Chapter 8

Macroinvertebrates

A History of Biomonitoring

Today many state agencies, environmental consultants, professors, and volunteer groups (Project SEARCH included) utilize benthic macroinvertebrates in an attempt to measure environmental conditions for a specific area. A standard technique used to evaluate environmental conditions is to collect a sample of organisms, identify the species present, and make inferences based upon the composition of the sample. A good aquatic ecologist should be able to provide information about the dominant energy source, potential human influence, and water quality after only a few minutes at a stream site. These conclusions are based on the tremendous amount of scientific studies both large and small which help us to understand the ecology of flowing waters. Today there are more than 1,000 publications concerning aquatic entomology annually. Although many of these may seem trivial (*i.e.* Rhythmic thermoregulation in larval crane fly), each helps to focus our view into the ecology of running waters. Although presently, there may seem to be a exceptional amount of research focusing on aquatic entomology, this was not case prior to the 1970's. The following is a brief description of the major events leading to the development of biomonitoring.

To get to the present use of macroinvertebrates in biological monitoring studies, aquatic entomology has gone through several phases, natural history, recreational, and scientific. Each phase is defined by the majority work being produced. The earliest work concerning aquatic entomology can be dated back to Aristotle. Like all other biological work at this time, publications were based on observation. Newly discovered organisms were observed, described, and documented. The focus was on the natural histories (the who, what, where, and when) of the organisms. A good example is *De Natura Animalium* published by Claudius Aelianus around 235 A.D. In this, he described strange organisms swarming over the River Astraeus. He further explains that a crude mimic of these organisms (later described as caddisflies) made of wool and hackles could be used to capture fish. Remnants of the natural history phase extend to the present day. Each time a new or remote area of the earth is discovered, many new species have to be observed, described, and documented just as was done in the third century.

The second phase started around 1496 with the publication of *A Treatyse of Fysshyne Wyth an Angle* by Dame Juliana Berners. Her publication was the first of what was to be a long series of works focusing on using manufactured flies to capture fish. Her book included instructions on how to build flies (specific colors and materials) and the technique used to fish with them. The most famous work of this phase is *The Compleat Angler* (1836) by Izaak Walton. This was a series of publications spanning many years. His publication was extremely comprehensive, undoubtedly spawning interest in the field of fly fishing. Many publications followed Walton all of which focused on the recreational experience called fly fishing. Several notable text include: *The Fly Fisher's Entomology* (1836) by Alfred Roland, *Floating Flies and How to Fish Them* (1886) by Theodore Gordon, *American Trout Stream Insects* (1920) by Louis Rhead, *Matching the Hatch* (1955) by Ernest Schwiebert, and *Aquatic Entomology* (1981) by Patrick McCafferty. Today there are hundreds of publications produced concerning fly fishing. It has become a major recreational activity especially in Connecticut. More importantly, fly fishing inadvertently caused people to focus on the benthic community in great detail. Fishing success (and bragging rights) was greatly increased by knowing the time and conditions which cause a particular type of mayfly hatch. This type of observations led to the scientific phase.

The scientific field of aquatic entomology is the newest phase. There was limited interest in studying aquatic insects until the Clean Water Act, passed in 1972, grabbed our attention. Prior to the act, only a few studies were being conducted. In 1908 a pair of Germans noticed different communities up and downstream of an industrial area. Their observation led to the development of the first method to assess water quality using macroinvertebrates, the *Saprobien System*. The other major work was the *Ecology of Aquatic Insects* (Macan 1962). This work was comprehensive covering all of the known information to date.

After the Clean Water Act, many state agencies began to develop methods to evaluate water quality. Several indices were the Hilsenhoff Biotic Index (HBI) and the Sequential Comparison Index (SCI). Additionally, many biologists developed mini-indices which would evaluate a particular aspect of the aquatic ecosystem. In 1989, the United States Environmental Protection Agency (USEPA) published a standardized method to assess water quality. Their method called, Rapid Bioassessment Protocols, includes a set of mini-indices or metrics and a comparison system. The Connecticut Department of Environmental Protection (CT DEP) has been evaluating water quality in this fashion for many years.

Future entomologists need to be able to interpret and understand the tremendous volume of work completed annually. The field is growing at a phenomenal rate with portions in each of the 3 phases. New species are being discovered daily, millions of angler hours are spent attempting to match the hatch, and all of the New England states are sharing data to develop regional biocriteria for water quality assessment. Despite the ever expanding volume of work, the fundamentals of biomonitoring still rely on field observation and how organisms relate to their environment, just as the early natural historians did in the third century.

What is bioassessment?

Bioassessment or biological assessment is defined by the United States Environmental Protection Agency (USEPA) as "an evaluation of the biological condition of a waterbody using biological surveys and other direct measurements of resident biota in surface waters" (Klemm *et al.* 1990). Due to the complexity of aquatic ecosystems, biological evaluations often involve examining only one organizational level (individual organisms, functional group, population, *etc*.) within a specific biological community (algae, fish, or macroinvertebrates). The results of these "small scale studies" are then extrapolated to the aquatic ecosystem as a whole. For example, the Hilsenhoff Biotic Index (HBI) is a weighted mean of the pollution tolerance values resulting from a collection of the macroinvertebrate population. This index score is then used to conclude water quality for the stream as a whole, regardless of chemical, fish or algal population data.

Biological monitoring or biomonitoring is the process of compiling data from many biological assessments over an extended period of time. Biomonitoring studies can help identify trends in water quality by indicating changes within the structure of the aquatic community. In most cases a biomonitoring program is established to assess and monitor the level of human influence on a particular stream system.

Chemical monitoring is valuable to determine the "normal range" of both organic and inorganic components which influence the biological communities. However, due to the continual uni-directional flow in rivers, the concentration of various substances in solution can fluctuate dramatically over a very short period of time. When attempting to determine the "actual" chemical water quality of a stream or river a large number of samples gathered with great frequency is necessary (Stevens *et al* 1994). The water sampling design of a monitoring program may call for grab samples taken bi-weekly, weekly, monthly, quarterly, or annually. However, studies have shown many chemical concentrations increase immediately following and during precipitation events. Therefore a limited sampling regime may miss isolated point and nonpoint source inputs linked to precipitation events.

Biological assessments are more efficient at quickly measuring overall stream health because the organisms being evaluated must continually remain in the water. Each organism acts as a mini round-the-clock water quality monitoring device, mirroring the combined effects of all pollution types (physical, chemical, biological, point and non-point) over an extended period of time (Figure 8.1).

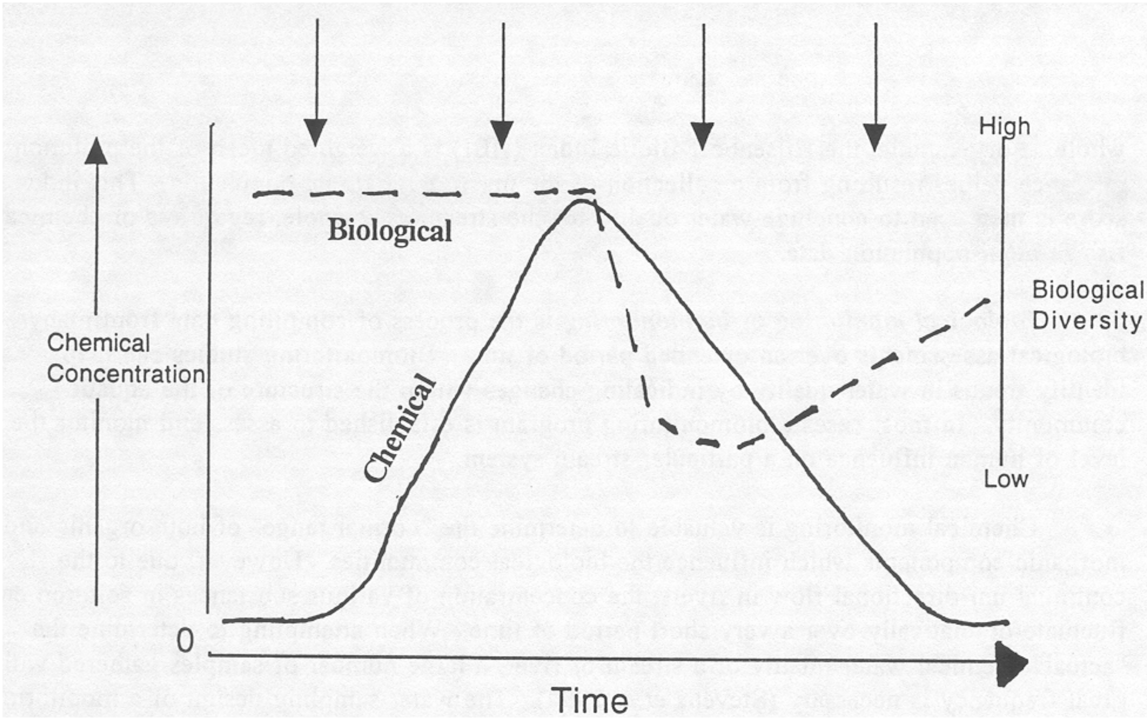

Figure 8.1. An example of how the discharge of a harmful pollutant moves through a riffle system and how chemical results and biological results can differ. Periodic grab sampling is indicated by the arrows at the top of the figure. Chemical concentration is shown by the solid line and biological diversity by the dashed line. A large number of samples are necessary to detect a chemical impact while fewer biological samples are needed.

Additional benefits of biomonitoring are it is relatively inexpensive, easy to do, and the results are meaningful to the public (Plafkin *et al.* 1989). Several communities uses as biological indicators are fish (Karr 1981), algae (Lange-Bertalot 1979), and macroinvertebrates (Bode 1991). Each community has advantages and disadvantages when attempting to assess water quality. Project SEARCH focuses on benthic macroinvertebrates because of several key advantages. The following are modified from the Maine Biological Monitoring and Biocritera Development Program (Davies *et al*. 1993).

1. Macroinvertebrates are generally limited in mobility and, therefore, less able to avoid the effects of pollutants. Sampling fish communities may provide unreliable results of localized influences because they are able to swim away from any isolated pollution.

2. Pollution tolerance levels for macroinvertebrates ranges from very low (affected by a very small amount of pollutant) to very high (actually thriving in large numbers in specific types of pollution). A single sample can provide a wealth of information.

3. The macroinvertebrate community is more diverse and encompasses a greater number of feeding strategies than fish communities.

4. Benthic macroinvertebrates have longer life cycles than bacteria or algae and therefore provide a better measure of long term ecological stability.

5. Macroinvertebrates can be found in almost any type of aquatic habitat, providing it has not been severely polluted. Fish, on the other hand, may be restricted by water depth, volume, or physical barriers.

6. Methods for sample collection, preservation, subsampling, and data analysis are well established and documented.

7. Macroinvertebrates can be captured by a single individual with relative ease and inexpensive equipment.

8. Macroinvertebrate populations rapidly recover from repeated sampling which is necessary to pinpoint a pollution source or event.

Once a target community has been selected, a sampling design must be developed. A ***sampling design*** is a plan which outlines the key components of who, what, where, when, and how concerning the data to be collected. The sampling design is the foundation of any scientific study. In order to develop an appropriate sampling design using living organisms to measure environmental health, a working knowledge of their ecology is crucial. Consider the following hypothetical example. A scientist wishes to assess the condition of the state forests in Connecticut using a forest health index based on redwood trees as an indicator of excellent forest health. After sampling every forest in the state, not a single redwood tree was found. The results of the study indicate the condition of the forests in Connecticut is dismal. Hopefully these results are not true. If the researcher had some basic knowledge of redwood tree ecology, the researcher would realize that this species is not native to Connecticut and therefore is an inappropriate environmental indicator for this study.

The same holds true for macroinvertebrates. A basic working knowledge of aquatic insect ecology is required in order to accurately design a biomonitoring study. Four key variables which must be considered in the sampling design are the habitat, the time of year (seasonality), available food type, and the water quality. In order to be confident that water quality is the primary factor affecting the macroinvertebrate community, the sampling design must consider the effects of habitat, season, and the available food sources. Collecting in either the wrong place or at the wrong time of year may lead to inaccurate water quality conclusions. The remaining subsections are dedicated to the basic ecology of macroinvertebrates so that an appropriate sampling design can be developed to produce accurate bioassessment results.

What are these organisms?

