

Blue Thumb Test Procedures

Items to take to Creek with you:

1. DO Kit (with scissors, thermometer & clear tubing)
2. Sample Bottle
3. Secchi Disk
4. Clip Board (with pen, instructions & data sheet)
5. Goggles, Gloves, Waste Container & Funnel
6. Trash Bags
7. First Aid Kit
8. Boots/Waders



A. Tasks Performed at Creek side:

1. Take air temperature in shade
2. Take water temperature
3. Record stream and testing conditions
4. Fill 2 DO Bottles upstream of a riffle (perform first part of DO test at Creek side, using packets 1, 2 & 3 to create two *fixed samples*.)
5. Fill Sample Bottle in zone of “mixed water” above/upstream of a riffle
6. Secchi Disk measurement in the deepest area of the creek

B. Back at Lab / best order to save time:

1. Finish DO Test first.
2. Ammonia Nitrogen Test (15 minutes)
3. Orthophosphate Test (15 minutes)
4. Nitrate/Nitrite Test (30 seconds)
5. pH Test
6. Chloride Test

Rinse procedures consist of rinsing the glassware 3 times with de-ionized water, then (if testing sample water) 3 times with sample water. Always rinse last with water you will test next.

WEAR
PROTECTIVE
GEAR!!!





I. Dissolved Oxygen Test

Steps for Creek side

1. Fill both DO bottles **upstream of a riffle**. Insert one end of the clear tubing all the way into and touching the bottom of one DO bottle (hold tube there). With your other hand put your thumb/finger over the other end of the tubing. Insert the DO bottle in the water upside down “wrist deep” (don’t touch the bottom of the creek) while your other hand at the end of the tubing stays up/dry out of the water. Remove your thumb/finger from the end of the tubing, this will allow the tubing to fill the DO bottle with water. When DO bottle is full (its ok to have a small bubble) and still under the water, pull the tubing out, invert the bottle, lift the bottle straight up to the surface. Fill 2nd bottle the same way.
2. Open one **DO #1** and one **DO #2** for each DO bottle. Add the contents of each pillow to one bottle. Stopper the bottle (over the waste container) to exclude air bubbles, pour off excess liquid into liquid waste.
3. Grip the bottle and stopper firmly; shake vigorously to mix. A flocculant (floc) precipitate will be formed. If oxygen is present in the sample, the precipitate will be brownish orange in color. A small amount of powdered reagent may remain stuck to the bottom of the bottle. This will not affect the test results. Repeat steps 2 and 3 for second bottle.
4. Allow the sample to stand until the floc has settled below the lines on the bottles, leaving the upper half of the sample clear or for five minutes.
5. Shake vigorously the bottles again so the floc is gone.
6. Allow the samples to stand again until the floc has settled below the line on the bottle.
7. Remove the stopper from one bottle and add the contents of one **DO#3**. Carefully restopper the bottle over the liquid waste container. To avoid trapping air bubbles in the bottle, drop the stopper from the top of the bottle. (If bubbles do become trapped gently remove stopper and retry dropping the stopper.) Pour off excess liquid in liquid waste container. Repeat for 2nd sample.
8. Grip the bottle and stopper firmly; shake vigorously to mix. The floc will dissolve and a golden color will develop if oxygen is present. (If the liquid in both bottles is not the same color, discard the samples and start again.) The samples are now “**fixed**” and you may take it to the location where you finish monitoring.

Steps for Lab

DO - High Range

9. Fill the skinny plastic measuring tube with a reverse meniscus with the prepared sample. Invert the square mixing bottle over the measuring tube and then pour the sample into the square mixing bottle.
10. Add two drops of **Starch Indicator** solution to the square mixing bottle. Swirl to mix. The sample will turn dark blue/black.
11. Add **Sodium Thiosulfate** standard solution drop by drop to the mixing bottle, swirling to mix after each drop. Hold the dropper vertically above the bottle and count each drop as it is added. Continue to add drops until the sample changes from blue to colorless. Multiply the number of drops by 1 to get mg/L.
12. Record your results on the data sheet. **If your result is 3 mg/L of oxygen or less, continue with the low range test** that follows. Use rinse procedure and repeat test for 2nd sample. Both samples should be about the same.

DO - Low Range

13. Pour off the remaining “fixed” sample so that 30 ml remains in the round bottle. The line on the bottle is at 30 ml.
14. Add two drops of **Starch Indicator** solution to the round bottle. The sample will turn dark blue/black.
15. Add **Sodium Thiosulfate** standard solution drop by drop to the round bottle, swirling to mix after each drop. Hold the dropper vertically above the bottle and count each drop as it is added. Continue to add drops until the sample changes from blue to colorless. Multiply the number of drops by 0.2 to get mg/L.
16. Record your results on the data sheet. Use rinse procedure and repeat test for 2nd sample.

