A Water Quality Assessment Plan to Determine Biological Health and as a Baseline for Future Studies and Monitoring on the West Branch Susquehanna River between Anderson Creek and the 879 Bridge east of Clearfield.

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ABSTRACT: Coal mining in Clearfield County prior to 1977 unearthed harmful chemicals which are entering into the West Branch Susquehanna watershed. Federal laws passed in 1977 have prevented further damage to the streams, but billions of dollars of damage have already occurred (Fayette County Website 2005). The chemical pollutants in the streams kill aquatic organisms and in many cases stain the water and streambed with ugly, unnatural colors.

Old Town Sportsman's Association and Pennsylvania Fish and Boat Commission have been stocking trout in this stretch of WBS for many years. Though information is limited regarding the capacity of WBS to support trout, stocked trout are known to survive for months or years. Some of the tributaries leading into WBS are known to have wild brook trout populations.

OTSA teamed up with Trout Unlimited to design and implement a stream health assessment plan to measure the ecological health of this section of WBS. The assessment will measure chemical loadings entering the stream, and will scientifically analyze the benthic macroinvertebrate populations. Initially, the assessment will be an inventory of the stream's health at this time, but will become a useful tool for identifying trends over future years.

INTRODUCTION: Acid mine drainage (AMD) is a major problem throughout Appalachia's coal mining region, and is the most extensive water pollution problem in Pennsylvania. Of the 13,000 miles of AMD-polluted streams in Appalachia (Todd 1997), 3,000 miles of affected streams are located in PA. These degraded streams in Pennsylvania result in an estimated loss to the state economy of $67 million per year in tourism and sport fishing (USGS 2006). Today Clearfield is the worst county for AMD in the state, with 617 problem mine areas identified by the PADEP's Bureau of Abandoned Mine Reclamations Inventory under the Abandoned Mine Lands program (Fayette County Website 2005).

AMD pollution is mostly the result of various coal mining techniques prior to the 1970s (Fayette County Website 2005). Coal mining is now regulated under strict federal and state laws. Under these laws, mining cannot begin if it is determined that it will harm the environment (Enviro Sci Inquiry 2004). Furthermore, water discharges now must be monitored during and after active mining to ensure that the pH, alkalinity, acidity, and content of dissolved aluminum, iron, and manganese fall within regulation guidelines.

Other elements have maximum allowable concentration limits as well (Science Direct 2007), but these elements have insignificant impact on aquatic life.

During coal mining, ground disturbance often progresses below the water table. Coal seams are usually located beneath layers of iron pyrite, which contains sulfur, iron, and other minerals. In the process of removing the coal, the iron pyrite is disturbed and allowed to contact water and oxygen. This exposure results in a four-step process of chemical reactions involving oxygen, water, and sulfur (Todd and Redick 1997). These reactions produce chemicals that contaminate the water directly, and by carrying unnaturally high concentrations of metals. The resulting AMD may go directly from the mine into groundwater, or may enter nearby streams.

The resulting chemicals will have an adverse effect on stream life. AMD affects the ecological productivity of plant, animal, and microbial life in impacted streams in several ways (Klemow 2007). AMD chemical reactions cause a variety of effects. Because of the complexity of these effects and their interrelation, it is often difficult to isolate an individual factor. Toxicity depends upon various factors including discharge volume, pH, total acidity, and concentration of dissolved metals. The effect of AMD on streams depends on its flow, pH, and ability to buffer through alkalinity. Streams' buffering capacity is indicated through the presence of bicarbonate or carbonate ions in the receiving stream. The stream's buffering capacity increases proportionally to increasing amounts of these alkaline materials in the water (Earle and Callaghan 1998).

pH is the most important factor in overall effects on a stream by AMD. Low pH has direct effects on organisms (Earle and Callaghan 1998) and indirect effects by influencing all other aspects of AMD. The direct effects include respiratory difficulties in aquatic animals due to increased permeability of gills (Earle and Callaghan 1998, SRHCES 2007) and impairment of the normal functioning of aquatic animals due to disruption of cellular metabolism (Klemow 2007). Cellular interference occurs when low pH unbalances the ratio of sodium and chloride ions in blood, and lowers oxygen levels in tissues. Under these circumstances sodium ions are expelled and hydrogen ions are absorbed, disrupting osmoregulatory functions and usually resulting in impairment or death of organisms (Earle and Callaghan 1998).

