Bioindicators of Strawberry Creek

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Students: Bring a calculator with you to lab!

Goals

The goal of this lab is to introduce you to the following:

- biological organisms as indicators of ecosystem health
- use of identification keys
- calculation of taxa diversity and other biological measures
- use of statistical tests to determine if two assemblages of organisms are different from each other.

Introduction

Biological organisms are commonly used to indicate ecosystem health. In streams, aquatic macroinvertebrates are often used by the E.P.A. and most foreign regulatory agencies to assess biological condition because these organisms are diverse, easy to sample, and have a wide range of tolerance to pollution. In addition, aquatic organisms can give a long-term view of stream conditions because of their long life cycles, while chemical and physical measurements only reflect the conditions when samples or measurements are taken.

In streams, biological condition is strongly influenced by water chemistry and habitat quality. Low dissolved oxygen, high bacterial, nitrate or phosphorous concentrations, and low pH can cause reduced water quality. Good habitat quality is generally characterized by a heterogeneous habitat with both slow and fast moving water, woody debris, substrate variety, and well-vegetated, stable banks. Impairment of habitat and/or water chemistry can lead to reduced diversity of aquatic macroinvertebrates.

Biological Measures

1. Taxa Richness

Richness measures represent the diversity within a sample. Taxa richness is calculated by counting the total number of taxa within a sample. A higher richness implies higher habitat diversity and better water quality conditions.

2. % EPT

The insect orders Ephemeroptera, Plecoptera, and Trichoptera (EPT) are often found in clean, pollution-free streams, because the majority of these taxa are very sensitive to poor water quality. This metric is calculated as the % of the individuals in the sample which belong to the aquatic insect orders Ephemeroptera, Plecoptera, & Trichoptera.

$$\text{\% EPT Abundance} = \frac{\text{Total \# of EPT individuals}}{\text{Total \# of individuals in the entire sample}}$$

3. Biotic Index

The Biotic Index is a measure that is representative of the tolerance of organisms to pollution. Tolerance values on a 0 (sensitive to pollution) to 10 (tolerance of pollution) scale are assigned to invertebrate taxa. The biotic index you use in this lab is based on the Family Biotic Index (FBI) developed by the U.S. Environmental Protection Agency, which was originally developed to detect the effects of organic pollution. The FBI is the average tolerance for the community, weighted by the number of individuals in each family. FBI is calculated by the following formula:

$$BI = \sum_{i=1}^{z} \frac{x_i * t_i}{n}$$

where, z = number of taxa (family) in the sample

x_i = number of individuals in a taxon

t_i = tolerance value of a taxon

n = number of organisms in the sample

This looks like:

$$= (x_1 * t_1)/n + (x_2 * t_2)/n + (x_3 * t_3)/n \ldots + (x_z * t_z)/n$$

where, 1= taxa1, 2=taxa2, 3=taxa3, …

Lower FBI values imply better water quality:

Family Biotic Index	***Water Quality***	***Degree of Organic Pollution***
0.00-3.75	Excellent	Organic pollution unlikely
3.76-4.25	Very Good	Possible slight organic pollution
4.26-5.00	Good	Some organic pollution probable
5.01-5.75	Fair	Fairly substantial pollution likely
5.76-6.50	Fairly Poor	Substantial pollution likely
6.51-7.25	Poor	Very substantial pollution likely
7.26-10.00	Very Poor	Severe organic pollution likely

Functional Feeding Groups

Feeding measures or trophic dynamics provide information on the balance of feeding strategies (food acquisition and morphology) in the benthic invertebrate assemblages.

Each of the following metrics is calculated by:

$$\%\ \text{FFG} = \frac{\text{Total number of individuals of the functional feeding group}}{\text{Total number of individuals in the sample}}$$

4. % Filterers: Filtering organisms use their body (in the case of the blackfly Simuliidae) or construct nets (in the case of the caddisfly family Hydropsychidae) to filter particles of organic matter out of the stream water. The predominance of filterers may reflect an unbalanced community response to an overabundance of a particular food source.

