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Life Science / Biology, Environmental Science, Marine / Ocean
Science Grades: 6-8**

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MARINE PARASITES AND FISH: HOW TO SAMPLE AND ANALYZE

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Grade Level
Middle School

Subject area
Life Science, Biology, or Environmental Science

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1. **Activity Title:** Marine Parasites and Fish: How to Sample and Analyze
2. **Focus:** Marine Fish Parasites: Importance, Sampling, and Analysis
3. **Grade Levels/Subject:** 7th grade Life Science
4. **VA Science Standard(s) addressed:**
 - a. LS.8: interactions among populations in a biological community, symbiotic relationship between parasites and hosts
 - b. LS.10: ecosystems, communities, populations, and organisms are dynamic, change over time, and respond to daily, seasonal, and long-term changes in their environment, factors that increase or decrease population size
 - c. LS.11: relationships between ecosystem dynamics and human activity, population disturbances and factors that threaten or enhance species survival
5. **Learning objectives/outcomes:**
 - a. Students will explain what a parasite is and three ways they can affect their host.
 - b. Students will know three types of parasites found in marine fish.
 - c. Students will be able to calculate prevalence, intensity, and abundance.
6. **Total length of time required for lesson:** one class length (45-50min)
7. **Key words, vocabulary:**
 - a. Parasite—organism that lives in or on another organism (its host) and benefits by deriving nutrients at the host's expense
 - b. Prevalence—proportion or percent of individuals in a population having a disease or characteristic
 - c. Incidence—the number of new cases of a specific disease occurring during a certain period in a population
 - d. Mean Intensity—average number of parasites per infected fish
 - e. Mean abundance—average number of parasites in all animals (includes uninfected fish)
 - f. Disease—impairment in the normal state of an organism that interrupts or modifies the performance of the vital functions and is typically manifested by distinguishing signs and symptoms
8. **Background information:**

Parasites are a common component in the marine environment. When the environment is in equilibrium, the hosts and parasites are able to coexist without one destroying the other. But when the environment becomes unbalanced due to changes in temperature, oxygen levels, fishing pressure, pollution, ocean acidification, the fish hosts are no longer able to tolerate the parasites, and disease can result. Diseases are evident through the detection of symptoms such as lesions, discoloration, changes in behavior, weight loss, lethargy, and death. Infections from parasites and the resulting symptoms can have important effects on a fish population such as increased predation on infected fish, increase catches of infected fish, reduced ability to tolerate environmental stressors, reduced value of infected fish, and the potential to make consumers sick. Therefore identifying, studying, and managing marine diseases is extremely important to keeping our fish healthy and economically valuable.

Some examples of common marine parasites are copepods, isopods, monogeneans, digeneans, nematodes, cestodes, and acanthocephalans. Viruses and bacteria are also considered to be parasites. See attached PowerPoint for examples of marine parasites.

The first step in studying a marine parasite is first identifying it and then usually some sort of survey that looks at the extent of infection in a population. This work usually entails going out into the habitats of the fish of interest and collecting the fish. These specimens are brought back to a laboratory to determine if it is infected and/or diseased, as well as the quantity of parasites it harbors. The methods for determining infection and the quantity of parasites can be done using molecular work, visualization under a microscope, or with the naked eye. Usually, the higher the quantity of parasites, the worse condition the fish is in.

This activity simulates the surveying of a fish population and quantifying basic infection metrics (prevalence, intensity, mean abundance) using the example of American eels and a parasitic nematode (*Anguillicoloides crassus*). This parasite is originally from East Asia where it infects the Japanese eel, and was brought to the U.S. in the early 1990s through the eel aquaculture trade. It has rapidly spread throughout the American eel range, and now can be found from Canada into the Gulf of Mexico. Severe damage to one of the eel's organs, the swimbladder, occurs during infection. A swimbladder is an air filled organ, similar to a balloon, which fish use to control their buoyancy and help them swim. This parasite lives inside the swimbladder where it feeds on the eel's blood and can cause so much damage that the organ no longer functions properly. Despite this knowledge of this parasite, we do not know how it affects the American eel population as a whole. We do not know the full extent of infection, nor if it causes the eels to die. To get at these questions, scientists need to do a lot of sampling of eels in the wild, and this activity will guide students through the steps needed to sample a population and analyze the results they obtain.

9. **Student handouts and other materials needed (worksheets, data tables, diagrams, websites, etc.):** At bottom of lesson plan.

10. **Materials and supplies, A/V/Tech Support:**
 - a. Pipe cleaners
 - b. Beads
 - c. Buckets
 - d. Calculators (or students can do the math by hand)

11. **Classroom/lab/field study setup:** Either push a few desks together to make groups or use larger lab benches to split up groups.

12. **Procedure:**
 - a. Introduction—10 min
 - i. Explain what a parasite is and why parasites are important to study in the marine environment
 - ii. Show examples of some marine parasites (PowerPoint)
 - iii. Introduce the example for the activity—You've just gotten word that there's a nasty nematode parasite infecting American eels! But we don't know anything about this parasite. We don't know how many eels are infected and how many parasites there are per eel. And we don't know the differences between various areas of where eels live. You are all tasked with discovering the infection level in

American eels. In different groups, you must go to different sites and count the number of infected eels and number of parasites per eel and report back.