Percent Oxygen Saturation

Enter the water temperature and the mg/L of dissolved oxygen into the chart in the data sheet. (Use the lowest mg/L DO. DO NOT average.) Draw a line with a straight edge between the two to calculate the % oxygen saturation.



II. Ammonia Nitrogen Test

(15 minutes for all three)

Low Range Test *(run the blank only at this Low Range, even if your sample water needs to be run at the Mid Range)*

1. Follow rinse procedure.
2. Fill one test cube to the mark with deionized water (the blank) and two test cubes to the mark with the water sample.
3. Open and add the contents of the **Ammonia Salicylate** reagent. Cap and shake gently to mix until reagent is dissolved.
4. Open and add the contents of the **Ammonia Cyanurate** reagent. Cap and shake gently to mix until reagent is dissolved. Solution should be yellow like the lowest color level on the cube.
5. Time 15 minutes for color development.
6. Match the color of the samples to the color on the cubes.
7. Estimate results if color of the samples falls between two color blocks.
8. Record your results on the data sheet.

Mid Range Test

1. Follow rinse procedure.
2. *Run the blank using the Low Range Test.*
3. Follow rinse procedure.
4. Use the dropper to put 1.0 ml of sample water in a **test tube**. Add deionized water to fill the test tube to the 5 ml mark. Shake well to mix.
5. Fill **test cube** to the mark with the diluted water sample from the test tube.
6. Open and add the contents of the **Ammonia Salicylate** reagent. Cap and shake gently to mix until reagent is dissolved.
7. Open and add the contents of the **Ammonia Cyanurate** reagent. Cap and shake gently to mix until reagent is dissolved. Solution should be yellow like the lowest color level on the cube.
8. Time 15 minutes for color development.
9. Match the color of the sample to the color on the cube.
10. Estimate results if color of the sample falls between two color blocks.
11. Multiply your reading by 5 to get mg/L. Record your results on the data sheet.



III. Orthophosphate Test

(8 minutes each, about 15 minutes total)

Low Range Test (0-0.3 mg/L)

(run the blank only at this Low Range, even if your sample water needs to be run at the Mid Range or High Range)

1. Follow rinse procedure.
2. Fill a square mixing bottle to the 20 ml mark with the water to be tested. (The first test should be the blank using deionized water. When it is more than half way through the color development phase start the first stream sample in another square mixing bottle. When it is more than half way through the color development phase start the second stream sample.)
3. Open one **PhosVer 3** phosphate reagent powder pillow. Add the contents of the pillow to the bottle (**start timing for 7.5 minutes**). Cap mixing bottle with plastic thimble and gently shake to dissolve reagent.
4. Insert the lengthwise viewing adapter (the mirror) onto the shelf in the black box comparator. The *glass* test tubes will rest on the mirror. Put the color wheel on the spindle.
5. Fill one *glass* test tube with untreated water to the top white line. Insert this tube in the opening of the color comparator behind the colored portion of the wheel.
6. At 7.5 minutes fill the empty *glass* test tube to the top white line with the prepared water sample and place in opening of the black box comparator behind the clear portion of the wheel.
7. **Read at 8 minutes.**
8. Orient the comparator with the tube *tops* pointing to a window or light source. View through the openings in the front of the comparator. When viewing, use care not to spill samples from the tops of the tubes.
9. Rotate the color wheel to obtain a color match. Read the concentration of the measured parameter through the scale window. Record the reading.
10. Divide by 150 to calculate mg/L and record your answer. For your convenience, the data sheet has some of the most common calculations in a table. If your reading is 40 or more and the calculated answer is 0.25 mg/L or more, do the mid-range test.
11. Repeat test for remaining samples.

OP - Mid Range Test (0-1.7 mg/L)

1. Follow rinse procedure.
2. *Run the blank first and use the Low Range Test.*
3. Follow rinse procedure.
4. Remove the lengthwise viewing adapter (mirror).
5. Fill both of the *glass* test tubes to the 5 ml mark with the water to be tested.
6. Insert one tube in the opening of the black box comparator behind the colored portion of the wheel. Be sure you have removed the lengthwise viewing adaptor (mirror).
7. Open one **PhosVer 3** phosphate reagent powder pillow. Add the contents to the other tube. Swirl to dissolve reagent. Place in the opening of the color comparator behind the clear portion of the wheel.
8. **Read at 1 minute.**
9. Hold the comparator up to a light source like a window or a lamp and view through the two openings in front.
10. Rotate the color wheel to obtain a color match.
11. Read the concentration of the measured parameter through the scale window. Record the reading.
12. **Divide by 30** to calculate mg/L and record your answer. If your reading is 40 and your calculated answer is 1.3 mg/L or more, do the high range test.
13. Repeat test for second sample.