5. % Predators: Predators represent a higher trophic level than filterers and grazers, and thus play an important ecological role. Predators also tend to be larger and longer-lived organisms; thus, they are more susceptible to short-term or infrequent disturbances.

Using Dichotomous Keys

Dichotomous keys are a frequently used method for identifying plants and animals that are constructed of organized statements called **couplets**. A couplet consists of two descriptions (usually labeled “a.” and “b.”) that represent mutually exclusive choices. Beginning at the first couplet of the dichotomous key, read both options and decide which description matches the organism you are trying to identify. At the end of the descriptions are numbers that indicate the next couplet to advance to. This process is repeated until an identification is made. At this point a verification step is important: compare the specimen with any details in the description and/or any available figures. If the description seems satisfactory, a correct identification probably has been achieved. If the description is not satisfactory in one or more important particulars, back up to some earlier couplet and start over, questioning each decision more carefully.

Assignment

You will use aquatic macroinvertebrates that live in streams to assess water quality of the North and South Forks of Strawberry Creek. You will determine whether the invertebrate community from each fork contains relatively pollution sensitive organisms or if it is dominated by pollution tolerant organisms. Because of the impact of sampling on the creek by a large class such as ours, insect samples were already taken and are stored in ethanol for preservation. You will assume that South Fork samples are taken from Strawberry Canyon (east of the football stadium) and that North Fork samples are taken from the North Gate area of campus. Look at the map to help you hypothesize about potential causes of differences in water quality and biological condition between the two forks.

Using a t-test (see page 13), you will determine if the measures describing aquatic insects found in the two forks of Strawberry Creek are significantly different. Based on your results, you should propose a null hypothesis and an alternate hypothesis to test these differences. You will then propose a field and laboratory experiment to test your hypotheses.

Methods for Biological Assessment

1. Each group should identify 50 organisms from both the North Fork and South Fork of Strawberry Creek. Each flask contains 5 organisms; thus, your group will need to examine 10 flasks from each fork. Identify all of the organisms in a flask before beginning the next flask. In order to ensure there are enough flasks available, each group should only take 5 flasks at any time.

2. Use a dissecting microscope in order to examine the organisms. Some of the features are hard to see with your naked eye.

3. Select one organism to identify using the dichotomous key. At the beginning of the key (couplet 1), decide which of the statements (a. or b.) matches the organism you are looking at. At the end of the descriptions are numbers that indicate the next couplet to advance to. Repeat this process until an identification is made. Record the name of the taxa on the appropriate worksheet (North Fork or South Fork).

4. As you identify more invertebrates, keep a tally of the number of organisms of each taxa you find on the data sheet.

5. Once you have identified 50 organisms, calculate the biological metrics as described above. When you are done, write your results for all five metrics on the chalkboard.

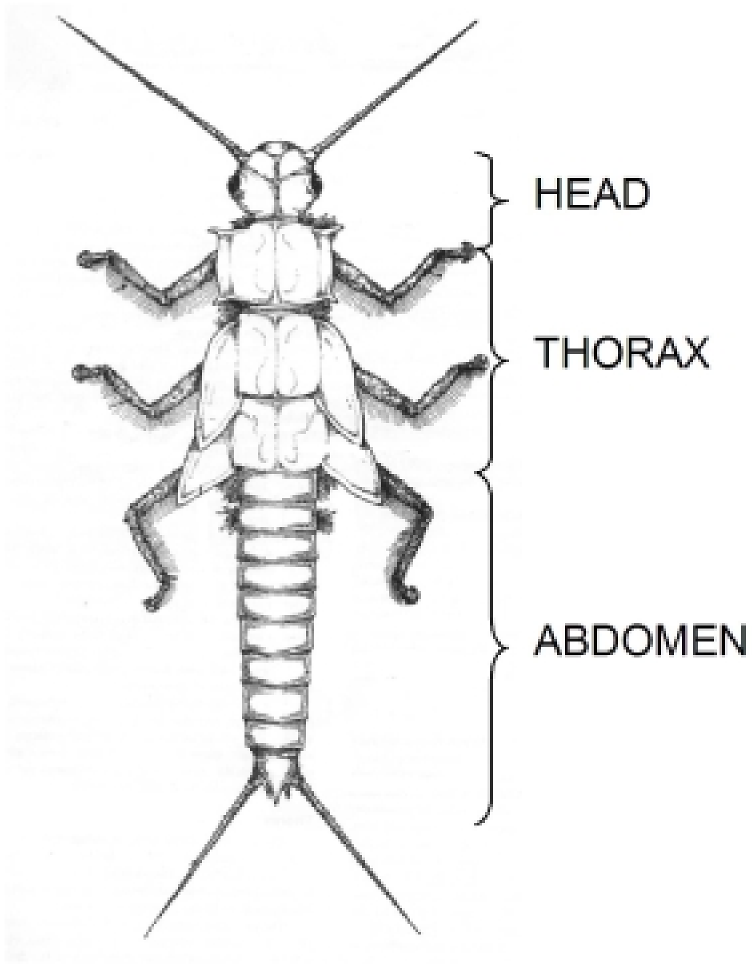
6. Copy down the values from the other groups in your lab.

