, and hepatitis A. The presence of fecal contamination is an indicator that a potential health risk exists for individuals exposed to this water. Fecal coliform bacteria may occur in ambient water as a result of the overflow of domestic sewage or nonpoint sources of human and animal waste.

PROCEDURES FOR BACTERIA COLIFORM

- 1) For agar powders, dissolve by microwaving (hot plate) 6.9 g of agar in 500 ml of water.
- 2) While still hot, pour the agar solution into a sterile Petri dish. Put the cap on the Petri dish and let the agar solidify for 10 minutes.
- 3) Obtain a water sample from the school.
- 4) Place your Q-tip in the water sample for 1 minute.
- 5) Gently streak the agar plates (procedure provided by teacher) with your Q-tip that has been soaked.
- 6) Throw away your Q-tip and tape up your agar plate.
- 7) Label your agar plate with your group name and the location of the water sample. Tape the petri dish along the side with masking tape
- 8) Let the agar plate sit for 2 days.
- 9) After 2 days, count the number of bacterial colonies on the agar plate.



STATION 8: SALINITY

Salinity is the concentration of salt in water, usually measured in parts per thousand (ppt). The salinity of seawater in the open ocean is remarkably constant at about 35 ppt (3.5% salt). Salinity in an estuary varies according to one's location in the estuary, the daily tides, and the volume of fresh water flowing into the estuary.

In estuaries, salinity levels are generally highest near the mouth of a river where the ocean water enters, and lowest upstream where freshwater flows in. Actual salinities vary throughout the tidal cycle, however. Salinity levels in estuaries typically decline in the spring when snowmelt and rain increase the freshwater flow from streams and groundwater. Salinity levels usually rise during the summer when higher temperatures increase levels of evaporation in the estuary.

Estuarine organisms have different tolerances and responses to salinity changes. Many bottom-dwelling animals, like oysters and crabs, can tolerate some change in salinity, but salinities outside an acceptable range will negatively affect their growth and reproduction, and ultimately, their survival.

Salinity also affects chemical conditions within the estuary, particularly levels of dissolved oxygen in the water. The amount of oxygen that can dissolve in water, or solubility, decreases as salinity increases. The solubility of oxygen in seawater is about 20 percent less than it is in fresh water at the same temperature.

PROCEDURES FOR SALINITY

- 1) Take out the salinity meter. Remove the gray cap from the bottom of the meter.
- 2) Place the meter into a 250 ml beaker that is $\frac{3}{4}$ filled with your water sample.
- 3) Allow the meter to become accustomed to your sample for three minutes.
- 4) After three minutes, record the amount of salt in your sample: ppm.

TABLE ON SOURCES AND EFFECTS OF WATER POLLUTANTS

Pollutant Name	Sources of Material/Pollutant	Effects on Environment From Material/Pollutant
Nitrates		
Phosphates		
Total Bacterial Coliform		
pH		
Dissolved Oxygen		
Turbidity		
Temperature		