OP - High Range Test (0-16.7 mg/L)

1. Follow rinse procedure.
2. *Run the blank first and use the Low Range Test.*
3. Follow rinse procedure.
4. Remove the lengthwise viewing adapter (mirror).
5. Fill one of the *glass* test tubes to the 5 ml mark with **deionized** water. This is the comparator tube. Place it in the opening of the color comparator behind the colored portion of the wheel.
6. Use the dropper to put 0.5 ml of sample water in the other glass test tube. **Add deionized water** to fill the test tube to the 5 ml mark.
7. Open one **PhosVer 3** phosphate reagent powder pillow. Add the contents to the tube with the sample water. Swirl to mix. Place in the opening of the color comparator behind the clear portion of the wheel.
8. **Read at 1 minute.**
9. Hold the comparator up to a light source like a window or a lamp and view through the two openings in front. Rotate the color wheel to obtain a color match.
10. Read the concentration of the measured parameter through the scale window. Record the reading.
11. **Divide by 3** to calculate mg/L and record your answer.
12. Repeat test for second sample.



IV. Nitrate/Nitrite Test Strips

1. Follow rinse procedure and fill one test tube with sample water.
2. Thoroughly saturate the pads on two test strips with sample water by dipping and swirling them in the test tube. Do not shake excess water from the strips.
3. Hold the strips level, with pad side up.
4. At **30 seconds** compare the **Nitrite** test pads closer to your thumb to the bottom color chart on the bottle.
5. At **30 seconds** compare the **Nitrate** test pads on the end of the strips to the top color chart on the bottle.
6. Estimate results if the color on the test pads falls between two color blocks.
7. Enter results on data sheet.



V. pH Test

1. Follow rinse procedure.
2. Fill both plastic test tubes to the 5 ml mark with the water sample. (5 ml is marked on the test tube where the tube turns from clear to frosted near the bottom of the tube.)
3. Add six drops of **Wide Range pH Indicator** solution to one of the test tubes and swirl to mix.
4. Insert the sample with reagent added in the inside opening of the black box comparator.
5. Insert the test tube with the untreated sample in the outside opening of the comparator. Put the color wheel on the spindle.
6. Hold the color comparator up to a light source such a window, a lamp, or something white and view through the two openings in the front. Rotate the color wheel to obtain a color match. Read the pH from the scale window. You may interpolate.
7. Record your data on the data sheet.
8. Repeat test for second creek water sample.



VI. Chloride Test

Low Range Test (0-100mg/L)

(run the blank only at this Low Range, even if your sample water needs to be run at the High Range)

1. Follow rinse procedure.
2. Fill the mixing bottle with deionized water (the blank) to the 23 ml mark.
3. Add the contents of one **Chloride 2** indicator powder pillow. Swirl to mix.
4. Add the **Silver Nitrate Titrant** drop by drop to the water in the mixing bottle. Hold the dropper in a vertical position and swirl the bottle to mix after each drop is added. Count each drop as it is added until the color changes from bright yellow to a slight orange. (An orange-red, rust color indicates the end point has been exceeded and the test must be redone.) **Do not add more than 20 drops.** If you need more than 20 drops, use the high range test.
5. Record your raw data on the data sheet.
6. **Multiply by 5** to calculate mg/L chloride and record your answer. If your calculated answer exceeds 100 mg/L, retest using the high range test.
7. Repeat test twice using sample water.

High Range Test (0-400 mg/L)

1. Follow rinse procedure.
2. *Run the blank first and use the use the Low Range Test.*
3. Follow rinse procedure.
4. Fill the plastic measuring tube with a reverse meniscus with sample water. Pour it into the rinsed mixing bottle.
5. Add the contents of one **Chloride 2** indicator powder pillow. Swirl to mix.
6. Add the **Silver Nitrate Titrant** drop by drop to the water in the mixing bottle. Hold the dropper in a vertical position and swirl the bottle to mix after each drop is added. Count each drop as it is added until the color changes from yellow to a slight orange. (Orange-red, rust color indicates the end point has been exceeded and the test must be redone.) **Do not add more than 20 drops.** If you need more than 20 drops, do a dilution.
7. Record your raw data on the data sheet.
8. **Multiply by 20** to calculate mg/L chloride and record your answer.
9. Repeat test with second sample.