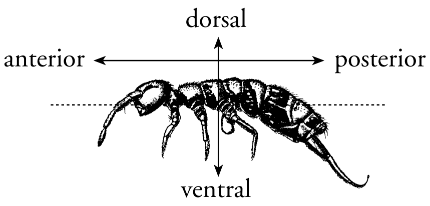
7. Do a t-test to determine if there is a significant difference in each of the metrics between the North Fork and South Fork. Each group's value represents one sample.

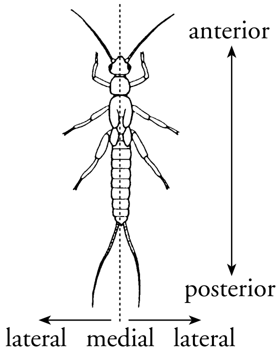
Common Insect Terms

head capsule - a clearly differentiated and hardened head segment
prolegs - little nubbed legs, without joints (like on a caterpillar).
labium - lower lip, as seen from underside of head.
caudal filament - long, slender tails of end of abdomen.
gills - can be small and in bunches or longer and plate-like.
tarsal claws - claws on very end of legs.

Insect Morphology







Strawberry Creek on the U. C. Berkeley Campus

▬ Natural Stream Channel

- - - Underground (Culverted) Stream

Taxonomy, Biotic Index Scores, and Functional Feeding Groups of Invertebrates in Strawberry Creek

(biotic scores and functional feeding groups given in parentheses)

Phylum Annelida
    Class Oligochaeta – aquatic earthworms (8, C)

Phylum Arthropoda
    Class Crustacea
        Order Amphipoda – scuds (4, C)

    Class Insecta
        Order Odonata
            Suborder Anisoptera - dragonflies (3, P)
        Order Ephemeroptera – mayflies
            Family Ephemerellidae (1, C)
        Order Plecoptera – stoneflies
            Family Perlidae (1, P)
        Order Megaloptera
            Family Corydalidae – hellgrammites (0, P)
        Order Trichoptera – caddisflies
            Family Hydropsychidae (4, F)
        Order Diptera
            Family Simuliidae – black flies (6, F)
            Family Tipulidae – crane flies (3, S)
        Order Coleoptera
            Family Psephenidae – water pennies (4, G)

Phylum Mollusca
    Class Gastropoda – snails (7, G)

    Class Pelecypoda – clams (8, F)

Key to the Functional Feeding Groups
C = Collector
F = Filterer
G = Grazer
S = Shredder
P = Predator

Key to the Benthic Invertebrates of Strawberry Creek

1. a. With a hard calcerous shell ………………………………………………………….……….. 2
b. Without a shell ……………………………………………………………..……..……. 3

2. a. Shell spiral-shaped ………………………………….……………… Mollusca-Gastropoda
b. Shell with two halves ……………………………….……………… Mollusca-Pelecypoda

3. a. Thorax with more than 6 jointed legs, body laterally compressed……. Crustacea- Amphipoda
b. Thorax with 6 or fewer legs …………………………………………………………………4