The first step in understanding the ecology of an organism is to identify the type (plant, animal, bacteria, *etc*.) and its habitat. The biomonitoring procedures followed by Project

SEARCH look at small organisms which have a long but descriptive name. Aquatic ecologists refer to these organisms as ***riffle dwelling benthic macroinvertebrates.*** By looking at the definition of each part of the phrase it will be easy to understand the who, what, and where behind these organisms.

Starting with the last part, ***macroinvertebrate***. An ***invertebrate*** is an organism that does not have a backbone. Several common examples found in or on a stream are leeches, snails, mussels, amphipods, isopods, crayfish, insects and spiders (Figure 8.3.1).

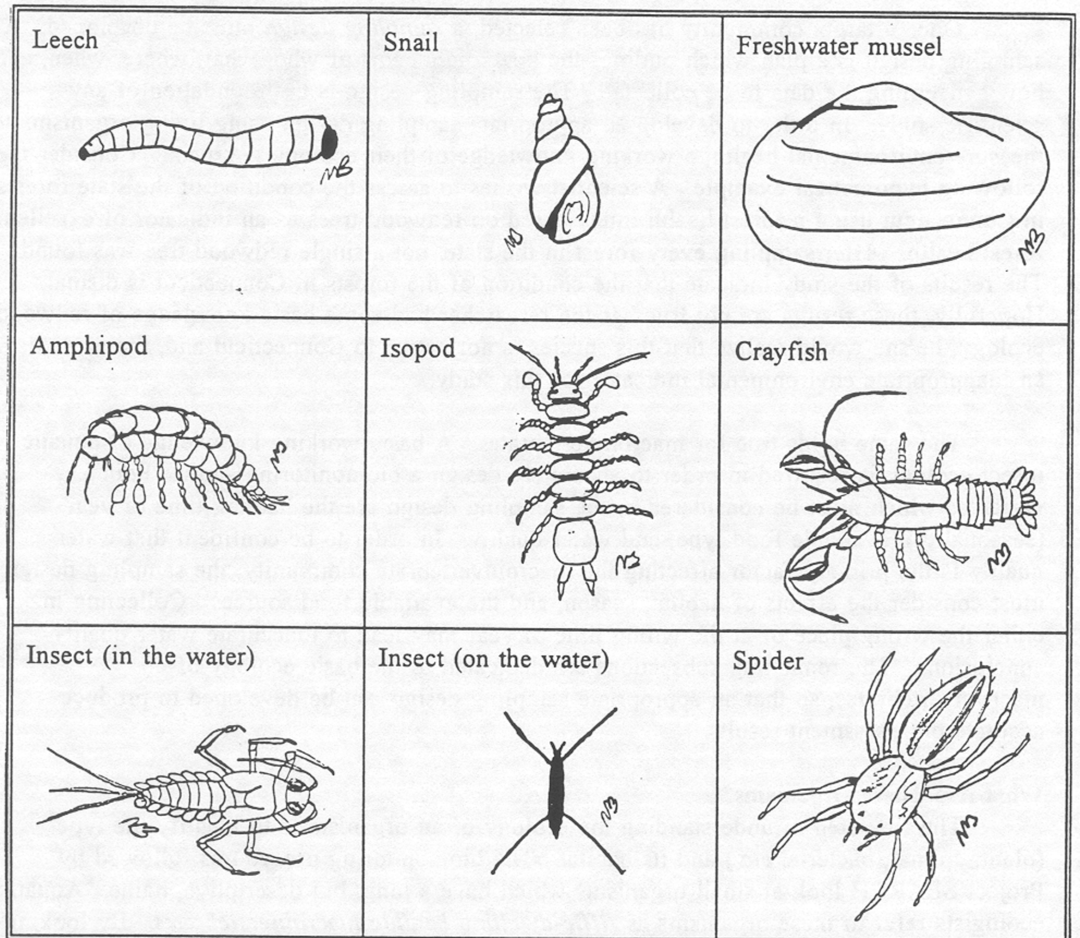

Fig. 8.2. Common invertebrates found in or around a stream.

The prefix ***macro-*** refers to the size of the organism. For example, if one wanted to examine a very tiny object (bacteria, plankton, individual cells, *etc*.) one would use a microscope. When asked to describe the size of the object, hopefully one would say it is microscopic. Macro- is the opposite of micro-. Macroscopic objects are those which can be seen with the unaided eye. What is the smallest object you can see with the unaided eye? Can you see a grain of sand, a bread crumb, or the period at the bottom of this question mark? These are very small objects to say the least but all are considered to be macroscopic.

According to the USEPA a macroinvertebrate is any invertebrate which is retained in a U.S. #30, 600 micrometer mesh sieve (Klemm *et al*. 1990). Along with macroinvertebrates sand, leaves, fine detritus, and a lot of other material is also retained in this sieve. Therefore, when collecting a macroinvertebrate sample a hand lens or dissecting microscope is useful to locate very small organisms mixed in the debris (Figure 8.3).

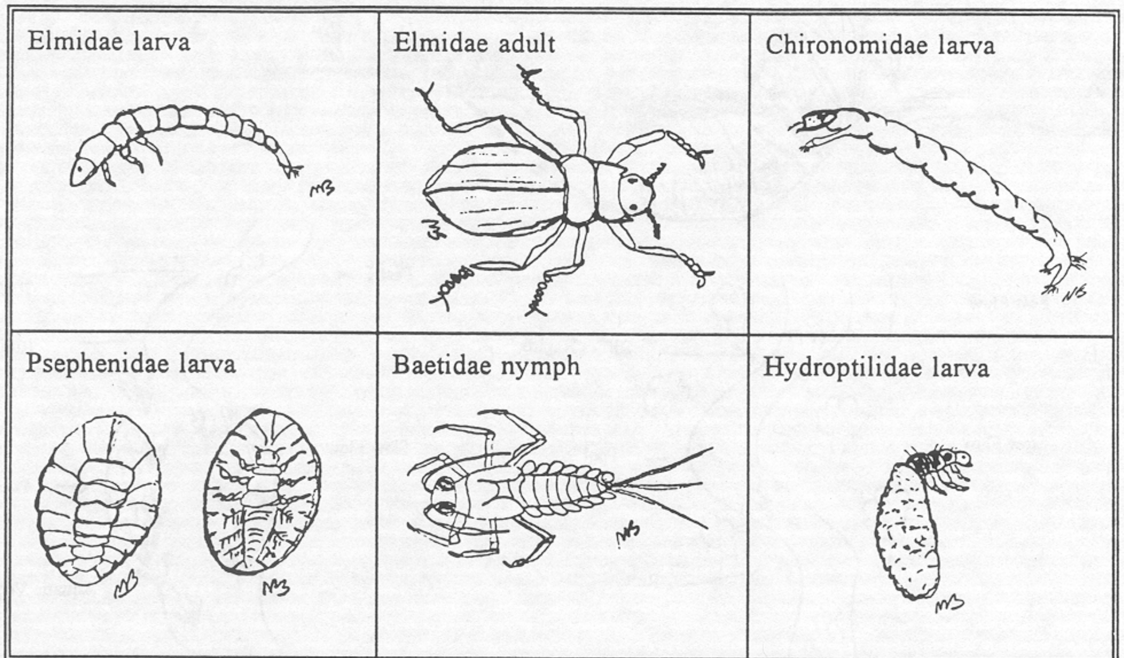

Fig. 8.3. Examples of small macroinvertebrates which are easily overlooked when picking the organisms from the sample debris.

The remaining parts of the term (riffle-dwelling and benthic) describe the organisms' habitat. A ***riffle*** is a specific section of a stream, brook, or river where the water is very turbulent. The turbulence results from the water flowing over a predominantly inorganic ***substrate*** (boulders, rocks, sand and gravel). Riffles are only found in flowing waters (***lotic***) like streams and rivers and are not found in still waters (***lentic***) like lakes and ponds (Figure 8.3.3). ***Dwelling*** is defined as the place where one lives. Riffle dwelling organisms then are those which live in fast flowing turbulent sections of a stream, brook or river.

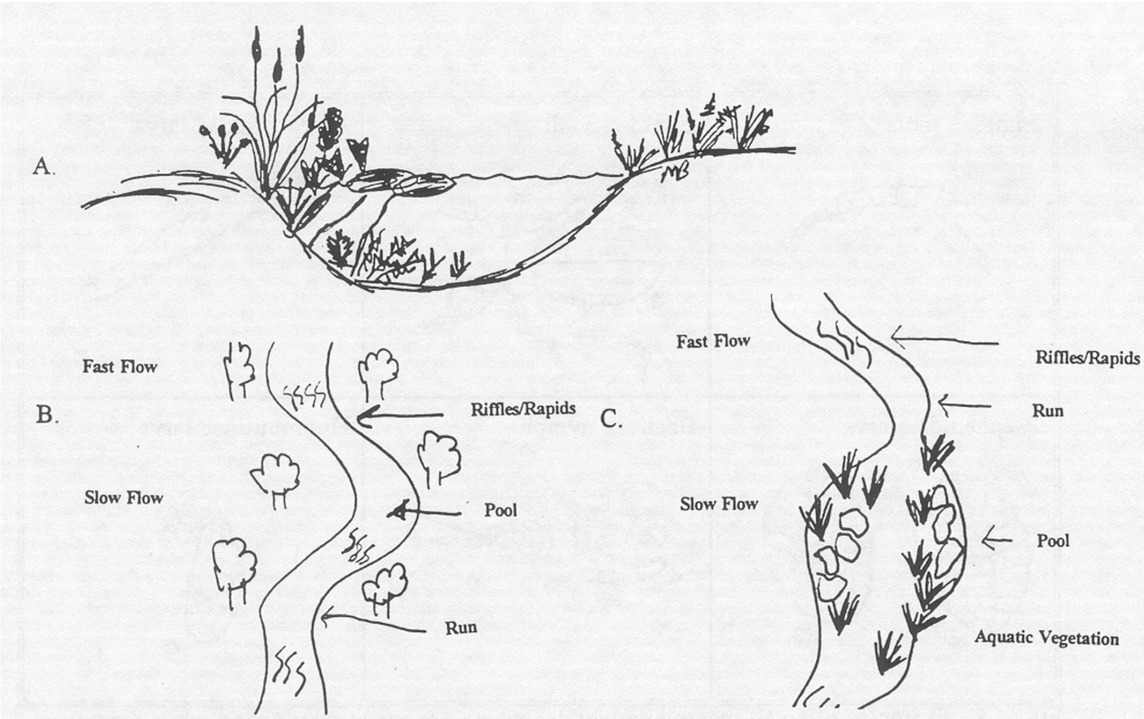

Fig. 8.4. A.) A cross section of a lentic system. B.) A view of a typical lotic system. C.) A lotic system with the inclusion of lentic characteristics.

To complete the phrase, a ***benthic*** organism is one which lives in, on, or attached to the bottom of an aquatic ecosystem (rocks, leaves, sticks, mud, plants, *etc*.). The entire community of such organisms is called the ***benthos***.

In summary, a ***riffle dwelling benthic macroinvertebrate*** is an organism visible to the unaided eye, which does not have a backbone, and lives on/in the bottom of a fast flowing turbulent section of a stream. Riffle dwelling benthic macroinvertebrate communities have many advantages for use in biomonitoring studies over both fish and algal communities (Davies *et al.* 1993). Two important factors to consider when collecting macroinvertebrates are the sampling location in a stream and the sampling technique. First, collections must be located in a riffle area. Even though these organisms may be able to survive in other types of habitats (wave swept lake shores) the populations which most accurately represent water quality are found in riffles. Secondly, when sampling the substrate must be vigorously disturbed by hand. Attempting to collect by lightly disturbing the substrate or sweeping the net through the water column will not capture these organisms. Erroneous water quality conclusions may be drawn if sampling is not thorough or occurs in the wrong habitat or during the wrong time of year (see section 8.4). With slight attention to detail, bioassessment results will be very rewarding to the students performing the tasks, the state DEP, local officials, and scientists who evaluate water quality.

Life cycles of aquatic insects

Insects, unlike many other animals, have relatively short life spans (most only 1-3 years) which leaves little time for growth and reproduction. The life cycle of an insect is the step-wise morphological and physiological progression from egg, to immature insect, to a reproductive adult insect, and back to egg. As a result of this cyclical life cycle, the size and abundance of the insects present in a riffle varies with the season. Therefore when developing a sampling design, it is extremely important to consider the life cycle in scheduling collection trips.