b. Activity—30 min

- i. Divide students into groups of four or five, each group is a “sampling site”. Have students write their site name on a sheet of paper to leave at the table or tape to the bucket.
- ii. Give each group a bucket, a big bag of beads (“nematodes”), 10 pipe cleaners (“eels”) for a total of 40-50 per group and as many recording sheets as rounds conducted.
- iii. Each group has a maximum number of parasites a fish can have (3-10 max). Teacher will assign these when groups are settled. This is to create groups with higher and lower intensities. Also groups can be assigned with how many of their eels can be infected (3 out of 10, 8 out of 10, etc.). This way you will have an idea of the expected prevalence and intensity should be and there will be variation among the groups. Tell each group individually so that the other groups don’t know before they “sample” that site.
- iv. Students get 7-10 minutes to “infect” their fish, i.e. put the beads on the pipe cleaners, making sure to leave some uninfected.
- v. Each student records on their sheet the percent of fish with parasites (prevalence), the average number of parasites in a sample (intensity), and the average number of parasites per infected fish (mean abundance).
- vi. Students put all their fish into the bucket and switch with another group.
- vii. **Alternatively, you can set this all up beforehand. I added the student involvement to cut down on prep time, but you may be able to get through more rounds if it’s already set up. If you choose to set it up beforehand, then start with step ix.**
- viii. Each student randomly (closes their eyes and sticks their hand in the bucket) chooses 7 fish and records prevalence, intensity, and mean abundance.
- ix. Students and groups total their amount.
- x. Return eels to bucket and switch buckets with another group or go to different “site”.
- xi. Repeat steps viii to ix one or two more times for a total of 3 rounds of sampling (i.e. 3 replicates).

13. **Assessment:**

Can be done as a whole group or small group discussion using think-pair-share. Or questions can be included in the activity worksheet and be passed in for grading. Suggested answers are below questions.

- a. What is a parasite?

A: Organism that lives in or on another organism (its host) and benefits by deriving nutrients at the host’s expense

- b. What are three ways a marine parasite can affect a fish?

A: Parasites can make their host sick (cause disease) or can kill it; they can change their host's behavior; they can become eaten by predators more than uninfected fish; they can have lower tolerance to environmental conditions such as temperature, oxygen level, and acidity; they can affect their ability to reproduce; they can be caught more by fishermen; they can decrease their ability to swim.

c. What are three types of marine parasites?

A: Copepods, isopods, monogeneans, digeneans, nematodes, cestodes, and acanthocephalans. Viruses and bacteria are also considered to be parasites.

d. Open ended questions:

i. Which group has the most infected population? How to define "the most infected" (which metric is most important)?

A: This question is meant to discuss the importance of the three infection metrics: prevalence, mean intensity, and mean abundance. Each metric tells us something different about the infected population. For example a population could have a high prevalence (i.e. many individuals could be infected) but have a low mean intensity (i.e. few parasites per individual). But depending on the severity of the infection. So in this example, there could be a big impact on the population if there doesn't need to be a high intensity to cause disease, but a small impact if there needs to be a higher number of parasites per fish before we see symptoms. This question shows the importance of using all three metrics to understand parasite infection levels.

ii. How did the individual students' samples compare to the groups totals? Why are these differences important to consider when studying a population?

A: You could have the students write their totals on the board to compare. All groups should have different answers. This result is supposed to demonstrate the variation in sampling, which is extremely important to consider when collecting data. Taking a sample multiple times from the same population does not always result in the same answers due to (as in this experiment) choosing different individuals or other factors such as time of day or year, temperature, salinity, etc. This result is also to show that a sample size of one does not necessarily capture the true population level status. Also this result gets at the reality of field sampling—you can't sample the whole population! So instead you must get smaller samples and infer what the true population is like. And if you collect many of these samples then you should get results that look more like the total population.

iii. What could cause the infection metrics to change over time?

A: Some reasons may be that more infected individuals are dying; leaving the less infected ones in the population. The individuals, through evolutionary time, could build up genetic resistance to the pathogen. More infected individuals could be eaten by predators or caught by fishermen. The parasite could become weaker due to changing environmental conditions so that it no longer can infect as well or cause as much disease. New parasites may enter a population and infect many individuals. A treatment could be developed to stop infection (most likely only in aquariums, hatcheries, or aquaculture).

14. References:

- a. <http://ocean.si.edu/blog/marine-parasites-crazy%E2%80%A6and-really-cool>
- b. <http://marinebio.org/oceans/symbionts-parasites/>
- c. <http://www.marineparasitology.com/MarineParasitology.htm>
- d. <http://parasite.org.au/para-site/introduction/introduction-essay.html>
- e. <https://parasiteecology.wordpress.com/>
- f. http://www.oxforddictionaries.com/us/definition/american_english/parasite
- g. <http://www.medicinenet.com/script/main/art.asp?articlekey=11697>
- h. <http://medical-dictionary.thefreedictionary.com/incidence>
- i. <http://parasite.org.au/para-site/introduction/introduction-essay.html>
- j. <http://www.merriam-webster.com/medical/disease>
- k. http://www.texasinvasives.org/pathogen_database/detail.php?symbol=4

Marine Parasites Activity

Sample site:

Individual Totals			Group Totals		
	Infected? (yes or no)	Number of Parasites		Total number infected	Total number of parasites
Eel #1			Student #1		
Eel #2			Student #2		
Eel #3			Student #3		
Eel #4			Student #4		
Eel #5			Student #5		
Eel #6			Total number of eels surveyed:	Grand total number infected:	Grand total number of parasites:
Eel #7					
	Total number infected:	Total number of parasites:			

$$Prevalence = \frac{\# \text{ of infected eels}}{\text{total \# of eels}} \times 100 =$$

Individual	Group
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$$Mean Intensity = \frac{\text{total number of parasites}}{\text{total \# of eels}} =$$

Individual	Group
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$$Mean abundance = \frac{\text{total number of parasites}}{\# \text{ of infected eels}} =$$

Individual	Group
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