4. a. 6 jointed legs and distinct head, thorax and abdomen …………………………………… 5
b. Fewer than 6 legs ………………………………………………………………………… 10

5. a. Abdomen with 2 or 3 tail-like projections ………….…………..……………………….. 6
b. Abdomen without a tail or with 1 single projection ……………..…………………… 7

6. a. Abdomen with 2 tail-like projections, thorax with gill
tufts between the bases of the legs (Fig.1) ……………...………..…... Plecoptera-Perlidae
b. Abdomen with 3 tail-like projections, paddle-like gills
on abdominal segments (Fig.2) …...……………………… Ephemeroptera-Ephemerellidae

7. a. Body disk-shaped with hardened plates covering the
head and legs ……..……………………………………….. Coleoptera- Psephenidae
b. Body not disk-shaped ……………………………………………………………………. 8

8. a. Abdomen lacks hardened plates, gill tufts on ventral
abdominal segments (Fig.3) ……………………..…………. Trichoptera-Hydropsychidae
b. Abdomen with hardened plates …………………………………………..………... 9

9. a. Abdominal segments with 1 long lateral filament
on each side (Fig.4) ……………..…………………………….. Megaloptera- Corydalidae
b. Lacks abdominal filaments, body thick and stout ……….……………..... Odonata-Anisoptera

10. a. Body with a distinct head, fan-like feeding structures
(Fig.5), bulbous posterior abdomen …….…………………………... Diptera-Simuliidae
b. Body with indistinct or retracted head ………………………………………………….. 11

11. a. Maggot-like body with fleshy protrusions at end of abdomen (Fig.6) …... Diptera- Tipulidae
b. Worm-like body with many segments …………………..….............. Annelida-Oligochaeta

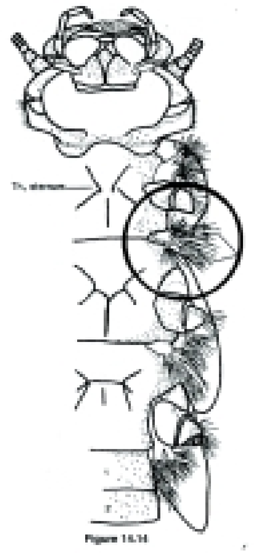


Fig.1. Gill tufts between the legs (ventral view)

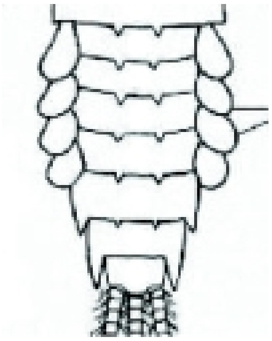


Fig.2. Paddle-like gills on abdominal segments (dorsal view)

Fig.3. Gill tufts on ventral abdominal segments (lateral view)

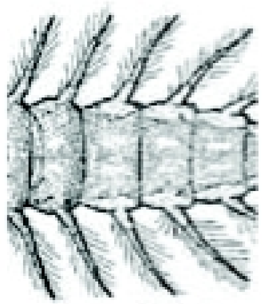


Fig.4. Lateral filaments on abdomen (dorsal view)

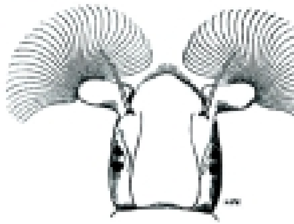


Fig.5. Fan-like feeding structures on head capsule (dorsal view)

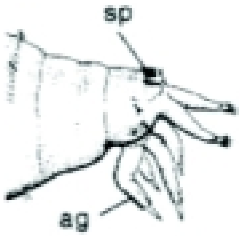


Fig.6. Fleshy protrusions at end of abdomen (lateral view)

North Fork Strawberry Creek Data Sheet

	A	**B**	**C**		**D**		**E**
	Taxa Name	**Functional Feeding Grp.**	**Biotic Score**		**# of Orgs.**		**Total**
1.				X		=	
2.				X		=	
3.				X		=	
4.				X		=	
5.				X		=	
6.				X		=	
7.				X		=	
8.				X		=	
9.				X		=	
10.				X		=	
Total:							