Several other direct effects of low pH also harm stream ecosystems. Acidic water hinders microbial decomposition of leaves by carrying unnaturally high quantities of dissolved metals. Acidity, along with dissolved metals such as iron and aluminum, decrease the population of these microscopic decomposers. With fewer decomposers available to break down detritus, nutrient cycling and retention is disrupted (Earle and Callaghan 1998, McTammany et al. 2007). Studies by McTammany et al. (2007) have revealed that AMD affected streams have a leaf decomposition rate two to three times slower than unaffected streams. With high acidity and low concentration of nutrients, the growth of algae is hindered in affected streams. Algae are the base of the food chain in most streams, and the main food for many macroinvertabrates. Besides unnaturally low populations of algae, benthic macroinvertabrate populations are also severely limited by the high concentrations of metals that are dissolved in the water as a result of low pH (Earle and Callaghan 1998, McTammany et al. 2007). This disappearance of biota at lower levels removes the main food source for fish (EPD 2008).

As mentioned above, low pH indirectly influences streams' ecosystems. The acidic water leaches harmful metals out of the surrounding clay and bedrock as it passes out of the mine and through the stream. These metals are deadly to a stream ecosystem, each affecting natural processes in different ways. The worst metals draining out of the bituminous coal fields of western Pennsylvania include iron and aluminum. These metals fall out of their water solution in the form of precipitates, creating a whole host of new problems for the stream (AMRC 2007). Both aluminum and iron decrease the amount of dissolved oxygen in water as they precipitate in the form of a hydroxide. This loss of oxygen is detrimental to stream animals (Earle and Callaghan 1998).

Aluminum is the most harmful metal found dissolved in high concentrations associated with acid mine drainage (AMRC 2007). At a pH between 5.7 and 6.2, aluminum precipitates out of solution. Aluminum hydroxide accumulates on the stream bottom and rocks. It appears as a whitish coating, usually found in areas where the pH of acidic water with large amounts of dissolved aluminum is increased by contact with water from a tributary with a higher pH. The coating destroys stream life, smothers eggs, and kills fish and macroinvertebrate by accumulating on their gills, thereby suffocating them (AMRC 2007). This coating prevents the growth of algae and makes a poor surface for macroinvertebrates to live on. A combination of dissolved aluminum in greater amounts than .5 mg/L and a pH below 5.5 generally results in loss of all fish and most macroinvertebrates. Aluminum is most toxic to fish in a pH between 5.2 and 5.4 (Earle and Callaghan 1998).

Another harmful metal dissolved by acidic water is iron. Iron begins to precipitate out of solution at a pH of 3.5, reacting with water and oxygen to form iron hydroxide, or "yellow boy" (Earle and Callaghan 1998). Yellow boy forms a yellow-orange coating on the stream bottom creating problems for all aquatic life. This coating prevents the growth of algae necessary as the base of a stream's food chain. It also directly affects benthic macroinvertabrate by preventing them from living on rocks, and by clogging their gills. Yellow boy increases water turbidity, blocking light required for photosynthesis of aquatic plants and lowering visibility for fish and other aquatic animals (Wheeling Jesuit University 2004).

Like aluminum, a high concentration of iron in conjunction with low pH kills aquatic life in streams. Iron is somewhat less deadly than aluminum, for more alkaline waters with iron contamination will support some life.

Dissolved manganese is often present in waters contaminated with AMD discharge. Manganese is difficult to remove from streams because it doesn't precipitate unless the water reaches a pH of 10, which is rare. Because of the high pH required for its precipitation and the fact that manganese usually occurs with other AMD metals, it is difficult to isolate and study its effects on aquatic organisms. Little is known about the effects of high manganese concentration levels on organisms, but research has determined that low levels of water hardness will buffer its deleterious effects.