A characteristic common to all insects is the presence of a rigid ***exoskeleton***. While the presence of an exoskeleton has contributed to the massive success of insects on earth, it poses a disadvantage for rapid growth. In order for the insect to grow, the old, smaller exoskeleton must be replaced by a new, larger one. This process is called molting or ***metamorphosis.*** As an insect progresses from egg to adult, it must undergo several molts. The period of growth in between each molt is called an ***instar***. The first instar is the newly hatched insect (from the egg), the instar number increases by 1 each time the immature insect molts (second, third, *etc*.) until the molt to the adult form (final instar).

All insects can be divided into one of two groups based upon the type of metamorphosis to the final instar, incomplete or complete. Insects characterized by ***incomplete metamorphosis (hemimetabolous)*** molt directly to the adult from the immature nymph (Figure 8.5A). Just prior to the final instar, the adult exoskeleton forms beneath the existing one. Once the organism is ready for the final molt, it splits the exoskeleton down the back and crawls out. The newly emerged adult form must allow time for the exoskeleton to harden before heading on its way. The cast off exoskeleton of stonefly, mayfly, and dragonfly/damselfly nymphs can often be found on emergent plants, rocks, or bridge abutments. The major aquatic insect orders which undergo incomplete metamorphosis are ephemeroptera (mayflies), plecoptera (stoneflies), odonata (dragonflies and damselflies), and hemiptera (true bugs).

Insects characterized by ***complete metamorphosis (holometabolous)*** have one additional step in the life cycle. In order to facilitate a complete change, the organism must go through the ***pupal stage*** (Figure 8.5B). Just prior to the pupal stage the insect builds a protective case (called a puparium), which is similar to the cocoon of a caterpillar. Inside this case, the tissues and organs are disassembled and reorganized into the structure of the adult. When the insect emerges, it is both morphologically and physiologically very different. The major aquatic insect orders which undergo complete metamorphosis are trichoptera (caddisflies), megaloptera (dobsonflies, fishflies, and alderflies), coleoptera (beetles), and diptera (true flies).

A. Hemimetabolous life cycle B. Holometabolous life cycle

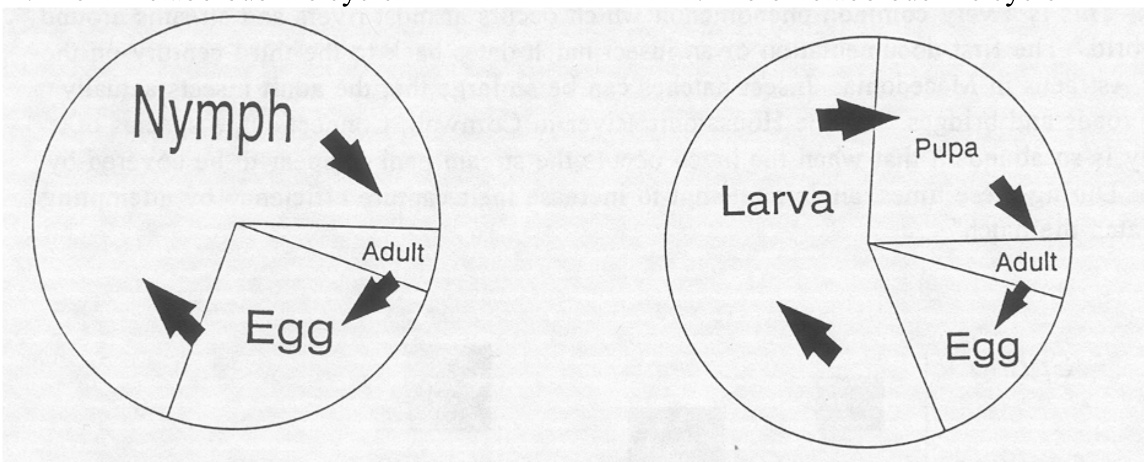

Figure 8.5. A.) The path of incomplete metamorphosis and B.) The path of complete metamorphosis. Each section of the pie represents the approximate amount of time spent in each particular phase.

The majority of the riffle dwelling macroinvertebrates collected are in the immature form (meaning they lack both wings and reproductive structures). Three terms commonly used to describe immature insects are: nymph, larva, and naiad. A ***nymph*** is the immature stage of and insect with incomplete metamorphosis (hemimetabolous), a ***larva*** is the immature stage of and insect with complete metamorphosis (holometabolous), and a ***naiad*** is simply an aquatic nymph.

A term often confused with life cycle is ***voltinism***. If a life cycle is the step-wise progression from egg to adult, voltinism, is the frequency in which the life cycles are repeated. In general, there are several time frames in which a life cycle can be completed. Usually a type of insect will maintain one life cycle frequency. However, this frequency can be altered by environmental conditions such as temperature and food availability. Each of the following terms describes the number of life cycles (generations) completed within a given period of time:

Uni-, Bi-, or Trivoltine = one, two, or three generation(s) per year

Multivoltine = more than one generation per year

Semivoltine = one generation completed in two years

Merovoltine = one generation completed in more than two years

Often the life cycles of individuals within a species are synchronous, meaning the majority of population is in the same life stage at the same time. For many riffle dwelling insects the eggs begin to hatch in late summer. The immature (naiads and larvae) feed and grow throughout fall and winter. The adults emerge in late spring and early summer (Figure 8.6). The simultaneous emergence of the adult form of a species of aquatic insect is called a ***hatch***. This is a very common phenomenon which occurs at most rivers and streams around the world. The first documentation of an insect hatch dates back to the third century on the River Astraeus in

Macedonia. Insect hatches can be so large that the adult insects actually cover roads and bridges. On the Housatonic River in Cornwall, Connecticut, a species of mayfly is so abundant that when the hatch occurs the stream banks appear to be covered by snow. During these times, anglers attempt to increase their capture efficiency by attempting to "match the hatch".

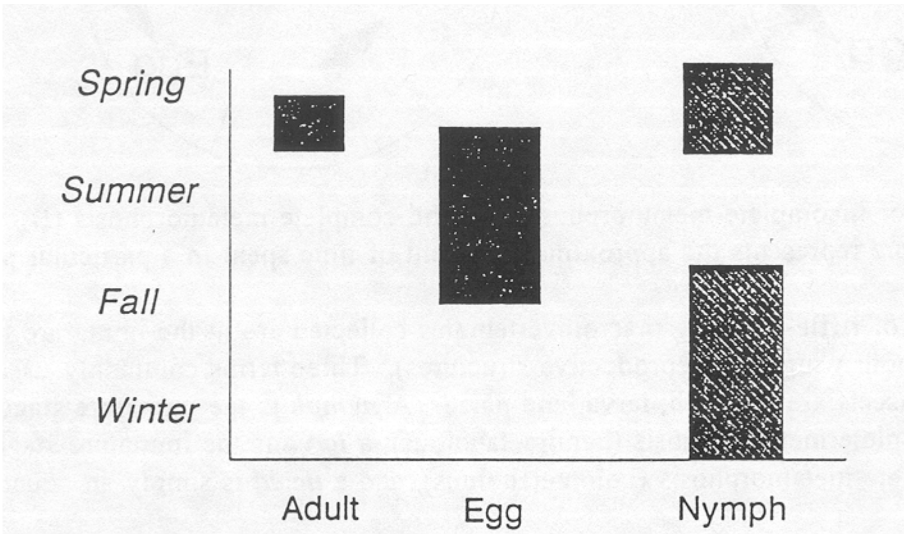

Figure 8.6. The seasonal distribution of the life stages of a univoltine riffle dwelling benthic insect.

In summary, knowledge of life cycles is very important for accurate bioassessment studies. Not only does sampling have to be in the correct habitat,but it must occur at the correct time of year. In order to obtain a representative population sample, collections must occur when the majority of the members of the insect community are present in the riffle. Figure 8.7 shows how riffle insect abundance, size, and biomass change throughout a typical year in Connecticut. Stream insect abundances are highest in the fall immediately following egg hatching, however, these insects are extremely small. During the winter months, abundance decreases while the size and biomass increase. By spring the abundance is much lower than fall, however, the insects are much larger in both size and mass. Following the emergence of the adults in the spring, abundance and biomass drop dramatically. During the summer months many insects are in the egg stage. Neither abundance nor biomass represent true population conditions and sampling to determine water quality should be avoided. Additionally, spring collections must be planned carefully. Many families begin to "hatch off" when the water temperature is approximately 15 degrees centigrade. Once the stream temperature is above this level some species may not be represented in collections. This may result in a bioassessment that falsely indicated impaired water quality.

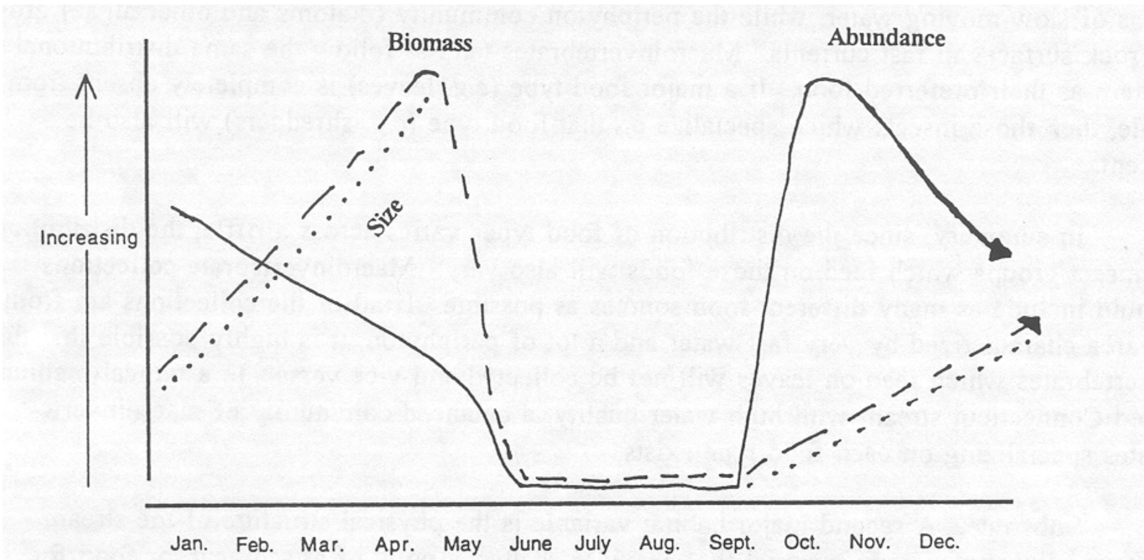

Figure 8.7. A seasonal time line of insect abundance (solid line), size (dotted line) and biomass (dashed line).

The riffle dwellers' habitat

Habitat can be defined as a specific area where a plant or animal naturally grows and lives. There are 3 major types of stream habitats: pools, runs or glides, and riffles. Each habitat type is comprised of a variety of ***biotic*** (living) and ***abiotic*** (non-living) parameters. The combination of the different habitat parameters often has a direct influence upon both the type and abundance of organisms present.

Several key riffle habitat parameters are food availability, substrate, current velocity, and dissolved oxygen levels. Assuming that a collection will be taken from a riffle in a medium-sized stream in Connecticut, lets look at how each of the preceding habitat parameters may influence the macroinvertebrate community. **Note:** Even though each habitat parameter may appear to have a distinct and identifiable influence on the macroinvertebrate community often it is very difficult to isolate the specific effect of one parameter on the benthic community.

Food availability: The overwhelming majority of the riffle dwelling aquatic insect life cycle is spent as an immature. The primary function during this time is to feed and grow rapidly enough so to hatch at the right time and reproduce. Therefore food (both type and abundance), influences the invertebrates (both type and abundance), within a riffle (Eggilshaw 1979). Food types can be either ***coarse particulate organic material, CPOM***, (leaves, sticks, algae, *etc*.) or ***fine particulate organic material, FPOM***, (suspended solids, insect feces, *etc*.). The presence and distribution of each depends upon the surrounding land use and the physical structure of the riffle. Riffles tend to have a several micro-habitats resulting from different current velocities and substrate types. Within each of the micro-habitats food resources may be slightly different. For example, coarse particulate material accumulates in areas of slow moving water, while the periphyton community (diatoms and other algae) grows on rock surfaces in the fast currents. Macroinvertebrates tend to follow the same distributional pattern as their preferred food. If a major food type is completely absent from a riffle (*i.e.* leaves), then those insects which specialize on that food type (shredders) will also be absent.

In summary, since the distribution of food types varies across a riffle, the distribution of insect groups which feed on those foods will also vary. Macroinvertebrate collections should include as many different food sources as possible. If all of the collections are from an areas characterized by very fast water and a lot of periphyton, it is highly possible that the invertebrates which feed on leaves will not be collected and vice versa. In a typical medium-sized Connecticut stream with high water quality, a balanced community of macroinvertebrates specializing on each food type exists.

