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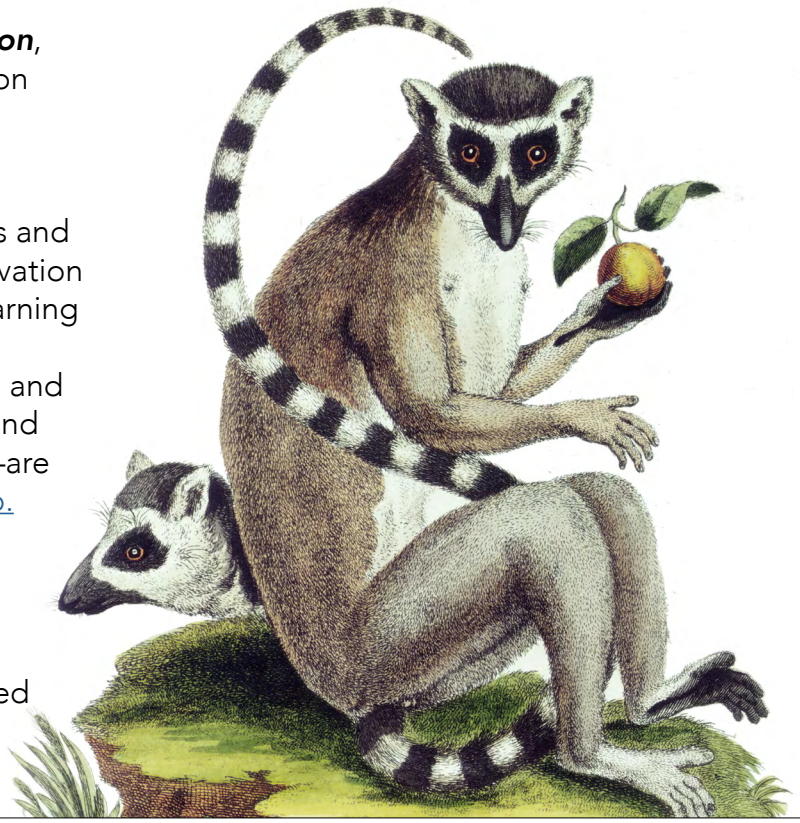

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Parasite Biodiversity: Community Data Analysis

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ABSTRACT

In this exercise, you will engage with real data to answer the question: how do human impacts on ecosystems change the abundance of parasites in wildlife? You will use data to assess whether human impacts on the environment increase or decrease the abundance of parasites in coral reef fish. Specifically, you will determine if fishing increases or decreases parasite abundance in coral reef fishes.

LEARNING OBJECTIVES

By the end of this exercise, students will be able to:
1. Calculate mean parasite abundance across six species of central Pacific reef fish.
2. Plot mean parasite abundance data and summarize patterns.
3. Discuss the results and how these results apply to similar and different ecosystems.

BACKGROUND

Coral reefs are among the most biodiverse ecosystems on the planet, and that biodiversity includes a large number of parasitic species that reside on and in reef-dwelling fishes. Many factors can influence which species of parasite are present in the community and the number of each species within each host fish, including characteristics of the host, parasites, and the wider coral reef environment. As humans modify the environment, particularly by fishing, we change which host species are present and how many individuals of each host species are present in a given area. These changes to the fish community structure alter the interactions between the host species, which can increase or decrease parasite transmission rates. By changing the overall environment in which the hosts and parasites live, humans may impact how long parasites can live outside their hosts, by providing extra nutrients in the water column or by altering the physical environment present for parasites as they search for hosts. Human influence may alter how receptive the hosts are to parasitic infection, as the fish left behind after fishing may be more or less able to defend themselves from infection. As a result of these changes, we expect human influence to shape the community of parasites present. In this exercise, you will use real data from coral reef fishes to determine how fishing pressure around each island can change the parasite community and you will start to examine potential reasons why this might happen.

