

**Complete document includes
the methods and manuals
for water analysis that
came with each instrument.**

Methods Protocol

Water Quality Assessment

General Procedures

Lycoming College interns operate out of a small research laboratory on the campus of Lycoming College in Williamsport. All operations are overseen by Dr. Mel Zimmerman. All data is recorded either on site or in the lab on standardized water chemistry sheets in pencil.

Custody of samples and data:

1. The intern is responsible for the proper collection of samples in the field. The water sample will be collected into plastic, square-screw cap, LEXAN bottles. The samples will be obtained from a site off shore of the river at a point which a visible current may be observed. Bottles will be filled and dumped two times to prevent contamination. Two bottles will be collected from each site to account for any possible errors. All samples will be stored in a cooler packed with ice for transport to the laboratory.
2. Once delivered to the lab the intern will analyze samples immediately or store them under refrigeration for a period of no more than 24 hours from the time of sample collection.
3. All data generated will be filed in appropriately labeled binders and cataloged in the research lab along with field data sheets and any other text concerning the project.
4. Reports will be reviewed by the project advisor and sent to all appropriate recipients.

Quality Assurance:

1. All interns undergo training concerning protocols and techniques.
2. Standardized data sheets are used to ensure data results are organized and legible.
3. Chemical reagents are checked for expiration dates; expired reagents are disposed of properly.

Quality Control:

1. Each instrument, when turned on, completes an automated or manual self test and/or calibration.
2. Where applicable, a minimum of 10% of all sample analyses will be conducted for quality control purposes.
3. Where applicable, one blank, one mid-point standard, one high-point standard, one spike, and one set of duplicates will be run with each batch of samples.
4. Data accuracy will measure how closely the results come to the actual value of the parameter being measured. Standards and spikes will evaluate test accuracy.
5. Data precision will measure how well the test results can be produced. Duplicates, standard sampling and standard deviation will evaluate test precision.

Safety:

1. All chemicals and acids are stored according to MSDS procedures on shelves in the lab or on tables adjacent to proper equipment.
2. All equipment is employed and cleaned according to the manufacturer's instructions as printed in the user manuals. All manufacturer's safety methods and procedures are strictly adhered to.

Instrumentation:

All equipment is employed, stored, and maintained according to the manufacturer's instruction. All instruments are used and calibrated on a regular basis.

pH:

Several types of pH meters are available, the first printed is the preferred and more commonly used.

1. OAKTON pH510 aided by the Fisher Scientific model 120S
2. HACH sensION 2
3. Orion portable pH/ISE 230A

Spectrophotometer:

1. HACH DR4000U
2. HACH DR2000
3. HACH DR3000

The preferred instrument for all testing is the HACH DR4000; for reasons of time conservation low-medium range nitrate and nitrite data may be analyzed in the HACH DR2000 and orthophosphate may be measured in the HACH DR3000.

Conductivity Meters:

1. HANNA instruments model HI8733
2. OAKTON model CON410

Pipettes:

1. HACH tenset pipettes

Dissolved Oxygen Meter:

1. YSI 55

Sample Preservation

Measurement	Container	Location of Data Collection	Holding Time	Preservation Method
pH	LEXAN	Lab	≤24	Cold Room Storage
Dissolved Oxygen	n/a	On Site	n/a	n/a
Alkalinity	LEXAN	Lab	≤24	Cold Room Storage
Nitrate	LEXAN	Lab	≤24	Cold Room Storage
Nitrite	LEXAN	Lab	≤24	Cold Room Storage
Orthophosphate	LEXAN	Lab	≤24	Cold Room Storage
Phosphorous	LEXAN	Lab	≤24	Cold Room Storage
Temperature	n/a	On Site	n/a	n/a
Conductivity	LEXAN	Lab	≤24	Cold Room Storage

Methods: (See Appendix 1 for copies of methods)

~ pH – according to OAKTON pH/ION 510 instruction manual

~ Dissolved Oxygen – according to YSI 55 operating manual

~ Alkalinity – as addressed in the OAKTON pH/ION 510 Instruction Manual

~ Nitrate – DR4000: HACH method 10020 program 2511 (0 to 30.0 mg/L NO₃⁻ -N)