Substrate: A second major habitat variable is the physical structure of the stream bottom. ***Substrate*** is any material that provides shelter, a point of attachment, or food for benthic organisms (Resh and Rosenburg 1984). Substrate can be either ***organic*** (leaves, sticks, aquatic plants, algae, *etc.*), or ***inorganic*** (rocks, concrete, shopping carts, cars, *etc*.). The substrate of most stream systems is usually a mixture of both. Since all riffle dwelling macroinvertebrates are benthic, the substrate has a major influence on the macroinvertebrate community.

Although the inorganic substrates tend to be more visible and dominant in riffles (lotic erosional systems), organic substrates can be very prevalent in localized areas. Organic substrates such as aquatic mosses, plants, filamentous algae, and leaf-packs provide large surface areas for attachment, while concurrently serving as a food resource.

Even though a substantial amount of organic substrate can occur in a riffle, the inorganic types have a greater influence on the benthos. Inorganic substrates are classified by the diameter of the particle (Table 8.1). Streams with a ***heterogeneous*** mix (many shapes and sizes) of large gravel and cobble have the most diverse communities, while ***homogeneous*** substrates, (all the same type and size) have a lower diversity of insects (Williams 1980). Consider a section of stream with one of the two substrate scenarios, a bedrock outcropping or entirely small gravel. In each case the substrate is homogenous and may limit insect diversity and abundance. The water quality in each stream may be excellent. However, due to the physical constraints of the substrate, invertebrate diversity might be low. Note: Streams containing large amounts of shifting sand are very unstable and poor habitat for the majority of riffle dwelling families.

Table 8.1. The Wentworth size and classification scale of inorganic substrates (Modified from Ward 1992).

Size	Category	Diameter (mm)	Size	Category	Diameter (mm)
Boulder		> 256	**Sand**	Very Coarse	1-2
Cobble	Large	128-256		Coarse	0.5-1
	Small	64-128		Medium	0.25-0.5
Pebble	Large	32-64		Fine	0.125-0.25
	Small	16-32		Very Fine	0.063-0.125
Gravel	Coarse	8-16	**Silt**		< 0.063
	Medium	4-8			
	Fine	2-4			

Another factor which affects the habitat quality of the substrate, is embeddedness. **Embeddedness** refers to the how much an object is surrounded, buried, or covered by smaller material. A person buried up to his/her neck at the beach could be described as 95% embedded. The percent embeddedness of the substrate has a direct effect on the quality of the riffle habitat. As the percent embeddedness increases, the spaces between the larger rocks (***interstitial spaces***) become filled. Interstitial spaces are an important component of the benthic habitat. These spaces protect the organisms from the stream current, predators, and traps organic material for food. As sand/silt fills these spaces, the quality of the habitat is substantially lowered (Chutter 1969). During a habitat assessment (See: Activity 4A) check the amount of embeddedness by trying to move the cobble-size rocks. If they are moved easily, the substrate has a low percent embeddedness. In such areas the interstitial space provides enough habitat to support a large insect population.

A final component of the substrate habitat, is called the Hyporheic zone. The **hyporheic zone** is the portion of the stream bottom below (hypo-) the flowing water (rheic) (Figure 8.5.1). In general it is the area immediately below the larger surface substrate and the stream banks. In order to collect macroinvertebrates from the hyporheic zone, it is important to "dig in" or "scrape down" several centimeters into the stream bottom. The hyporheic zone can extend a meter or more, depending upon the geology of the stream bottom (Stanford and Gaufin 1974). The higher the quality the hyporheic zone, the more diverse the invertebrate community will be (Resh and Rosenburg 1984). The hyporheic zone serves as a refuge area in times of extreme drought conditions and also as a safe environment for immature aquatic insects. In streams with heavy embeddedness, the hyporheic zone is effectively eliminated.

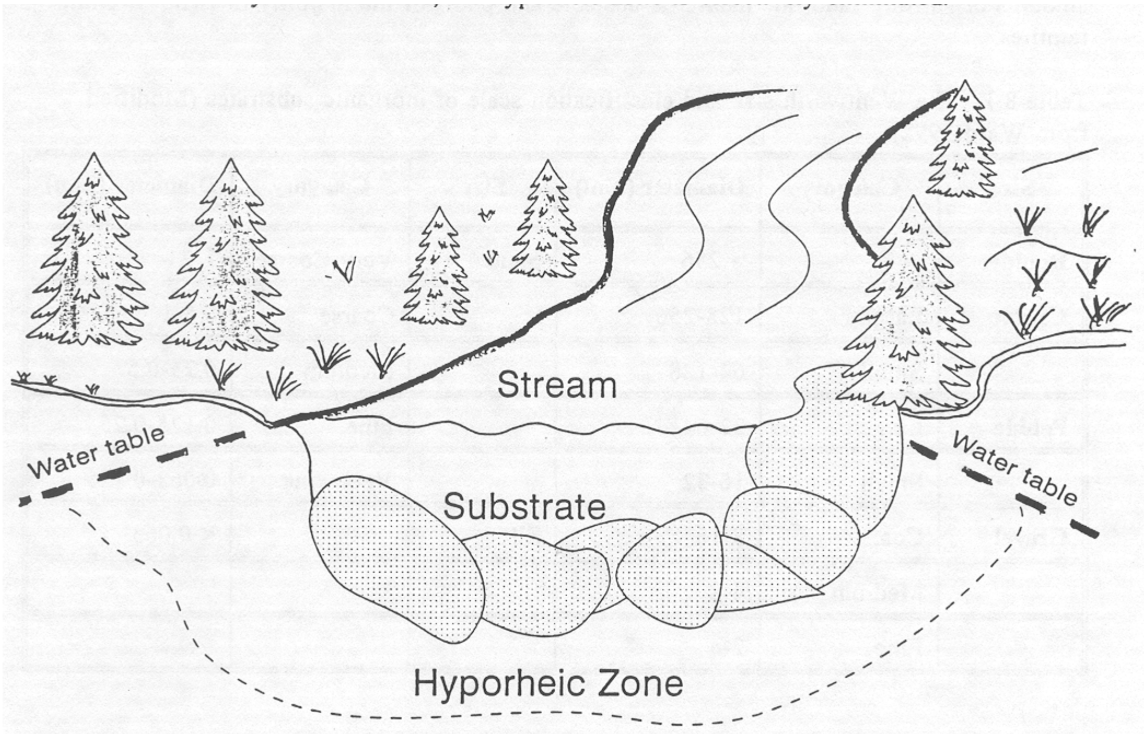

Figure 8.8. A cross-section of a stream bottom with a high quality hyporheic zone.

Imagine a stream flowing through a drain pipe or concrete culvert (Figure 8.9). What is the quality of the hyporheic zone? Does a hyporheic zone even exist? What percent of the substrate is embedded? Obviously, this situation has a negative impact on the organisms which normally live in the hyporheic zone. What happens to the hyporheic zone when a stream becomes muddy? What causes the stream to become muddy? What long term effects to the hyporheic zone do you see happening here?

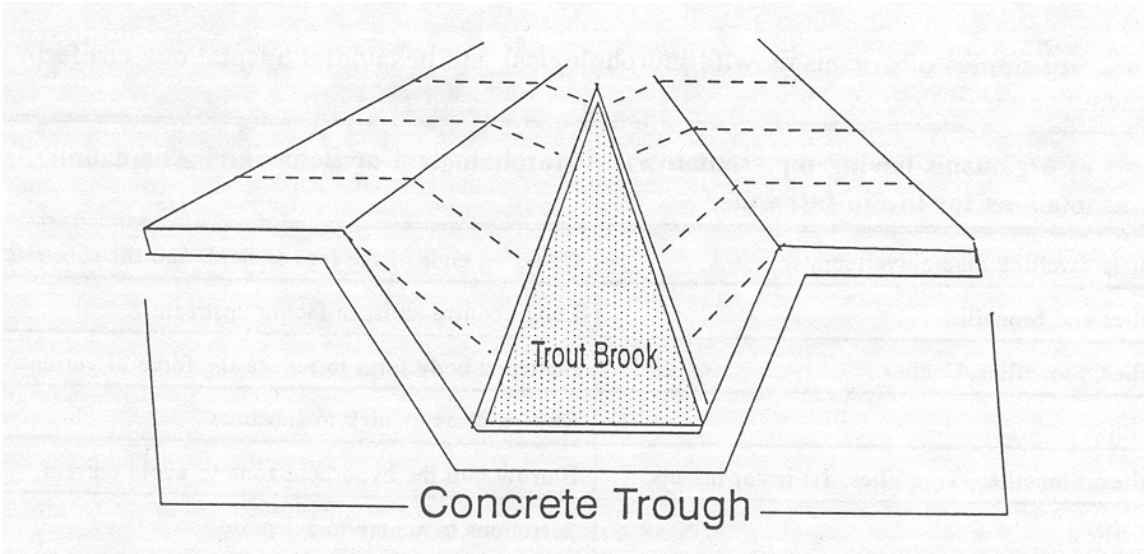

Figure 8.9. A cross sectional view of Trout Brook in West Hartford, Connecticut.

In summary, the riffle substrate is very important to consider in water quality studies. As the size and type of substrate particles becomes less heterogeneous, insect diversity tends to decrease. Before drawing conclusions about water quality it is important to consider the effects of substrate on insect diversity. Each of the three substrate components, composition, embeddedness, and hyporheic zone may affect the benthos. Consider the following scenario: a few insect families are collected from a small, high quality woodland stream. An evaluation using taxa richness may conclude possible impacts to the water quality. However, the low number of families collected may be an artifact of the limited habitat provided by the large boulder substrate typical of small woodland streams.

Current velocity: Despite the apparent harsh erratic conditions caused by torrential flows, fluctuating water levels and temperatures, on a geologic time line lotic waters are very stable. The continual uni-directional water flow provides a predictable and stable habitat. In response to the challenge posed by the force of fast flowing water, macroinvertebrates have developed morphological, physiological, and behavioral adaptations (Table 8.2). Morphological adaptations are structures which help the organism remain in or on the substrate and not be swept downstream. Several include tarsal claws (toes), suction discs, long cerci (tails), and fattened and streamlined body forms. Physiological adaptations are changes in metabolic body functions specifically for an aquatic existence. Two major physiological adaptations are passive respiration and "anti-freeze like" chemicals. Behavioral adaptations include current avoidance, limited mobility, and drifting.

Table 8.2. Examples of organisms with morphological and behavioral adaptations to life in fast-flowing water.

Groups of organisms with representatives with adaptations for life in fast water	**Morphological or Behavioral Adaptation**
All riffle dwelling macroinvertebrates	Claws on ends of the legs to hold onto the substrate
Mayflies and Stoneflies	Long cerci to assist in facing upstream
Mayflies, Stoneflies, Beetles	Flattened body form to reduce the force of current
True Flies	Suction discs to stick to substrate
Mayflies, Stoneflies, True Flies, 1st instar insects	Burrow into the hyporheic zone to avoid current
Caddisflies	Secretions to adhere to substrate

As a result of adaptation to life in fast flowing water, riffle dwelling macroinvertebrates have become so specialized that they are functionally restricted to riffle areas. Although these organisms may be able to develop isolated populations in other aquatic environments which may have some riffle characteristics, permanent stable populations occur only in riffle areas. Unlike many other adaptations, passive respiration can actually limit an organisms ability to live in different habitats.

Riffle dwelling insects use external gills to obtain oxygen directly from the water without expending any metabolic energy. The stream current determines how much oxygen is available to insect. Generally, in very fast-turbulent water dissolved oxygen levels are near saturation. As water velocity and turbulence slows, so does the amount of oxygen passing over an insect per unit time. If the current slows too much, insects may not be able to extract enough oxygen and may eventually suffocate. (See the following heading **Dissolved oxygen** for further discussion.)

Stream current is also intimately linked to substrate composition and food availability (Rabenini and Minshall 1977). Current influences the substrate composition by sorting the substrate by size. For example fast currents scour at the stream bottom, transporting substrates downstream until the decreasing velocity can no longer move each particular size substrate. The larger the substrate the greater the force required to move it downstream. In sections of a riffle with very fast currents the substrate will consist of large boulders, areas of moderate current will consist of cobbles, gravel, and sand, while areas with little or no current (behind large boulders or the margins of the stream) will consist of fine sediments and detritus.