The data you will use is from a study conducted in the northern part of the Line Islands archipelago (Figure 1). The Line Islands are a group of eleven total islands formed by volcanic activity in the Pacific Ocean, and they get their name from the fact that they sit on the International Date Line. The northern part of the Line Islands includes three unfished islands (Kingman, Palmyra, and Jarvis) and three fished islands (Teraina, Tabuaeran, and Kiritimati). The unfished islands have never been permanently inhabited by humans or intensively fished, and are therefore some of the least human-impacted coral reef systems in the tropical Pacific. Only a few hundred kilometers away from these unfished islands are islands forming part of the Republic of Kiribati, and they experience intensive fishing pressure, which has depleted the abundance of large-bodied fishes. The fishing in the area is artisanal fishing, which is fishing undertaken at a community or household level using low technology

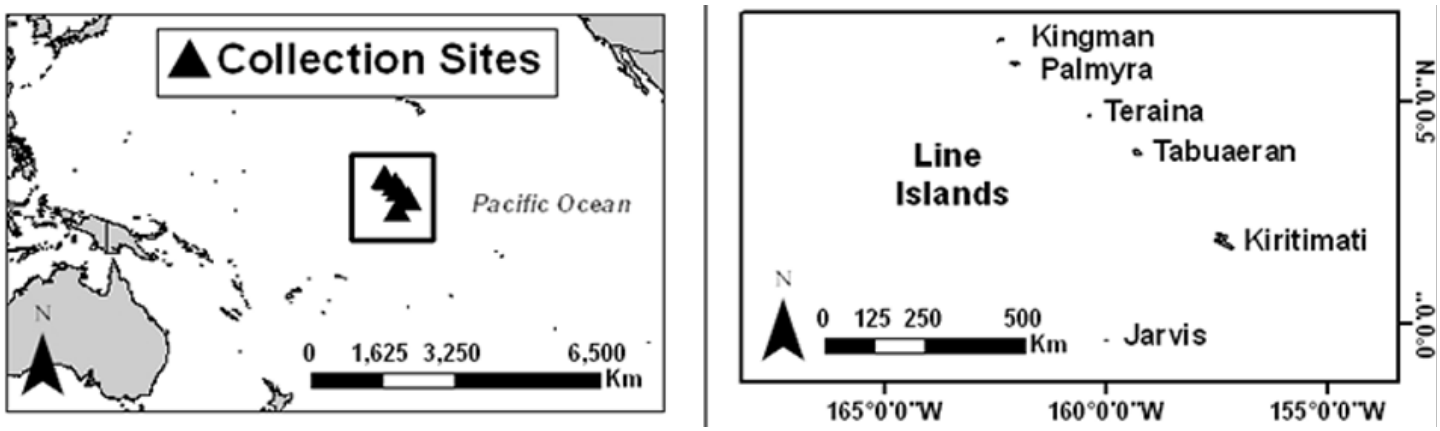


Figure 1. Map of the Northern Line Islands. Image credit: Wood et al. 2014; CC Public Domain.

methods.

We know that fishing can change the number and types of fish present around the islands that are fished. Now we want to know how those changes in host biodiversity (number and type of host fish present) might, in turn, influence what parasites are present and how many of each type of parasite there are. Two possible options include that (1) maybe fishing reduces the number of fish present, reducing the number of possible hosts, and potentially reducing the number of parasites or (2) higher diversity of fish in unfished areas might be linked to higher diversity of parasites.

A group of researchers wanted to know how fishing and human settlement might impact parasite communities. They joined up with a larger group of researchers and travelled all the way to the Line Islands to conduct this study. A total of 821 fishes (Figure 2) were collected by researchers while scuba diving at the six islands in October and November 2010. Fish were caught with a spear, placed in a bag underwater, and brought to the surface. Once the scuba divers were back on the boat, each fish was identified, numbered, and placed into an individual baggie. Fish were then frozen and transported back to the lab in the USA. Each fish was dissected and comprehensively examined to detect, identify, and quantify the abundance of metazoan parasites (parasites within the animal kingdom). Each parasite was classified according to its broad taxonomic group (Figure 3).

For this study, the researchers were specifically interested in the differences between two different types of metazoan parasites: trophically transmitted parasites (i.e., parasites with complex life cycles that must use hosts of multiple species, such as trematodes, cestodes, and nematodes) and directly transmitted parasites (i.e., parasites that can be transmitted among conspecific hosts, such as parasitic crustaceans and monogeneans), in terms of their response to fishing pressure. Of the six major groups of parasites they studied, three were trophically transmitted parasites (trematodes, cestodes, and nematodes) and three were directly transmitted parasites (monogenes, copepods, and isopods). These parasites tend not to be lethal, but they do take energy from their hosts. For example, isopods extract blood and fluids from their host by latching onto their host with piercing mouthparts, and cestodes (also called tapeworms) absorb important nutrients from the digestive system of their host.