DR2000: HACH method 8171 program 353 (0 to 4.5 mg/L NO₃⁻ -N)

~ Nitrite – DR4000: HACH method 10019 program 2630 (0 to .500 mg/ NO₂⁻ -N)

DR2000: HACH method 8507 program 371 (0 to .300 mg/ NO₂⁻ -N)

~ Orthophosphate – DR4000 HACH method 8048 program 3035 equivalent to USEPA method 365.2 and Standard Method 4500-PE (0.00 to 5.00mg/L PO₄⁻³; 0.00 to 1.60mg/L P)

DR3000 Standard Method 4-108 (0 to 5.00 mg/L PO₄⁻³)

~ Phosphorous – DR4000 HACH method 8180 program 3037 (0.00 to 5.0 mg/L PO₄⁻³; 0.00 to 1.60 mg/L P)

~ Temperature – according to OAKTON 35608 series operating instructions manual

~ Conductivity – according to OAKTON 35608 series operating instructions manual

Sampling Equipment and Methods

Data Recording: To be recorded on appropriate pre-fabricated data sheet in pencil only.

Sample Takers: Label water chemistry sheets with last name and record date; label bottles with site location, date, and last name

Dissolved Oxygen: To be recorded in mg/L with YSI 55 dissolved oxygen meter

Stream Temperature: To be recorded in degrees centigrade with YSI 55 dissolved oxygen meter

Stream pH: To be obtained by (preferably) the OAKTON pH510 or HACH sensION 2

Conductivity: To be obtained by OAKTON CON410 – 35608 series or HANNA instruments model HI8733

Water Sample: The water sample will be collected by the hands of the sample taker and LEXAN, square, screw cap, and bottles. The bottles will be rinsed and dumped two times before the final sampling. The bottles will then be immediately packed in ice in preparation for transport to the lab.

Appendix I

1. Background Information on Chemicals to be Tested
2. Water Chemistry
3. OAKTON pH/ION 510 Instructions Manual
4. Orion pH/ISE Instructions Manual
5. HACH sensION 2 Instructions Manual
6. HACH DR4000U Nitrate Methods
7. HACH DR2000 Nitrate Methods
8. HACH DR4000U Nitrite Methods
9. HACH DR2000 Nitrite Methods
10. HACH DR4000U Orthophosphate Methods
11. HACH DR3000 Orthophosphate Methods
12. HACH DR4000U Phosphate Methods
13. OAKTON Conductivity Instructions Manual
14. HANNA H18733 Conductivity Instructions Manual

Background Information on Chemicals to be Tested

Dissolved Oxygen

In water oxygen is present in a dissolved form and measured as milligrams of gas per liter (mg/L) which can also be referred to as parts per million (ppm). The amount of oxygen present in water is dependent on a number of variables including temperature, flow, aquatic plants, bacteria, altitude, and dissolved or suspended solids. Human activities such as urban development, dams, removal of riparian vegetation, and organic and nutrient inputs may also affect dissolved oxygen levels.

The amount of dissolved oxygen in a stream is a direct result of gas transfer from the atmosphere to surface waters. This is aided by wind, waves, and any other turbulence churning the water. Oxygen is also introduced into the water in great amounts by aquatic plants through photosynthesis. However, at night the same plants decrease the oxygen levels through respiration.

Water's ability to hold dissolved oxygen is decreased as the temperature rises. Thus, during the hottest time of the year and when plant respiration is greatest, the dissolved oxygen levels of streams are generally at their lowest. For these reasons, temperature is very important when analyzing water for dissolved oxygen content.

Human activities may directly or indirectly affect the dissolved oxygen content of streams. Organic pollution and nutrients introduced to the water by human use of fertilizers, manure piles, lawn waste (when dumped in or near streams), and inadequate sewage treatment facilities stimulate the growth of algae and bacteria which in turn use up the oxygen. Dams built by humans also decrease the oxygen levels by increasing water temperature.