The stream current also influences the distribution of food types across a riffle. In the fastest sections of a riffle, diatoms and other algae grow attached to the surface of the rocks. Non-attached organics like CPOM and FPOM are carried through the fast sections of the riffle and tend to accumulate in slow sections of the riffle such as backwater eddies and stream margins. The greatest diversity of insects occurs in riffles characterized by several different current velocities which allows for a mix of substrate and food sources (Ward 1975).

In summary, current velocity is an important consideration in the selection of a site for water quality studies. Since the target organisms for a bioassessment are highly adapted for life in fast flowing water, it is extremely important to sample in a riffle area. Because of the fast flowing turbulent water, riffles are easily differentiated from other stream sections like runs and glides (intermediate flow) and pools (very little flow). The long-term stability of river and stream systems is created by the continual uni-directional flow. As a result of the predicable nature of the lotic environment, a very specialized macroinvertebrate community has developed. The distribution and diversity of this community is often directly related to both the substrate composition and food availability within this habitat.

Dissolved oxygen: ***Dissolved oxygen (D.O.)*** is the amount of oxygen in solution. The dissolved oxygen level is influenced by several factors, most notably temperature. As the temperature of a solution increases the solubility of a gas decreases (see Chapter 7 Chemical Parameters). In riffles areas turbulence, caused by water flowing over rocks, physically forces large volumes of oxygen into the water. As a result dissolved oxygen levels are at or near saturation most of the time.

In response to this oxygen-rich environment, riffle dwelling insects have evolved to be ***osmoconformers***, using external gills to passively remove oxygen from the water. The major advantage to being an osmoconformer is that through passive respiration no metabolic energy is required. The disadvantage is the insect can not adjust or regulate the amount of oxygen entering its body. Therefore the insect must remain in an oxygen-rich environment. If a riffle dwelling insect becomes dislodged and floats into an area of slow water, over time the dissolved oxygen level surrounding the insect is depleted. Since the organism is now in an area with little flow, the oxygen level is not being replenished. As surrounding oxygen level continues to decline, an insect may begin to do "push-ups". These movements are an attempt to actively force oxygen containing water over the gills. If an organism remains in this environment, eventually all available oxygen is depleted, and the organism will suffocate.

Dissolved oxygen levels are usually very high in riffle areas (7-14 ppm). However, there are two situations when the D.O. level may be too low to support pollution sensitive families. The first is during the summer months. As the water temperature increases with summer heat and the stream flow is reduced, the D.O. levels can become low (< 7ppm). Many riffle-dwelling insect families avoid this problem by leaving the riffle completely. The adults hatch in late spring, mate and lay eggs. The insect eggs then remain in or around the stream throughout the summer months when D.O. levels are the lowest. The insect eggs have structural features which prevent desiccation (drying out). The eggs hatch at the end of the summer when the D.O. levels are increasing.

The second situation where D.O. levels may become low is when there is a high Biochemical Oxygen Demand (B.O.D). B.O.D. is the amount of oxygen consumed through the decomposition of organic material (primarily bacterial respiration). The direct effect of high B.O.D. levels on riffle insects is not well known. However, streams with high B.O.D. levels also tend to have increased organic loads which has been shown to decrease insect diversity.

In summary, the major effect of the dissolved oxygen level on riffle insects is that, because they are osmoconformers unable to regulate the rate of oxygen uptake, they must remain in an oxygen rich environment like a riffle. Although some of these insects can survive in slow water (pools, runs, and glides) for short periods of time, the greatest numbers are found in riffle areas. Generally D.O. is not a limiting factor in a riffle system but may influence the community in the following situations, summer drought, intermittent streams, groundwater seeps, and streams with high B.O.D. levels.

Functional feeding groups: The primary energy source in a stream ecosystem is detritus (decomposing organic material). This is much different than other ecosystems in which primary productivity (photosynthesis) is the major energy source. Despite the reduced input from primary productivity, rivers and streams are able to support a large biomass of secondary and tertiary consumers (macroinvertebrates and fish).

Detritus can originate from either outside the stream (allochthonous) or from within the stream (autochthonous). Each source is divided into 2 major forms, particulate organic matter (POM) and dissolved organic matter (DOM). Particulate matter in turn can be either coarse particulate organic matter (CPOM: leaves and other large detritus) or fine particulate organic matter (FPOM: feces, colloidal material, broken down leaves). The relationship of food sources and insect consumers is shown in Figure 8.10.

Since rapid and efficient growth is extremely important to reproductive success, insects have morphological and behavioral adaptations to take advantage of one of three broad categories of food resources (detritus, living plants, or other organisms). Aquatic ecologists place all aquatic insect families into one of five major functional feeding groups. Each group, scrapers, shredders, collector-gatherers, collector-filterers, and predators are differentiated by the type of food utilized as well as the feeding method (Cummins1984).

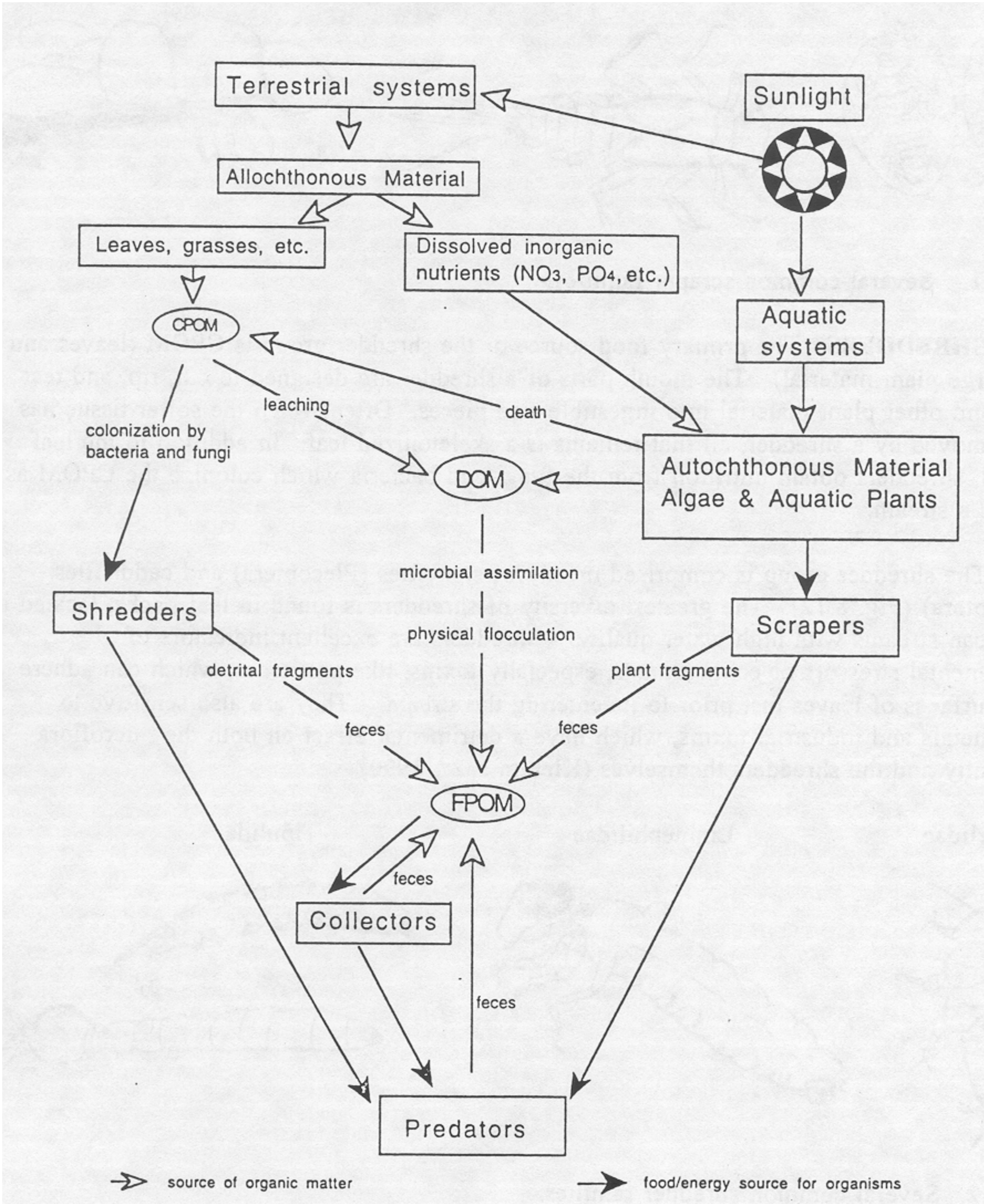

Figure 8.10. The relationship between food source and the feeding group of riffle insects. Adapted from Merritt and Cummins 1984. FPOM is fine organic matter, CPOM is coarse particulate organic matter, DOM is dissolved organic matter, POM is particulate organic matter.

Two feeding groups, scrapers and shredders, use CPOM as the primary food source. Both groups tend to be intolerant to pollution inputs and found in streams characterized by good to excellent water quality.

SCRAPERS: Scrapers are the only macroinvertebrate feeding group whose main food source is based on primary productivity. Similar to terrestrial herbivores, scrapers feed on living plant material. In streams, the community of algae and diatoms growing on the surface of the substrate is called ***periphyton***. Scrapers have specialized mouth-parts which enable them to pry or scrape the periphyton from the substrate. Two examples of the modified mouthparts are short rigid bristles (like those on a wire brush) and chisel-like mandibles. Additionally, since most periphyton communities are on the exposed surfaces of rocks in very swift currents, many scrapers have very flattened body forms (*i.e.* water penny beetles). The extra flat body enables the organisms to feed on the surface of rocks without being swept away by the current.

Several aquatic insect orders contain families belonging to the scraper feeding group (Figure 8.11). The greatest diversity of scrapers are found in fast, cold, clean streams which contain large diatom populations. Because FPOM and filamentous algae tend to decrease the periphyton community, scraper diversity also tends to decrease with increasing organic influence.

Psephenidae Heptageniidae Glossosomatidae

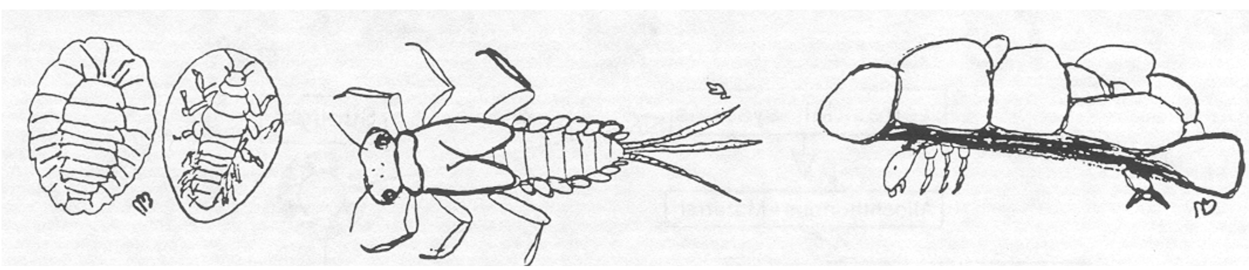

Figure 8.11. Several common scraper families.

SHREDDERS: The primary food source of the shredder group is CPOM (leaves and large plant material). The mouth parts of a shredder are designed to cut, rip, and tear leaves and other plant material into ingestible size pieces. Many times once the softer tissue has been removed by a shredder, all that remains is a skeletonized leaf. In addition to the leaf material, shredders obtain nutrition from the fungi and bacteria which colonize the CPOM as it enters a stream.

Shredders group is comprised mostly of stoneflies (Plecoptera) and caddisflies (Trichoptera) (Figure 8.12). The greatest diversity of shredders is found in leaf packs, located in cold, clean streams with high water quality. Shredders are excellent indicators of environmental stressors or contaminants, especially toxins (for example: pesticides) which can adhere to the surfaces of leaves just prior to its entering the stream. Shredders are also sensitive to heavy metals and industrial toxins, which have a detrimental effect on both the microflora community and the shredders themselves (Klemm et al. 1990).