Metric Scores

1. Taxa Richness = Total number of taxa present = __________

2. % EPT = Total number of taxa in EPT divided by total of column D = ____________

3. Biotic Index = Total of column E divided by total of column D = __________

4. % Filterers = Total number of filterers divided by total of column D = __________

5. % Predators = Total number of predators divided by total of column D = __________

South Fork Strawberry Creek Data Sheet

	A	B	C		D		E
	Taxa Name	Functional Feeding Grp.	Biotic Score		# of Orgs.		Total
1.	____________	________	_____	X	________	=	______
2.	____________	________	_____	X	________	=	______
3.	____________	________	_____	X	________	=	______
4.	____________	________	_____	X	________	=	______
5.	____________	________	_____	X	________	=	______
6.	____________	________	_____	X	________	=	______
7.	____________	________	_____	X	________	=	______
8.	____________	________	_____	X	________	=	______
9.	____________	________	_____	X	________	=	______
10.	____________	________	_____	X	________	=	______
Total:			________		______		

Metric Scores

1. Taxa Richness = Total number of taxa present = _________

2. % EPT = Total number of taxa in EPT divided by total of column D = ___________

3. Biotic Index = Total of column E divided by total of column D = _________

4. % Filterers = Total number of filterers divided by total of column D = _________

5. % Predators = Total number of predators divided by total of column D = _________

How do I do a t-test?

We do a t-test to determine if there is a significant difference between groups or if the difference is due to chance.

The basic idea is to test the null hypothesis, H_0, which states that two sample means are equal.

You will need to calculate the following from your data:

n_1 = number of FBI scores in sample 1 (North Fork)
n_2 = number of FBI scores in sample 2 (South Fork)
$N = n_1 + n_2$
$\bar{X}_1$ =mean of sample 1
$\bar{X}_2$ =mean of sample 2
SS_1 = sum of squares for sample 1
SS_2 = sum of squares for sample 2

To calculate the sum of squares: first, subtract the mean from each of the individual scores in a sample; then, square the differences and add them together. This is the sum of squares (SS).

For example: North Fork

FBI Score	**Mean FBI Score**	**(Score – Mean)**	**Square of Difference**
3.5	3.64	-0.14	0.0196
4.0	3.64	0.36	0.1296
3.0	3.64	-0.64	0.4096
3.5	3.64	-0.14	0.0196
4.2	3.64	0.56	0.3136
Mean$_1$ = 3.64			**SS$_1$** = 0.8920

Next, calculate the t-statistic:

$$t = \frac{\left| \bar{X}_1 - \bar{X}_2 \right|}{\sqrt{S^2_p \left(1/n_1 + 1/n_2\right)}} \qquad S^2_p = \frac{SS_1 + SS_2}{(n_1 + n_2) - 2}$$

Look up the critical t in a statistical table (provided below) to see if you can reject H_0. If your $t > t_{critical}$ then the probability that H_0 is true is <0.05 (very small) and the two samples are significantly different. The “N” value in the table below has already been adjusted for the degrees of freedom.

N	$\mathbf{t_{critical}}$
5	2.353
6	2.132
7	2.015
8	1.943
9	1.895
10	1.860
11	1.833
12	1.812
13	1.796
14	1.782
15	1.771
16	1.761
17	1.753
18	1.746
19	1.740
20	1.734

Questions

1. Which metrics were significantly different, using the t-test, between the North Fork and South Fork? Do these differences suggest that water quality and/or habitat conditions are different between the two sites?

2. Why might it be beneficial to use multiple metrics in a biomonitoring program?

3. Design a new field study to assess whether differences between the two sites are due to water quality conditions (e.g. pollutants) or habitat differences. If you could sample from additional sites, how would you try to assess differences in benthic macroinvertebrate communities?

4. The samples in this lab were collected in April. How do you think the invertebrate community would change if you sampled at different times of year?

5. Many stream insects have aquatic larval stages and terrestrial adult stages. Hypothesize some potential life cycles for these insects, given what you know about ecological conditions in different seasons.