Species: *Acanthurus nigricans*
Common name: whitecheek surgeonfish
Family: Acanthuridae (surgeonfishes)
Distribution: Indo-Pacific
Diet: herbivore, eat filamentous algae

Species: *Ctenochaetis marginatus*
Common name: blue-spotted bristletooth
Family: Acanthuridae (surgeonfishes)
Distribution: Indo-Pacific
Diet: detritivore

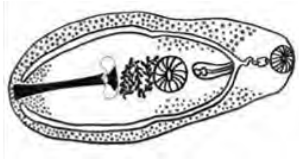
Species: *Cephalopholis urodeta*
Common name: darkfin hind
Family: Serranidae (sea basses)
Distribution: Indo-Pacific
Diet: predator, small fishes & crustaceans

Species: *Paracirrhites arcatus*
Common name: arc-eye hawkfish
Family: Cirrhitidae (hawkfishes)
Distribution: Indo-Pacific
Diet: benthic predator

Species: *Pseudanthias bartlettorum*
Common name: Bartlett's anthias
Family: Serranidae (sea basses)
Distribution: Pacific
Diet: zooplanktivore, feeds on plankton

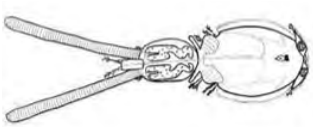
Species: *Stegastes aureus*
Common name: golden gregory
Family: Pomacentridae (damselfishes)
Distribution: Pacific
Diet: herbivore

Figure 2. Fish hosts collected from the Northern Line Islands, present in the dataset provided, and descriptions of their characteristics. Image credits (L-R): *A. nigricans*, D. Ross Robertson/Smithsonian Institution (public domain); *C. marginatus*, NOAA Photo Library (CC BY 2.0); *C. urodeta*, Rickard Zerpe (CC BY 2.0); *P. arcatus*, Richard Ling (CC BY-SA 2.0); *S. aureus*, NOAA Photo Library (CC BY 2.0).

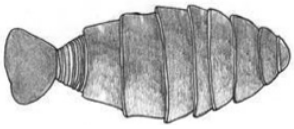


Trematode
Parasitic flatworms
Complex life cycle, trophic transmission

Monogene
Ectoparasitic flatworms
Direct life cycle



Copepod
Parasitic crustaceans
Direct life cycle



Isopod/Gnathiid
Parasitic crustaceans
Direct life cycle, Gnathiids are protelean parasites

Cestode
Tapeworms
Most have complex life cycles with trophic transmission

Nematode
Roundworms
All in this study have complex life cycles

Figure 3. General classes of parasites found associated with the fish hosts. Image credit: Danielle Claar.

PROCEDURE

Now you'll be provided the data collected from the above research project. First you'll explore the data, then you'll calculate the mean abundance and the standard error of the mean for the abundance of each parasite within each host, on both fished and unfished islands. You'll finish by plotting the results and considering the potential drivers of the patterns you find by responding to questions.

Explore The Data

1. Open the .csv or .xls file named "ParasiteBiodiversity_DATA" in Excel or other spreadsheet software (available from ncep.amnh.org).

DATA QUESTION ONE: How many rows does the file have? How many columns?

2. Below are the descriptions of the column headers and the associated data. There are questions embedded within to encourage you to get familiar with the data. Consider sorting or filtering data to help answering the questions, but if sorting, be sure to select all columns and all rows before sorting to ensure you do not scramble the dataset.
   - fish_sp : Fish species. The species of host are:
     aca_nig: *Acanthurus nigrans* (137 fish; 2 parasite taxa)
     cep_uro: *Cephalopholis urodeta* (162 fish; 4 parasite taxa)
     cte_mar: *Ctenochaetus marginatus* (141 fish; 4 parasite taxa)
     par_arc: *Paracirrhites arcatus* (129 fish; 2 parasite taxa)
     pse_bar: *Pseudanthias bartlettorum* (109 fish; 2 parasite taxa)
     ste_aur: *Stegastes aureus* (see Data Question 2)

DATA QUESTION TWO: How many *Stegastes aureus* fish were collected? How many different parasite taxa were found in/on this species?