Peltoperlidae Limnephilidae Tipulidae

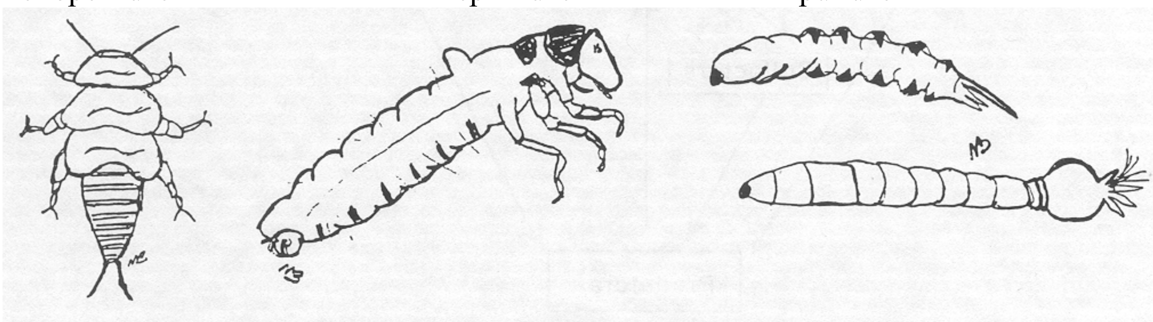

Figure 8.12. Several common shredder families.

PREDATORS: Predators are the only carnivorous feeding group. Macroinvertebrate predators are similar to predators from any other food web in that they feed upon other organisms in the ecosystem. The predators are commonly the largest macroinvertebrates and may take several years to reach the adult stage (Figure 8.13).

Although every order which is commonly found in riffle areas (except ephemeroptera) contains at least 1 predatory family, all of the families in odonata (damselflies and dragonflies) and megaloptera (alderflies and dobsonflies) are predaceous (Table 8.3). As a group predators have a range of pollution tolerances. Additionally, because they are not linked to any one food source, predators can be found throughout a riffle.

Gomphidae Perlidae Corydalidae Athericidae

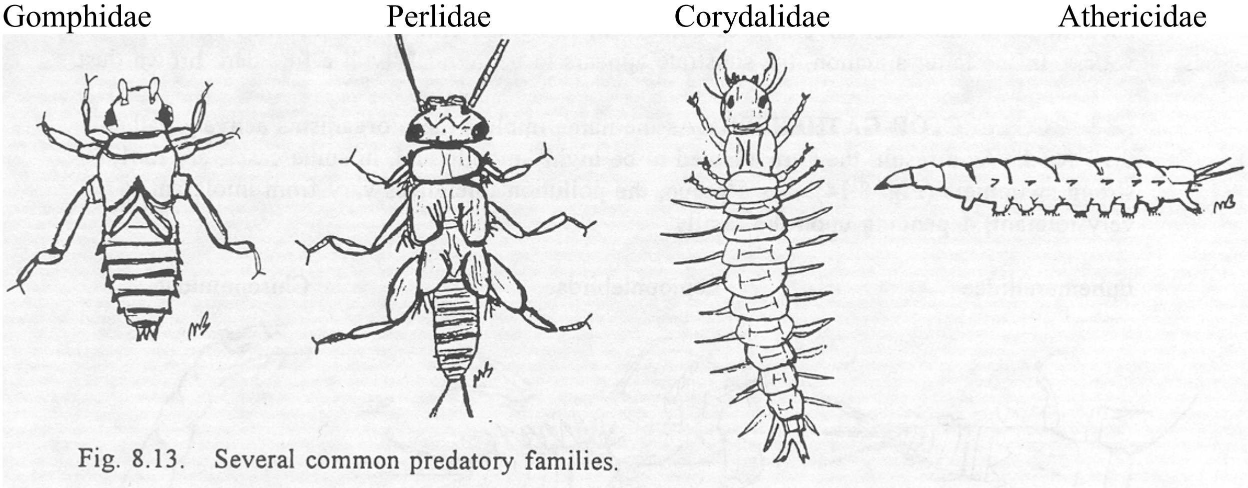

Fig. 8.13. Several common predatory families.

Table 8.3. Insect orders and representative predatory families.

Insect Order	**Representative family(ies)**
Coleoptera (beetles)	Dytiscidae, Haliplidae
Diptera (true flies)	Athericidae, Empididae, Tabanidae
Ephemeroptera (mayflies)	None
Megaloptera (dobsonflies and alderflies)	All families (only 2, Corydalidae and Sialidae)
Odonata (damselflies and dragonflies)	All families (*i.e*. Coenagrionidae, Gomphidae, Aeshnidae)
Plecoptera (stoneflies)	Chloroperlidae, Perlidae, Perlodidae
Trichoptera (caddisflies)	Rhyacophilidae

The two remaining functional feeding groups both collect FPOM. Fine particulate organic matter is considered to be less than 1 mm in diameter. It can be formed naturally from the break down of CPOM and decomposition or it can be a product of increased organic loading. Fine particulate organic matter tends to accumulate on the outer coverings of diatoms in fast water and on the substrate in slower water. In the latter situation, the substrate appears to be covered in a fine dark brown dust.

COLLECTOR/GATHERERS: As the name implies these organisms actively gather their food items. Since these organisms actively search for food items, they are adapted to be highly mobile and in some cases are fairly strong swimmers (Figure 8.14). As a group the pollution tolerances vary from intolerant to very tolerant depending upon the family.

Ephemerellidae Leptophlebiidae Chironomidae

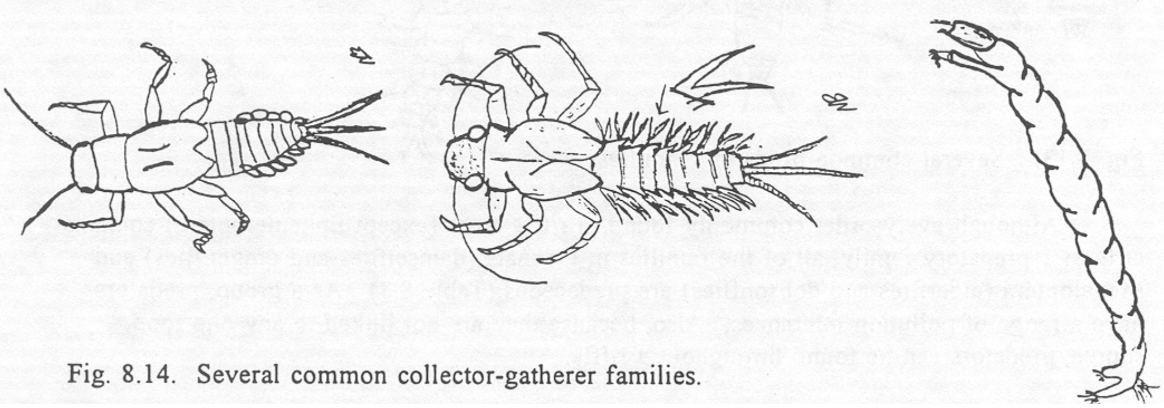

Fig. 8.14. Several common collector-gatherer families.

COLLECTOR/FILTERERS: Unlike gatherers, filterers feed passively (wait for food to come to them) by straining the water. The most common filterers are the net spinning caddisfly families (Hydropsychidae, Philopotamidae, and Polycentropodidae). Members of these 3 families build small funnel shaped webs on the stream bottom. As stream water flows past, some of the FPOM carried by the water becomes trapped. The insect then feeds on the captured particles. Other filterers have developed special structures to strain FPOM from the water. Simuliidae (black flies) have feathery appendages near the mouthparts and a posterior suction disc which enables the insect to remain attached to the substrate. The mayfly family Oligoneuriidae (minnow mayfly), has a double row of long hairs on the inside surface of the front legs. Both of these insects strain the water flowing through their legs, effectively filtering the FPOM (Figure 8.15).

As a group, the collector/filterers tend to have moderate to high pollution tolerances and tend to be the most opportunistic and prolific of all macroinvertebrates. In streams with increased organic loads and high quantities of FPOM, collector/filterers can be extremely abundant (hundreds of organisms in 11 kicks).

The two most common filterer families in Connecticut are Hydropsychidae and Simuliidae (Beauchene 1994). The greatest abundance of collector/filterers are often found immediately downstream of both wastewater treatment plants and eutrophic lakes and ponds. Filterers are a good indicators of organic enrichment. When a stream sample is dominated by Hydropsychidae (greater than 60%), organic pollution is very likely. It is important to note that there are some members from both of the above families which have low tolerance to pollution and live in high quality, low nutrient waters.

Web-building filterers		Attached filterer	Free-moving filterer
Hydropsychidae	Philopotamidae	Simuliidae	Oligoneuriidae

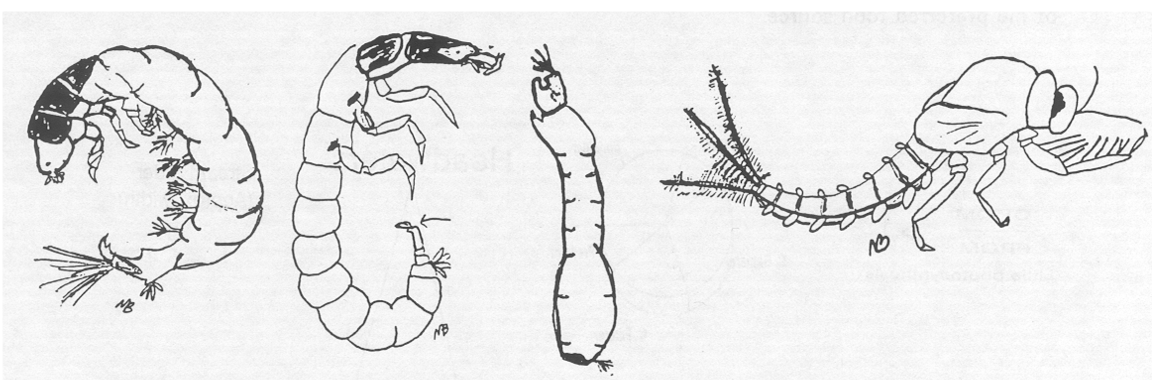

Figure 8.15. Several common collector-filterer families including web-building, attached, and free-living types.

In summary, feeding groups provide valuable information about the types of energy inputs driving a stream ecosystem. Since each feeding group targets a particular food type and the distribution of the food types is determined by the stream current, it is important to sample representative areas across the riffle. If areas harboring a particular food type are not sampled then an entire feeding group may be missed. For example, scrapers (feed on periphyton) are found in fast current on the surface of the substrate, shredders (feed on dead leaves) are found in slower waters behind large rocks or under larger rocks, collectors (feed on FPOM) and predators (feed on other organisms) are found scattered throughout a riffle. If all areas are sampled thoroughly, the absence or dominance of a particular functional feeding group provides information about water quality.

Functional feeding groups are the focus of a major ecological theory. Baring any water quality impacts, the river continuum concept (Vannote et. al. 1980) relates changes in the community structure from headwaters to the stream mouth to the changes in the dominant food source. The theory states that the food resources of headwater streams are almost entirely from outside the stream (leaves, twigs, *etc*.). Since most headwater areas are heavily forested, CPOM dominates the food supply. Forested banks shade the substrate and limit primary productivity. Since the headwaters are the uppermost reaches of a stream, nutrients levels and FPOM tend to be low. Therefore the benthic community will be comprised mostly of shredders, collector-gatherers, and predators. As the size of the stream (stream order) increases, the amount of CPOM input decreases and primary productivity increases. Additionally, much of the CPOM from the headwater areas is physically and chemically reduced to FPOM through decomposition and invertebrate feeding. In the lower reaches the benthic community shifts toward scrapers, collector-gatherers, and collector-filterers. Finally near the mouth of the system, FPOM is the dominant food source and the benthic community consists mainly of collector filterers and collector-gatherers (Figure 8.16). Therefore, one must check to see if the absence of a particular feeding group is related to lack of the preferred food source or actually water quality.