As dissolved oxygen content levels drop animals that inhabit the streams begin to feel stressed. At a level of 5.0 mg/L organisms become stressed to a point that a noticeable decrease in diversity is recognized. Hardier species of animals may be able to survive in levels below 4.0, however 2.0 mg/L is considered an absolute lethal level at which even the hardier species die off.

pH

pH is a measure of hydrogen ions and hydroxyl ions determining how acidic or basic a solution is. The pH scale runs from 0-14 with 7 being a neutral number, higher numbers signify a more basic solution and lower a more acidic; average stream pH is generally between 6 and 9.

Stream pH may be affected by a number of variables including rainfall (both acidic and natural unpolluted), soil types, aquatic plants, sediments containing metals, and mine drainage.

Natural unpolluted rain may have a pH as acidic as 5.6, as it falls through the atmosphere, it absorbs carbon dioxide and forms carbonic acid. Where soil types are alkaline, as in limestone streams, the pH may be greater than 7, which gives the stream a greater buffer level. The buffer level allows the stream to maintain a constant pH even when large amounts of acid or base are introduced into the water.

Aquatic plants may also impact pH levels. As plants undergo photosynthesis during the day and take in carbon dioxide, the pH increases. However, at night during respiration, the plants give off carbon dioxide decreasing the pH.

Aquatic organisms prefer a range of 6.5 to 8.0. pH levels around 4 or below may destroy larva and eggs; result in fish kills, and/or cause mutations. Although not as well studied or documented, it is likely that high pH levels also cause mutations. In regions where acid mine drainage is present, pH levels are decreased and often result in streams being intolerable to wildlife. Acid rain is also known to create these situations and decrease pH levels, especially in Pennsylvania which receives some of the most acidic rain of any state.

Nitrogen

In aquatic systems nitrogen is found in various forms including nitrate, nitrite, and ammonia. In water nitrogen is converted into usable forms by bacteria and then taken up and used by algae. It is essential to plant growth in the water. However, an over abundance of nitrogen directly causes an increase in plant growth and may cause and overgrowth. The overgrowth of plants will result in more live plants as

well as more dead plant matter in the water. The live plants block the sunlight from the water column while the decomposing plant matter provides food for algae, increasing the amount of algae in the water decreases the dissolved oxygen content.

Nitrogen levels are affected by various natural occurrences such as acid rain, the freezing and thawing of soils, forest fires, recycling by vegetation, as well in many human influences. Human influence such as improperly treated sewage, leaky septic tanks, over fertilization of fields, lawns and golf courses, improper disposal of pet/livestock waste, detergents, and industrial effluent all increase levels of nitrogen in streams. These increases have been linked to causing blue baby syndrome and methemoglobinemia. Both illnesses are a direct result of the high levels of nitrogen in the drinking water interfering with hemoglobin's ability to carry oxygen through the blood.

In areas rich in agriculture and industry, nitrogen levels in well water may increase. 10mg/L nitrate-nitrite is considered a level at which water is no longer safe to consume, any level above 1.0mg/L signifies an unnatural input of nitrogen into a stream.

Phosphate

Phosphate is found in two forms in water: organic phosphate which is attached to particles in the water such as plant or animal matter and inorganic phosphate also known as ortho-phosphate which is dissolved in the water and thus is more readily available to plant life. Normally phosphate levels are low, around 0.01mg/L; levels reaching 0.1 mg/L or above indicate pollution.

Phosphate is affected by various human activities including improperly treated sewage, leaky septic tanks, over fertilization, commercial cleaning operations, deforestation, and draining of wetlands. Each of these activities increases phosphate levels which sequentially increase plant productivity. When the plants die, the process of decay increases algae population levels and thus deprives the stream of dissolved oxygen.

Turbidity

Turbidity refers to the clarity of water in relation to levels of total dissolved solids (TDS) in the water. TDS along shores consists of particles of clay and silt, and throughout the stream re-suspended bottom sediments stirred up by wind, weather, and human activities.