   - fish_id: Unique fish ID.

DATA QUESTION THREE: How many unique, individual fish were collected in total?

   - island: Island where the fish was caught. The islands are:
     Jarvis
     Kingman
     Kiritimati
     Palmyra
     Tabuaeran
     Teraina

   - fishing_status: Either "Fished" or "Unfished"

   - depth: Depth in feet where the fish was caught.

DATA QUESTION FOUR: What was the deepest collection point? What was the shallowest?

- habitat: Habitat where the fish was caught. Either: "backreef," "forereef," or "patchreef"

- total_length: The total length of the fish in mm

- fork_length: The fork length of the fish in mm

- standard_length: The standard length of the fish in mm

DATA QUESTION FIVE: Find the shortest fish (by standard length) that was caught. How long is it in mm? What species of fish is it? Did it have any parasites?

- parasite: Which parasite is being counted. They are:
  Stephanostomum: *Stephanostomum* sp. Trematodes with trophic transmission
  FinMetacercariae: Fin metacercariae. Trematode larvae from the fish's fins with trophic transmission
  Grandiunguid: Grandiunguid sp. Copepod with direct transmission.
  Neobenedenia: *Neobenedenia* sp. Monogenean with direct transmission
  LarvalNematode: Nematode larvae with trophic transmission
  Tetraphyllidean: Tetraphyllidean sp. Cestodes with trophic transmission
  GillMetacercariae: Gill metacercariae. Trematode larvae from the fish's gills with trophic transmission
  Microscaphiid: Microscaphiid sp. Trematodes with trophic transmission
  Lepeophtheirinae: Lepeophtheirinae sp. Copepods with direct transmission
  Hatschekia: *Hatschekia* sp. Copepods with direct transmission
  Ancyrocephalid: Ancyrocephalid sp. Monogenean with direct transmission

- parasite_class: Classification of the parasite into the groups shown in Figure 3.
  cest: cestodes
  cope: copepods
  mono: monogeneans
  nema: nematodes
  trem: trematodes

- transmission: Transmission type. Either: "trophic" or "direct"

- count: The number of each parasite within that specific fish.

DATA QUESTION SIX: What does each row correspond to? Explain why there are multiple rows with the same unique fish ID.

3. In the next section, you'll be calculating the mean abundance and the standard error of the mean for the abundance of each parasite within each host, on both fished and unfished islands.

DATA QUESTION SEVEN: Which data columns do you think will be most important for these calculations? Which columns will most likely not be used?

Data Analysis & Visualization

1. Make a table of the mean ± standard error of the abundance of each parasite taxon/fish species combination. Include rows for "Unfished," and "Fished" islands. For each parasite, you should note underneath it whether it is directly transmitted or trophically transmitted. You should make one table for each fish species.

   Example:

Table 1. *Acanthurus nigricans* mean parasite abundance ± standard error for unfished islands and fished islands.

	Gill Metacercariae	**Neobenedenia**
Transmission	Trophic	Direct
Unfished	6.28 ± 1.37	
Fished		

   – Mean abundance ($\bar{x}$) is the average number of parasites per fish. It can be calculated by taking the sum of all the individual observations (x_i) and dividing by the number of observations (n). In Excel, you can type the command "=average()" and then select the cells you'd like to see the average of.