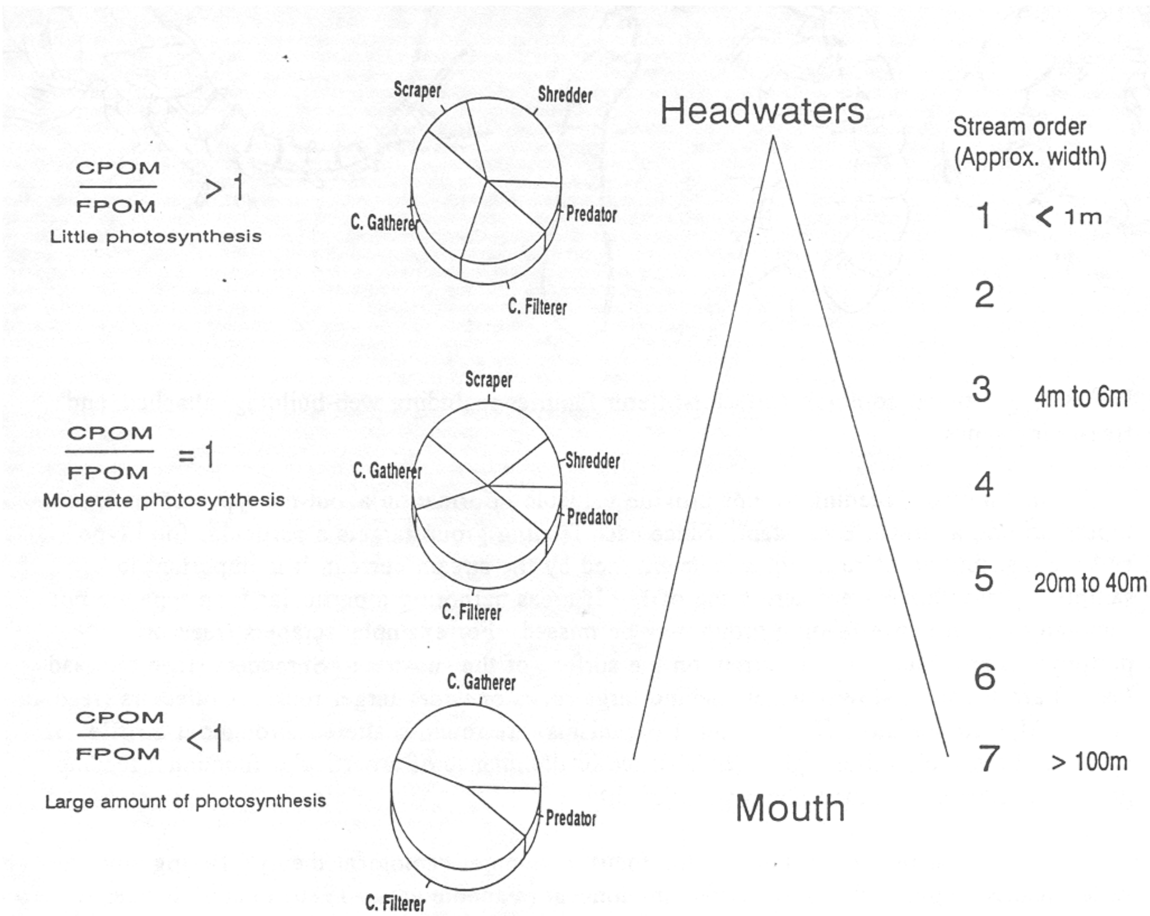

Figure 8.16. A simplified version of the River Continuum Concept (Modified from Vannote *et. al* 1984).

Using metrics and bioassessment procedures to determine water quality

Calculating metrics:

A ***metric*** is a numerical measure of a particular component of the biological community. Metrics are commonly used to determine community balance, diversity, environmental conditions in general, dominant food sources, and potential water quality influences (organic and inorganic). A bioassessment involves a mathematical comparison of the metrics from a study stream and a reference stream in order to determine the level of biological impairment.

The following are brief descriptions of the metrics used by Project SEARCH. Specific calculation instructions for each metric are found in Student Activity 8 D: Macroinvertebrate metric calculation Sheets. Table 8.5 presents relative water quality conditions for Connecticut streams and corresponding metric values.

Taxa Richness: *measure of:* community balance
interpretation: higher number = higher water quality

description: Taxa are units of biological classification that form a hierarchy made up of seven different levels (Figure 8.17). Each level is made up of organisms having a number of shared traits. The species level is the finest level of identification, and it is a group of organisms which share a unique morphological or physiological trait. Closely-related species are grouped into a genus, similar genera are grouped into a family, and so on. The taxa richness values calculated by Project SEARCH are to the family level.

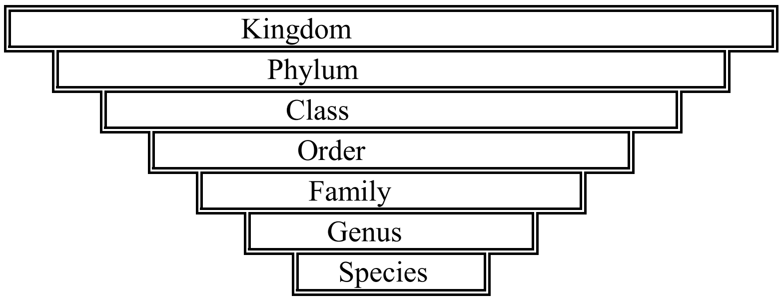

Figure 8.17. The taxonomic hierarchy. Each level contains representatives from the lower level. For example a genus is made up of a species or several species, a family is made up of a genus or several genera, and so on.

ecological background: streams with high water quality tend to be more physically and chemically stable. The stability of a habitat is directly related to the diversity of organisms. A stable habitat can support a very diverse community of benthic organisms. As water quality decreases, so does the stability of the habitat. The decreased habitat stability leads to a decrease in types of organisms present. A listing of the number of arthropod groups which can potentially be collected in Connecticut is presented in Table 8.4.

Table 8.4. A partial listing of arthropods which can be collected from the streams of Connecticut. All organisms listed belong to the Kingdom *Animalia*, Phylum *Arthropoda*. The list was developed from the CT DEP 1994 cumulative taxa list.

Taxon	Common name	Orders	Families	Genera	Species
Class=Hydrachnida		1	6	7	?
Order=Acari	water mites	1	6	7	?
Class=Crustacea		3	4	4	8
Order=Isopoda	aquatic sow bugs		1	1	2
Order=Amphipoda	scuds		3	3	4
Order=Decapoda	crayfish		1	1	2
Class=Insecta		7	73	248	396
Order=Ephemeroptera	mayflies		13	31	74
Order=Odonata	dragonflies/damselflie		9	28	36
Order=Plecoptera	stoneflies		9	32	67
Order=Megaloptera	dobsonflies/fishflies		2	3	3
Order=Trichoptera	caddisflies		17	54	69
Order=Coleoptera	beetles		8	24	37
Order=Diptera	true flies		15	76	120

EPT Index: *measure of:* diversity of pollution sensitive orders
interpretation: higher number = higher water quality

description: This metric is similar to taxa richness except the EPT index is only the number of families from 3 orders (Ephemeroptera, Plecoptera, and Trichoptera).

ecological background: The majority of families grouped in Ephemeroptera, Plecoptera, and Trichoptera are considered to be very sensitive to pollution. Therefore, as water quality decreases, the number of the families in these orders also decreases.

Scraper/Filterer Ratio: *measure of:* dominant food type
interpretation: larger ratio = higher water quality

description: This metric is based on the number of individuals present in 2 of the 5 functional feeding groups (FFG) - scrapers and filterers. The ratio of scrapers to filterers indicates the most available food source in an aquatic system. **Note:** If collector/filterers are absent from the subsample, the ratio will have a zero as the denominator. If this occurs, make a note of it in the appropriate box on the data sheet and do not use this metric in the bioassessment procedure. If scrapers are absent from the subsample there will be a zero in the numerator, and the resulting metric value will equal zero. Enter zero on the bioassessment sheet for this metric.

ecological background: Functional feeding groups (FFG) are an excellent window into a stream ecosystem because: first, each FFG is made up of members from many different taxonomic groups and second, the number of individuals belonging to the dominant FFG reflects the most prevalent food type.

Scrapers feed primarily on periphyton and filterers primarily on FPOM. High quality streams tend to have a diverse periphyton community comprised of microscopic algae and diatoms. Low quality streams tend to have a limited periphyton community comprised mainly of filamentous algae. Additionally, lower quality streams tend to have ample FPOM due to organic enrichment. Therefore the scraper/filterer ratio is used to determine water quality in the following way. A high ratio indicates a predominance of scrapers, a healthy periphyton community, and therefore high water quality; a low ratio indicates predominance by filterers, ample FPOM, and therefore low water quality.

EPT/Chironomidae Ratio: *measure of*: community balance
interpretation: larger ratio = higher water quality (see note #2)

description: This metric relates the number of individuals from the 3 pollution sensitive orders (E,P, & T) to the number of individuals of a pollution tolerant family (chironomidae). The ratio of EPT individuals to chironomidae individuals indicates which group is dominant, pollution sensitive or pollution tolerant. **Note #1:** If the family Chironomidae is absent from the sample, the ratio will have a zero as the denominator. If this occurs, make a note of it in the appropriate box on the data sheet and do not use this metric in the bioassessment procedure. If the EPT value equals zero, there will be a zero in the numerator, and the resulting metric value will equal zero on the bioassessment sheet.

ecological background: High quality streams tend to support members from all 4 taxa but have much higher numbers of individuals belonging to E,P and T. In streams with low water quality the abundance of E,P and T individuals tends to decrease while the number of chironomidae may increase. Additionally, a sample dominated by chironomidae and a very low EPT index may indicate contamination from heavy metals. **Note #2:** This metric is prone to underestimating the level of water quality impacts. Since the order Trichoptera includes the family Hydropsychidae, which is very abundant in organically enriched systems, a large ratio may lead to erroneous conclusions. If the ratio is large check to see whether or not Hydropsychidae is the dominant family. If it is, the metric may not be indicative of high water quality.

Percent Contribution by the Dominant Family:
measure of: variety of environmental conditions
interpretation: varies with the family

description: This metric is the percentage of the sample comprised of the family with the greatest number of individuals. A considerable amount of information can be gained by knowing the tolerance, the functional feeding group, and life history traits of the macroinvertebrate family that dominates a site.

ecological background: A particular family of aquatic insect tends to be very abundant whenever the environmental conditions are favorable for that family. Water quality conclusions can be drawn based on the specific set of conditions (*e.g.*, food preferences, temperature, dissolved oxygen) optimal for the dominant family. For example, if the filter-feeding Hydropsychidae (tolerance =4) is the dominant family, fine particulate matter is abundant. This can indicate organic enrichment and lower water quality. If the shredder family Peltoperlidae (tolerance =0) is dominant, then water quality must be very high with low nutrient input. Additionally, stressed communities tend to be represented by high abundance of a single organism, while non-impacted communities have an even distribution of many types of organisms.

Hilsenhoff Biotic Index (Modified): *measures:* potential for organic pollution
interpretation: lower number = higher water quality

description: This metric indicates the potential for organic pollution influences in a stream as a weighted mean of the pollution tolerances of all individuals in the sample. A lower HBI indicates low organic enrichment and higher water quality. As the HBI increases so does the level of organic enrichment. The index is said to be *modified* because tolerance values are assigned at the family level, not at the genus and species level.

ecological background: The HBI was developed based on data from 2,000 Wisconsin streams. It assumes each family of riffle dwelling invertebrates has a unique tolerance to organic pollution (0=least tolerant to 10=most tolerant). As the amount of organic pollution increases, many of the biological and chemical reactions change resulting in a reduction in the number of low tolerant families.

Community Balance: *measure of:* community balance
interpretation: even distribution = high water quality

description: This metric is not officially used in the bioassessment statement calculations but is related to the percent contribution by the dominant family. Community balance provides important information about the distribution of the benthic community. In streams with high water quality, all major taxa are fairly evenly represented. If one or two families, such as the pollution-tolerant Hydropsychidae or Chironomidae, comprise a large percentage of the macroinvertebrate community, water quality is generally lower.

ecological background: In general the more diverse a community is the more stable the environment surround that community. In this situation a diverse benthic community indicates a stable aquatic environment with a variety of food resources and little fluctuation in water chemistry due to little or no organic inputs. An optimal community structure for New York is shown in Figure 8.18. The community is comprised of 40% Ephemeroptera, 5% Plecoptera, 10% Trichoptera, 20% Diptera (mostly chironomidae), 10% Coleoptera, 5% worms, and 10% other arthropods (Bode 1991).