Manganese forms a black hydroxide coating on the streambed during its rare occurrences of precipitation. Its effects may be concealed by the more harmful effects of aluminum, acidity, and iron. Until more is learned about its impact on stream ecosystems, issues surrounding dissolved manganese levels are mostly confined to drinking water discoloration (Earle, J., T. Callaghan. 1998).

The stretch of the West Branch Susquehanna located between Curwensville and Clearfield has had limited monitoring to gage its potential to support aquatic life. Many watershed organizations are undertaking projects to improve quality of the small streams that contribute to the West Branch Susquehanna. It has become necessary to develop a plan to determine the water quality at the present time and that can be used to measure changes of water quality in the future.

The main factor affecting the health of West Branch Susquehanna at this time is acid mine drainage that was brought about by years of coal mining in Clearfield County. Several watershed organizations are working to improve the water quality of tributaries leading into WBS.

Another factor that will impact West Branch Susquehanna is the operation of the upcoming ethanol plant in Clearfield, PA. Construction of the Bionol Clearfield LLC Ethanol Plant is expected to be completed early in 2010 and operating shortly thereafter. The plant will produce 108 million gallons of ethanol per year and use approximately 378 million gallons of water taken from the WBS (Courier Express 2008). Data collection as outlined according to this assessment plan will begin before the ethanol plant starts operations, and may reveal geomorphological and hydrological changes that result in changes to the make up of the aquatic biological community in WBS due to the water loss (USGS 1994).

Various types of data are necessary to determine the health of streams. Of these, the most important gages include water chemistry and biological data. The four important chemical parameters used to test the water composition include water pH, and concentration of aluminum, iron, and manganese.

In the case of AMD remediation, biomonitoring generally analyzes benthic macroinvertebrate assemblages. Macroinvertebrates are better than fish for this type of assessment for various reasons. Macroinvertebrates are less mobile than fish; they can't migrate to avoid polluted discharges (NYSDEC 2008); therefore, they are constantly exposed to chemicals. Species of benthic macroinvertebrate have a distinct range of sensitivity to pollutants; therefore, they indicate various degrees of environmental degradation (USGS 1994).

Of the highest value to stream condition researchers are macroinvertebrates from the "EPT taxa". EPT taxa refer to species belonging to the taxonomic orders of *Ephemeroptera* (mayflies), *Plecoptera* (stoneflies), and *Trichoptera* (caddisflies). Members of the EPT taxa are the most pollution-sensitive benthic macroinvertebrates found in streams in this region, with *Ephemeroptera* being the single most sensitive. The presence of members of EPT taxa indicates a stream with high quality water. Streams with poor quality water usually won't have EPT taxa, but instead will be dominated by pollution-tolerant orders of organisms, such as *Tubificdae* (earthworms), *Chironomidae* (midge larvae), *Sialis* (alderfly larvae), *Nigronia* (fishfly larvae), and non-benthic insects, such as *Dytiscidae* (predaceous diving beetles) (Earle and Callaghan 1998).

METHODS: We identified 13 locations for chemical sampling, benthic sampling, or both at various sites along the WBS between Anderson Creek and the Rt. 879 Bridge east of Clearfield. Two locations were chosen to be tested for both benthic and chemical sampling, a site just upstream from Anderson Creek and a site just downstream of the Moose Run tributary. These are the farthest upstream and the farthest downstream of our chosen sampling locations. In future sampling we can compare the chemical content of the two locations to better analyze the chemical load and acidity contributions of the five tributaries between them. The other chemical sampling sites include one location in each of the five tributaries within the 10.5 mile sampling area. The tributaries we will sample include Anderson Creek, Hogback Run, Hartshorne Run, Montgomery Run, and Moose Creek. Testing the tributaries themselves will help us identify the worst AMD contributors to the WBS. This assessment will be performed in April and May each year. After May most of the various larva that we collect will metamorphisize into their flying or terrestrial phase and leave the stream.