$$\bar{x} = \frac{\sum_{i=1}^{n} x_i}{n}$$

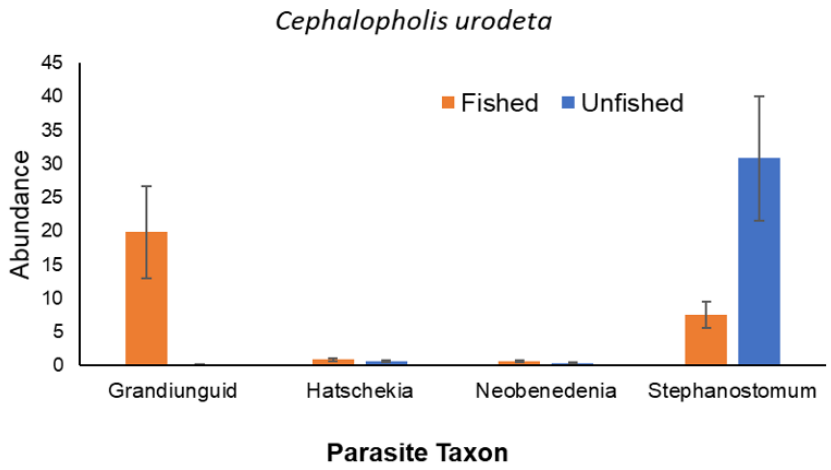
   – Standard error allows us to estimate the accuracy of our mean value. To calculate standard error, you first need to calculate the variance and standard deviation. First, you calculate the difference between the mean ($\bar{x}$) and each individual observation (x_i) and square that number. You then sum these squared differences and divide that by the number of samples minus one to get the variance. The standard deviation is the square root of the variance. The standard error is the standard deviation divided by the square root of the number of samples. In Excel, you can type the command "=stdev()/sqrt(count())" and within each (), select the cells you'd like to see the standard error of.

     Standard error = (standard deviation)/square root (sample size).

$$\frac{\sqrt{\frac{\sum_{i=1}^{n}(\bar{x} - x_i)^2}{n-1}}}{\sqrt{n}}$$

2. Plot mean ± standard error for each parasite taxon/fish species combination in both fished and unfished islands. To make a bar chart in Excel, you can select the cells with the data you'd like to plot. You then click insert along the top ribbon, followed by 2D column within the chart area. You can add your calculated error by clicking the "Chart Elements" button on the right of the chart (it looks like a plus sign). From there, click "custom" and select the values you'd like to appear as error bars. If you're unsure how to make the plots, Microsoft Office has an excellent help section which can guide you in the right direction, or you can search the internet for tutorials on making plots in your preferred spreadsheet software application.

Example:



*At the end of the session, you should have tables and charts for each fish species, as well as answers to the data questions.

DISCUSSION AND CRITICAL THINKING QUESTIONS

Answer the following questions based on the data you have generated.

1. Describe how fishing changes the mean parasite abundance. Give some examples from your results.
2. In the charts you made, you noted whether parasites were trophically transmitted (cycle through many hosts who eat each other) or directly transmitted (passed through the environment with fewer hosts). Looking at your summary data and the charts you've produced, does the impact of fishing differ between directly and trophically transmitted parasites? Do you notice any trends? Why do you think that you are seeing these patterns?
3. Did all host species have similar numbers of parasite taxa and abundances present? If there are differences, what about the hosts or parasites could cause them (consider reviewing Figure 2 and 3 in Background)? If there are no differences, provide one or two reasons that specific associations between parasites and their hosts may not happen?
4. These fish were collected by scuba divers underwater, brought to the surface, frozen, and then brought back to the lab to be dissected. How could collection methods alter the results? Which of the steps above could change the findings?
5. There were data columns that you did not use for the above calculations. Come up with a question that you could answer with the additional data that was collected. Do you think it's an interesting or helpful question for the researchers to ask? Why or why not?
6. Are there any additional data you would have collected within this study? Could any other stressors or environmental variables be linked to the patterns seen in the parasite communities?
7. These samples were collected in 2010. If these data were collected again today, do you expect the same patterns to be seen or would there be changes? If not, why not and if so, why?

FURTHER EXPLORATION

For more background information about this study and a more in-depth discussion of results, read:

– Wood, C.L., S.A. Sandin, B. Zgliczynski, A.S. Guerra, and F. Micheli. 2014. Fishing drives declines in fish parasite diversity and has variable effects on parasite abundance. Ecology

95(7):1929–1946.
– Wood, C.L., J.K. Baum, S.M.W. Reddy, R. Trebilco, S.A. Sandin, B.J. Zgliczynski, A. Briggs, and F. Micheli. 2015. Productivity and fishing pressure drive variability in fish parasite assemblages of the Line Islands, equatorial Pacific. Ecology 96(5):1383–1398.