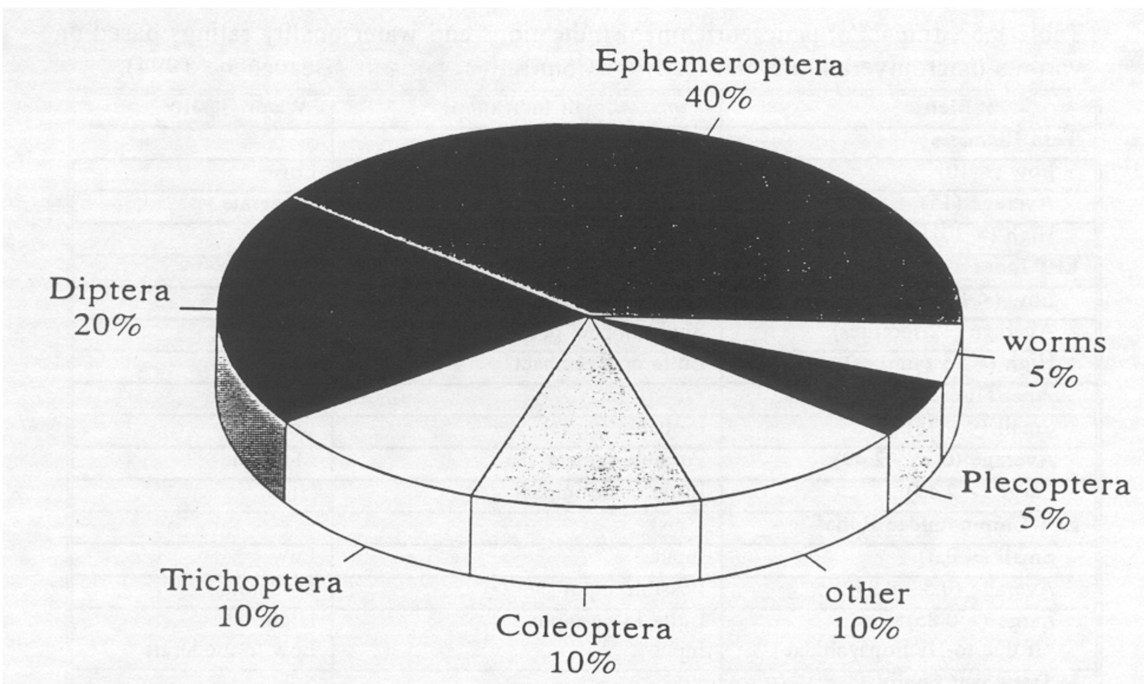

Figure 8.18. The distribution of the riffle dwelling macroinvertebrate community in a high quality stream (Bode 1991).

Table 8.5. Impact/organic enrichment indications and water quality ratings based on various macroinvertebrate metrics from Connecticut streams (Beauchene 1994).

Metric	Impact/Input Indications	Water Quality
Taxa Richness		
Low (< 10)	Impacted area	Low
Average (15)	Possible impact	Moderate
High (> 20)	No impact	High
EPT Index		
Low (< 10 families)	Impacted area	Low
Average (12 families)	Slight impact possible	Moderate
High (> 15 families)	Little or no impact	High
Scraper/Filterer Ratio		
Small (< 0.75)	Probable impact	Low
Average (0.75 - 2.49)	Possible impact	Moderate
Large (> 2.49)	Little or no impact	High
EPT/Chironomidae Ratio[1]		
Small (< 0.5)	Input	Low
Average (0.5 - 0.85)	Possible input	Moderate
Large (> 0.85)* *If due to Hydropsychidae	Little to no input Input	High Low to moderate
% Dominant family		
Large (> 70% of sample)	Impacted area	Low
Medium(40-69% of sample)	Potential impact	Moderate
Small (< / = 39% of sample)	Little or no impact	High
Hilsenhoff Biotic Index**		
0.00 - 3.75	Input unlikely	Excellent
3.76 - 4.25	Slight input possible	Very good
4.26 - 5.00	Some input probable	Good
5.01 - 5.75	Substantial input probable	Fair
5.76 - 6.50	Substantial input likely	Fairly poor
6.51 - 7.25	Very substantial input	Poor
7.26 - 10.00	Severe input	Very poor

[1]The most common discrepancy is when a stream contains predominantly Hydropsychidae and very little else. In this situation, all of the metrics indicate moderate water quality influence except the EPT:chironomidae ratio. Since Hydropsychidae is a family belonging to T (trichoptera) and the ratio is determined by totaling the number of individuals in each group and dividing by the number of chironomidae. The metric results will indicate excellent water quality. However, through evaluating the other metrics and preforming the bioassessment comparison a more accurate representation of the water quality results.

** ranges and values from Hilsenhoff 1987.

Metric limitations/contradictions:

Each metric has limitations due to the identification of these organisms to only the family level. Most macroinvertebrate families contain several genera. The pollution tolerance of a family is determined by averaging the tolerances of all the genera. For example if family X contains 2 genera, one with a tolerance of 10 and the other with a tolerance of 0, the family tolerance equals 5. As a result some metrics may potentially overestimate water quality impacts at high quality streams and underestimate water quality at low quality streams. Therefore if there is a discrepancy in any metric it is important to determine if the result is an artifact of the level of identification.

When assessing water quality using macroinvertebrate metrics, contradictions can sometimes occur. If this happens, do not assume that an error was made and discard the metric. Instead, look at the dominant family and its percent contribution. This information may tell you if one family is biasing a calculation. Keep all data and attempt to explain any contradictions. Remember, a zero in the numerator of a ratio will equal zero, while the metric is not used if there is a zero in the denominator of a ratio.

Bioassessment procedures

The bioassessment statement is like a jigsaw puzzle with each metric representing an individual piece. Even though most of the pieces of a puzzle are roughly the same size, some provide more information than others. For example if 2 sides of the piece are straight, then one can conclude that it is a corner piece. Once the piece is identified as a corner, then the print on the surface of the piece provides additional information as to which corner it is. The remainder of the puzzle pieces are then analyzed and put together to complete the puzzle. Regardless of the amount of information contained from any one piece, the picture is not complete until all of the pieces have been used.

To assess water quality the metrics from the benthic community of a study stream are usually compared to those of a reference stream. A ***reference stream*** is an area selected to represent the best attainable situation for a particular geographic area. Reference streams generally have very little human influence on the water quality. They usually are found in drainage basins with a high percentage of forest, little agriculture, industry, or human settlement. The benthic community of the reference stream then reflects the highest water quality. The completed bioassessment results in 1 of 4 statements about the condition of the biological community (Table 8.6). Any differences between the two benthic communities may be a result of water quality differences.

Table 8.6. The condition and attributes of a benthic community based on the percent similarity of one data set to another based on bioassessment procedures.

% Comparison to Reference	**Condition**	**Attributes**
Greater than 83%	Nonimpaired	Comparable to the best situation to be expected in the major basin. Optimum community structure and balanced trophic assemblage for that stream size and habitat.
54-79%	Slightly impaired	Community structure less than expected. Loss of some of the intolerant families. Percent of tolerant forms increasing.
21-50%	Moderately impaired	Fewer families due to a loss of most of the intolerant families. Reduction of the EPT index.
Less than 21%	Severely impaired	Few families present. If there are high densities of organisms it is by a few tolerant families.

Potential study designs:

Bioassessment studies can be set up to evaluate a variety of different water quality questions. The most common study design is an initial water quality assessment at a previously unsampled site. Other designs include, upstream and downstream of a suspected input, comparison of 2 different sites, and comparing 2 different dates at the same site. The following are brief descriptions about each type of experimental design.

Initial water quality surveys: The main purpose is to assess water quality of a site which has not been studied. It involves comparing the metrics of the unknown water quality to the metrics of a reference site (Table 8.7). The bioassessment statement indicates the level of biological impairment as compared to an optimal situation. **Note:** It is assumed that the study site will have lower water quality than the reference site. This may not be the case in all situations. It is very possible the study site can rank "better" than the best (reference site). This situation can be identified if the final bioassessment percentage for the study site is greater than 100%. If this should occur, switch the metrics on the bioassessment sheet so to compare the reference site to the study site.

Upstream/downstream of a potential pollution source: The purpose of this study design is to compare the benthic community upstream (above the source) and downstream (below the source). The bioassessment statement indicates the level of influence from the suspected source. If the comparison indicates that the downstream site is nonimpaired then the source has no effect on water quality. If the comparison indicates that the downstream site is impaired at some level, then further investigation is required to confirm the water quality impacts. The upstream site should be compared to a reference site to first document water quality above the suspected influence. The upstream site can then be used as the reference site for the downstream site since it is located above any potential influence.

Comparison of 2 different sites: The purpose of this study design is to compare water quality of two different sites. The sites may be tributaries entering along a main stem, several sites along the main stem, streams within the same major basin, *etc.* The bioassessment statement indicates the degree of similarity between sites. **Note:** The results of this study design must be interpreted carefully. Since it is not necessary to use a reference site in the comparison, the bioassessment statement may not reflect water quality. For example, two study sites are chosen below two separate sewage treatment plants. Samples from both sites consist completely of Hydropsychidae, which would normally indicate severe impact. However, bioassessment calculations conclude no impact because the comparison is between the biotic integrity of the two streams. If two streams have similar community structure, bioassessment procedures will conclude no impact.

Comparison of 2 different dates at the same site location: The purpose it to see if the biological community changes between sampling events or between seasons. The bioassessment statement indicates the degree of similarity between events or seasons. ***Note:*** see note in comparison of 2 different sites for cautions in data analysis.

In summary, bioassessment calculation procedures compare the metrics from at least two samples to formulate a blanket statement about water quality. The two streams being compared will vary depending upon experimental design. Regardless of the experimental design, each data set should be compared to a reference stream first to determine water quality. After this initial comparison is complete, then secondary evaluations such as upstream/downstream, 2 different sites, and seasonal differences can be performed. Remember unless a study site is compared to a reference stream, water quality can not be accurate determined.

When performing any bioassessment please note two special cases: First, if the metrics for the study site are consistently higher than for the reference site the study site is more diverse than the site chosen to represent "the best". Second, when comparing two streams and neither is a state reference stream use the stream whose metrics indicate higher water quality as the reference, and continue as normal. If the streams are at least 83% comparable, then their biotic integrity is very similar. This does not necessarily mean that the streams have excellent water quality, only that the communities are very similar to each other.

Table 8.7. Reference stream metric values to be used in comparisons of (A) fall data and (B) spring data.

(A) Fall data

metric	Shepaug R.	Salmon R.	Natchaug R.	Eightmile R.	Saugatuck R
taxa richness	19	17	18	13	13
EPT index	13	12	12	7	10
Scraper : Coll. Filterer ratio	1.68	1.6	1.18	1.09	0.22
EPT : Chironomidae ratio	5.5	37.5	undefined	39	undefined
Contribution of dominant family	17 %	31 %	24 %	31 %	37 %
HBI(modified)	3.03	2.44	3.10	3.20	3.08

(B) Spring data

metric	Shepaug R.	Salmon R.	Natchaug R.	Eightmile R.	Saugatuck R
taxa richness	14	24	21	12	14
EPT index	9	16	13	7	8
Scraper : Coll. filterer ratio	1.16	2.46	2.13	0.9	1.86
EPT : Chironomidae ratio	3.03	15.4	33	6.61	5.13
Contribution of dominant family	43 %	19 %	16 %	45 %	32 %
HBI(modified)	3.15	3.39	3.34	2.75	3.25

Summary of the major components of a bioassessment program:

1.) Initial office work:

-Determine purpose of the study: can be gathering initial baseline information, comparing current conditions to historical data, upstream/downstream of potential source

-Set up sampling schedule, organizing equipment, and selecting study groups.

2.) Initial field work:

-*Determining an appropriate study site by*: using a map, making field visits, and performing a habitat assessment (See: **Activity 4A**: *Site Selection,* for details).

3.) Field work:

-*Collection of organisms:* involves kick sampling, field sorting of debris, and sample preservation (See: **Activity 8A**: *Collecting Macroinvertebrates*, for details).

4.) Laboratory work:

-*Subsampling:* laboratory screening of sample to obtain a 100 organism (minimum) subsample. (See: **Activity 8B**: *Subsampling Lab*, for details).

-*Identification:* use of keys to determine the taxonomic composition of the sample. (See **Activity 8C**: *Identification Lab*, for details).

5.) Office work:

-*Calculation of the metrics:* several indices and ratios used to transform the biological data to a numerical format. (See: **Activity 8D & E**: *Metric and Bioassessment Calculations*, for details)

-*Bioassessment comparison:* comparing the metrics of the study site to a second site. The second site will vary depending upon the purpose of the study. For example, if the purpose of the study is to assess water quality, then a reference site is used. If the purpose is to compare to historical data, then the historical data is used. (See: **Activity 8D & E**: *Metric and Bioassessment Calculations,* for details).

-*Analysis and reporting of data:* interpreting results in relation to the purpose of the study. Several components include evaluating each metric, making a bioassessment conclusion, explanation of potential sources of error, evaluating site conditions, generating graphs and tables, comparing and integrating invert data with chemical data and writing text (See Chapter 10 for details).