We will take chemistry samples at the same time we take benthics samples. At each location, we will fill two sampling containers with water that we will have tested at a lab. For the first sample, we will fill a two fluid ounce vial with water. The two ounce vial is tested for dissolved metals, including aluminum, iron, and manganese. After filling, we will add five drops of diluted nitric acid to each two-ounce sample. The diluted nitric acid is added to ensure that the dissolved metals stay in solution for testing. Next, we will fill a 500 ml bottle with water from the same testing site. The 500 ml bottle is tested for pH. We will place the water samples in a cooler with ice immediately after removal from the stream. The cool temperatures prolongs the storage time to 24 hours. After we complete the sampling, we will deliver the samples to Mahaffey Laboratory, LTD, in Curwensville, PA.

During chemical sampling, we will measure stream flow rate. Using a flow meter, we will determine the amount of water flowing through the stream, measured in cubic feet per second. This information will be used later in conjunction with the lab reports on metal concentrations to calculate chemical loading of the stream. Chemical loading is a calculation of pounds of metal per day contributed by tributaries. We will use this information to determine which tributaries are the biggest contributors of AMD to WBS.

Our assessment plan includes eight benthic macroinvertebrate sampling locations along various points of WBS. The two benthic sites already listed above are located at the most upstream, and at the most downstream sampling points of our assessment plan. We can compare the point farthest downstream to the most upstream point to determine the effects of the tributaries on aquatic life in WBS. We chose the other six benthic sampling locations based on their location downstream of tributaries. Benthic sampling results at each of these locations along with chemical loading data will allow us to measure the effect of each tributary on the aquatic life in WBS. Biological monitoring greatly enhances assessment when combined with chemical monitoring; aquatic organisms are indicators of overall, integrated water quality. Their presence can indicate the combined effects of chemicals that are below detectable limits (NYSDEC 2008).

We will collect benthic samples and analyze them according to the PA DEP's "Index of Biological Integrity for Wadeable, Freestone Streams in Pennsylvania" at each of the eight benthic sampling locations. This biological index is designed to measure the extent to which anthropogenic stressors impair the capability of a stream to support a healthy aquatic community.

In keeping with the Index of Biological Integrity (IBI), we will use a D-Frame net with 500 micron mesh, 95% ethanol as a sample preservative, two 18" x 12" x 3.5" pans marked with 28 four-square inch grids, and a four-square inch circular "cookie cutter".

At each sample location, we will perform six "kicks" within a 100-meter stream reach. For each kick, we will disturb approximately one square meter immediately upstream of the net for approximately one minute to an approximate depth of 10 cm. We will place all specimens that we collect from a location in one bottle, and label them with the sampling location number. We will fill the bottle with ethanol to preserve the specimens. Later we will place each sample in a grided pan for analysis. We will randomly select four grids from the sample pan and remove debris from them using the cookie cutter. We will place the debris in an identical empty pan, and pick organisms from randomly selected grids until we have an approximately 200-organism sub-sample. We will identify and count the organisms in the sub-sample. The Biological Index uses six metrics to calculate the IBI. After we count and identify the samples, we will use several calculations to measure stream health based on our samples.

We will use the Modified Beck's Index. This metric is designed to indicate the loss of pollution sensitive species. The score for each sample will decrease in value with increasing levels of pollution. We will assign a pollution tolerance value to each taxon within our subsample, based on their resistance to pollution. Pollution tolerance values can be 0, 1, or 2. We will count the number of individuals within the sub-sample that have pollution tolerance values of 0, 1, and 2, and we will multiply the number of individuals by 3, 2, and 1, respectively. We will find the sum of these products. The formula is shown below.