Activities that affect turbidity include dredging operations, increased flow rates, floods, increased populations in bottom-feeding fish, and increased erosion, organic matter and nutrients due to poor land usage. These actions cause increased levels of turbidity which may cause shallow areas to fill in faster, as well as impairment of aquatic habitat. Increased turbidity negatively affects aquatic life by smothering benthic habitat (suffocating larva etc), interfering with particle feeding, introducing disease, damaging gill structure, and reducing light penetration needed for photosynthesis.

As far as effecting humans, increased turbidity makes the water aesthetically unpleasing, and requires greater processes to clean up water for human consumption.

Alkalinity

Alkalinity is also known as the buffering capacity of water or the capacity of acids to neutralize bases. Alkalinity data is important in determining a streams ability to maintain a constant pH and neutralize acidic pollution from rainfall or waste water. Primary alkaline buffer capacities consist of various bases including bicarbonate, carbonate, and occasionally hydroxide, borates, silicates, phosphates, ammonium, sulfides, and organic ligands.

Streams lows in alkalinity are susceptible to changes in pH while those higher in alkalinity resist these changes. When the pH reaches levels of 6.0, a rapid drop in the pH level ensues as the bicarbonate buffering capacity is consumed. At a pH of 5.5 there is a very low buffering capacity and at 4.0 there are no bicarbonate or carbonate ions left and thus there is no alkalinity.

Alkalinity may be affected by soil and rock that contains carbonate material, sewage outflow, and waste water from houses. Each of these effectors increases alkalinity levels in stream water.

Alkalinity levels between 20 and 200 mg/L are typical of freshwater streams, levels between 100

and 200 mg/L will stabilize the pH of a stream. Alkalinity levels below 10 mg/L indicate a poorly buffered stream.

Acidity

Acidity is a measure of the strong and weak acids that react with hydroxyl ions that indicates the water degree of corrosiveness. Acidity is measured through titration in units of mg CaCO₃/L. The most likely cause of high acidity in water is Carbon Dioxide. However, increase acidity levels may also be caused by sulfuric and hydrochloric acid.

Other causes of increased acidity levels are improper disposals of sewage and industrial waste, mine drainage and acid rain. High acidity in water utilized by humans is likely to be corrosive to copper pipes and may cause problems and leaks in plumbing. Acidic water is also a health hazard when consumed.

References:

Delaware Riverkeeper Network's Volunteer Monitoring Program. 2004. Delaware Riverkeeper. 14 July 2004
<http://www.delewareriverkeeper.org/monitoring/monitoring.htm>.

Murphy, Sheila. BASIN Water Quality Information. 15 June 2002. BASIN – City of Boulder – USGS Water Quality Monitoring. 14 July 2004
<http://bcn.boulder.co.us/basin/data/nutrients/info.alk.html>.

WOW. 2003. Water on the Web – Monitoring Minnesota Lakes on the Internet and Training Water Science Technicians for the Future – A National On-Line Curriculum using Advanced Technologies and Real-Time Data.
(<http://wow.nrri.umn.edu>) University of Minnesota – Duluth, Duluth MN 55813

Water Chemistry Standards

**WATER CHEMISTRY STANDARDS
FOR HIGH QUALITY STREAMS**

Note:

1. Roughly based on Chapter 91, Title 25 of PA Code
2. To be used in concert with Itach CCL899 Advanced Portable Laboratory

Acidity (as CaCO₃)

Alkalinity (as CaCO₃)

Natural ranges from 20 - 200 mg/L
Tend to be higher in calcium carbonate geology (limestone areas)
Below 20 mg/L should raise concern
230 mg/L at Donegal Creek, Lancaster Co. was not uncommon
75 mg/L = Limestone stream

Ammonia (as N)

0.002 mg/L	general limit
0.06 mg/L	fish gill damage
0.1 mg/L	indicates polluted water
0.2 mg/L	trout and salmon begin to die
2.0 mg/L	kills carp

Danger to aquatic life depends on temperature, pH and length of exposure along with dissolved oxygen and carbon dioxide levels. The higher the pH and the warmer the temperature, the more toxic the ammonia. Also, ammonia is much more toxic to fish and aquatic life when water contains very little dissolved oxygen and carbon dioxide.