Modified Beck's Index = (3 x (number of taxa with pollution tolerance value of 0)) +
(2 x (number of taxa with pollution tolerance value of 1)) +
(1 x (number of taxa with pollution tolerance value of 2))

Next we will use the Ephemeroptera Plecoptera Trichoptera (EPT) Taxa Richness metric. This is a community structure metric and it counts the number of taxa belonging to the highly pollution-sensitive orders listed above. This value will decrease as pollution levels increase. We will count the number of individuals in each of the EPT orders and add them to find this metric. See the example below.

EPT Taxa Richness = number of taxa belonging to the insect orders Ephemeroptera, Plecoptera, or Trichoptera

The next calculation we will perform is the Total Taxa Richness metric. This is a count of total number of taxa in a sub-sample. This number decreases in value with an increase in pollution. Pollution intolerant species will disappear and the stream will become dominated by pollution-tolerant taxa. To find this value, we will simply count the number of taxa in our sub-sample.

We will measure taxonomic richness and evenness of our sub-samples by using the Shannon Diversity Index metric. This formula will indicate increasing dominance of pollution-tolerant taxa, and loss of pollution-sensitive taxa at increasing pollution levels. The value will decrease with increased pollution. See the formula below.

$$\text{Shannon Diversity Index} = -\Sigma_i \,(ni / N) \ln (ni / N)$$

where,
ni = the number of individuals in each taxa (relative abundance)
N = the total number of individuals
Rich = the total taxa richness

Then we will use the Hilsenhoff Biotic Index to calculate the average pollution tolerance value. The formula is weighted by the number of individuals of each taxon in the sub-sample. This index increases with increasing pollution. The formula is listed below.

$$\text{Hilsenhoff Biotic Index} = \sum_{i=0}^{10} [(i * \text{nindvPTVi})] / N$$

where,
nindvPTVi = the number of individuals with pollution tolerance values of i
N = the total number of individuals

Finally, we will use the Percent Intolerant Individuals metric to determine the percent of individuals with pollution tolerance values of five or less for each sub-sample. This number will decrease with increasing pollution. The formula is shown below.

$$\text{Percent Intolerant Individuals} = \left(\sum_{i=0}^{5} \text{nindvPTVi}\right) / N * 100$$

where,
nindvPTVi = the number of individuals with pollution tolerance values of i
N = the total number of individuals

After calculating the values for these six metrics, we will integrate the values to find the IBI score. To do this, we will standardize all the scores to a maximum value of 1.00. The five metrics that decrease in value with increasing pollution are standardized to the 95th percentile from the 451 samples in the IBI development dataset. The Hilsenhoff Biotic Index is standardized to the 5th percentile from the IBI development dataset. After standardization, we will average the six scores and multiply by 100. This will produce an index score between 0 and 100. Lower numbers indicate greater deviation from the expected reference condition. Table 1 shows an example of the standardization and averaging of the six IBI metrics (DEP 1999).

Metric	Standardization Equation	Observed Metric Value	Standardized Metric Score	Adjusted Standardized Metric Score Max = 1.000
Modified Beck's Index	observed value / 39	41	1.051	1.000
EPT Taxa Richness	observed value / 23	20	0.870	0.870
Total TaxaRichness	observed value / 35	28	0.800	0.800
Shannon Diversity Index	observed value / 2.90	2.69	0.928	0.928
Hilsenhoff Biotic Index	(10 – observed value) / (10 – 1.78)	1.39	1.047	1.000
Percent Intolerant Individuals	observed value / 92.5	92.5	1.000	1.000
Average of adjusted standardized core metric scores * 100 = **IBI Score =**				93.3

Table 1= Example of metric standardization equations and index calculation (DEP 1999)

DISCUSSION: This assessment plan is designed to be used from now into the future to monitor the health of WBS. The information gathered in this assessment may be used in identification of tributary treatment priorities, acquisition of grant money for remediation projects, long-term monitoring of the health of West Branch Susquehanna to track changes as watershed groups and government agencies work to eliminate the harmful effects of AMD. Furthermore, publication of trends may lead to increased public interest and support for AMD remediation projects on WBS.

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