Carbon Dioxide

1.0 - 6.0 mg/L	fish tend to avoid the water
12 mg/L	few freshwater fish can survive in water at this concentration
30 mg/L	kills the most sensitive fish immediately
45 mg/L	maximum limit for trout
50+ mg/L	trout eggs won't hatch

Chloride

150 mg/L	maximum level for fish and aquatic life
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Chlorine, free and total

0.3 ppm	detectable odor
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Free chlorine test measures only the amount of free or dissolved chlorine in water
Total chlorine test measures both free and combined forms of chlorine
Free chlorine is toxic to fish in very small amounts (as compared to total chlorine)
Free chlorine rapidly reacts with other substances thus creating a combine formed typically less harmful to fish.
If water contains a lot of decaying materials and free chlorine is introduced, compounds called trihalomethanes or THMs can form. Some THMs in high concentrations are carcinogenic to people. THMs are persistent once formed and can pose a long-term health threat.

0.006 mg/L of total chlorine	kills trout fry in two days
0.01 mg/L of total chlorine	recommended max. for all fish and aquatic life
0.01 mg/L of total chlorine	starts to kill Chinook and Coho Salmon
0.02 mg/L of total chlorine max.	Brook and Brown Trout can withstand
0.05 mg/L of total chlorine max.	amount tolerated by young Pacific Salmon in the ocean
0.25 mg/L of total chlorine	only the hardiest fish can survive
0.37 mg/L of total chlorine max.	remaining hardy fish can withstand
1.0 mg/L of total chlorine	kills oysters

Chlorine Dioxide

Color

50 units	warm water fishery (example - Codorus Creek, York Co.)
75 units	max. for warm water fishery

Conductivity/TDS/Salinity

Inorganic compounds are good conductors (salts, acids, ammonia)
Organic compounds are bad conductors (sugar, oil, alcohol, organic matter)
Distilled water is a poor conductor because of the lack of ions
Conductivity usually somewhere between 50 - 1500 µmho
TDS for unpolluted lakes range from 17 - 30 mg/L
TDS for polluted lakes = 400 mg/L
TDS for drinking water should be < 750 mg/L

mho	unit of conductivity for water
µmho	micromho - unit for low conductivity waters
0.64 µmho/cm = 640 mho/cm = 640 µS/m = 64.0 cS/m = 6.4 mS/m	
TDS (mg/L) = 640 mho/cm = 0.64 µmho/cm	

Copper (Cu)

6.5 ug/L @ water hardness < 100 mg/L
0.0065 mg/L
12 ug/L @ water hardness 100 - 199 mg/L
0.012 mg/L
21 ug/L @ water hardness > 200 mg/L
0.021 mg/L

Hardness (as CaCO₃)

< 75 mg/L	soft water
75 - 150 mg/L	moderately hard
150 - 300 mg/L	hard
> 300 mg/L	very hard

Limestone streams from 300 - 350 ppm not uncommon

Iron, total (Fe)

1500 ug/L	not to be exceeded
1.5 mg/L	

Manganese (Mn)

1000 ug/L	not to be exceeded in drinking water (color problem)
1.0 mg/L	

Nitrate (NO₃-N)
 10.0 mg/L human consumption limit
 90.0 mg/L warmwater fishery limit
 > 20 mg/L health hazard to juvenile mammals

Nitrite (NO₂-)
 1.0 mg/L human consumption limit
 5.0 mg/L warmwater fishery limit
 Rarely measurable in unpolluted natural waters

Oxygen, dissolved (DO)
 3 - 4 mg/L stressful to aquatic life
 6 mg/L fair
 8 mg/L good
 > 7 mg/L no problem for river/lake bass
 > 8 mg/L no problem for river/lake trout

Species of fish	Lowest DO level in mg/L at which fish survive for 24 hours (summer)	Lowest DO level in mg/L at which fish survive for 48 hours (winter)
Northern Pike	6.0	3.1
Black Bass	5.5	4.7
Sunfish (general)	4.2	1.4
Yellow Perch	4.2	4.7
Black Bullhead	3.3	1.1

pH
 7.0 neutral
 8.5 biological productivity (CO₂)
 5.5 damaging, indicates acid rain, AMD problems
 6.0 - 9.0 ok for aquatic life

Limiting pH values		Effects of some scientific studies
Minimum	Maximum	
3.8	10.0	Fish eggs could be hatched, but deformed young were often produced.
4.0	10.1	Limits for the most resistant fish species
4.1	9.5	Range tolerated by trout
4.3	---	Carp died in five days
4.5	9.0	Trout eggs and larvae develop normally
4.6	9.3	Limits for perch
5.0	---	Limit for Sockeyeback
5.0	9.0	Tolerable range for most fish
---	8.7	Upper limit for good fishing waters
5.4	11.4	Fish avoided waters beyond these limits
6.0	7.2	Optimum range for fish eggs
1.0	---	Mosquito larvae were destroyed
3.3	4.7	Mosquito larvae will live within this range
7.5	8.4	Best range for algae growth

Phosphate
 0.1 mg/L recommended max. for rivers and streams
 0.025 mg/L accelerates the eutrophication process in lakes
 0.01 - 0.03 mg/L amount of phosphate-phosphorus in most uncontaminated lakes

Silica (Si)
 1 - 10 mg/L natural waters
 > 60 mg/L rarely exceeded in natural setting

Sulfate (SO₄)
 5 - 50 mg/L natural waters
 250 mg/L should not be exceeded (drinking limit as well)

Sulfide

Suspended Solids (TSS)
Lakes
 < 25 mg/L clear water
 25 - 100 mg/L intermediate
 > 100 mg/L muddy
Rivers
 < 25 mg/L high level of protection
 80 mg/L moderate level of protection
 400 mg/L low level protection
 > 400 mg/L very low protection
 75 - 100 mg/L harmful to fish eggs

25 - 80 mg/L good fisheries
 Walleye are sensitive to TSS w/ death rates @ < 200 mg/L (reduced sight affects feeding ability)

Temperature

Time Period	Coldwater Fishery (°F)	Warmwater Fishery (°F)
January 1 - 31	38	40
February 1 - 29	38	40
March 1 - 31	42	46
April 1 - 15	48	52
April 16 - 30	52	58
May 1 - 15	54	64
May 16 - 31	58	72
June 1 - 15	60	80
June 16 - 30	64	84
July 1 - 31	66	87
August 1 - 31	66	87
September 1 - 15	64	84
September 16 - 30	60	78

October 1 - 15	54	72
October 16 - 31	50	66
November 1 - 15	46	58
November 16 - 30	42	50
December 1 - 31	40	42

Species	Upper limit (°C)	Optimum (°C)
Spotted Bass		
adult	34	24
spawning	n/a	18 - 21
Smallmouth Bass		
adult	32	21 - 27
spawning	n/a	12.8 - 21
Largemouth Bass		
adult	36	24 - 30
spawning	30	21
Brown Trout		
adult	27	12 - 19
juvenile	27	7 - 19
spawning	27	2 - 13
Rainbow Trout		
adult	25	12 - 18
spawning	n/a	10 - 15.5
Brook Trout		
adult	24	11 - 16
spawning	n/a	4.5 - 10
Lake Trout		
adult	23.5	4 - 18
spawning	n/a	4.5 - 14

Turbidity

< 100 NTU	limit for rivers
< 25 NTU	10 day average for aquatic life - no problem
< 50 NTU	starting to raise concern
5 NTU	max. for swimming

Zinc

180 ug/L @ water hardness 50 mg/L
0.18 mg/L
320 ug/L @ water hardness 150 mg/L
0.32 mg/L
570 ug/L @ water hardness 200 mg/L
0.57 mg/L