Coral Growth and Survival Lab

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Part I: Pre-activities for the classroom

Science background

Corals are marine invertebrates in the class Anthozoa of phylum **Cnidaria**. They have a simple body structure (Figure 1) and like other cnidarians such as jellyfish, sea anemones, and zoanthids, they utilize **nematocysts** or “stinging cells” to catch potential prey (Figure 2). Polyps feed in two ways: either by utilizing nematocysts in the polyp’s tentacles to capture zooplankton, or by using a symbiotic, single-celled algae that lives in the coral’s tissue called **zooxanthellae** to photosynthesize (Figure 3). Like plants, zooxanthellae have internal structures called chloroplasts that are able to convert carbon dioxide and water, using sunlight, into sugar and oxygen. In return for food energy from the zooxanthellae, the coral provides shelter. Corals exist in either solitary or colonial forms. **Solitary corals** survive as a single, independent coral polyp. **Colonial corals**, which are the structure most people think of when picturing a coral reef, consist of many polyp clones living communally, often sharing skeletal or digestive systems among polyps.

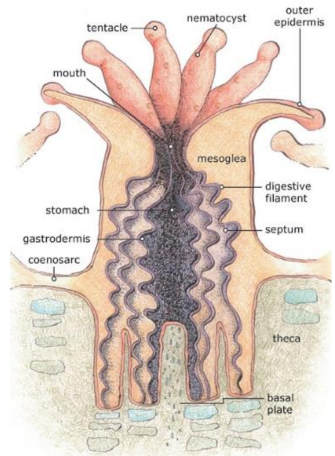

Figure 1. Anatomy of a coral polyp. (oceanservice.noaa.gov)

Like all animals, corals reproduce to generate new offspring. Coral larva (juvenile coral, Figure 4) are either genetically identical or genetically different from their parent(s) depending on the mode of reproduction. During **sexual reproduction**, corals combine sperm cells and ova (egg cells) to form **zygotes**. If the fertilization occurs internally, allowing the female coral to house developing larvae until they are ready to metamorphose, the process is called **brooding**. If the fertilization occurs externally, after the male and female corals release their gametes into the water column, the process is called **broadcast spawning**.

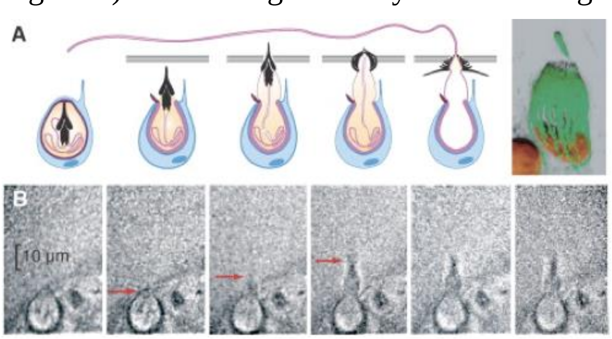

Figure 2. Schematic (A) and high-speed camera images (B) of a discharging nematocyst. The charged (ready-to-fire) nematocyst is on the left, the fully-discharged nematocyst on the right (Nüchter et al. 2006).

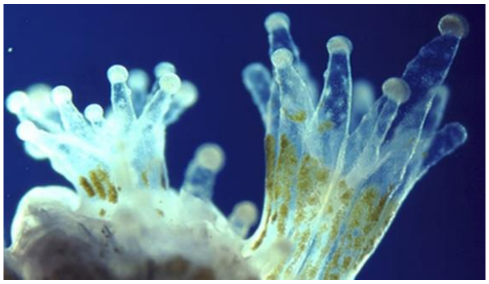

Figure 3. Coral polyp with clearly visible zooxanthellae (green structures in the clear coral tissue) (ocean.si.edu).

Corals can also utilize **asexual reproduction**, which does not require an exchange of genetic material (i.e., no combination of eggs and sperm). **Fragmentation** occurs when a branch of coral breaks off the parent colony (as can often happen during a storm event). The broken branch, under right conditions, reestablishes itself and becomes a new coral colony. During **budding**, a new polyp develops, or buds off, from an outgrowth or mature polyp (Highsmith 1982). The new polyp separates from the parent polyp when it is mature. The polyps created from fragmentation or budding are clones and genetically identical to the parent coral since there is no mixing of genetic material during reproduction.

After fertilization and many rounds of cellular division occur, a free-swimming coral larva, called a **planula** (Figure 4), is able to start looking for a suitable place to settle and continue development. The planula is covered in many small hairs, called **cilia**, which are used for locomotion. Planula can also use their cilia to detect sounds and orient themselves to the source of the sound (Vermeij et al. 2010). This is helpful for locating healthy reef environments which tend to be noisy with wave energy and animal activity.

The planula has a set of sensory organs, called the **apical sensory organs,** which detect favorable chemical cues for settlement. These sensory organs are located on the aboral end of the planula, opposite of the region where the mouth is located. The apical sensory organs are located along the outer layer of skin on the planula along with nematocysts and other secretory gland cells. When a planula finds a suitable place to settle, it attaches to the surface, flattens out, and metamorphoses into a polyp (this first stage is also called a "spat," Figure 5). The polyp begins secreting a calcium-carbonite skeleton known as a **calyx** (Tran & Hadfield 2013). As the polyp grows, buds, and continues secreting its rocky skeleton, it contributes to the growth of the reef structure.

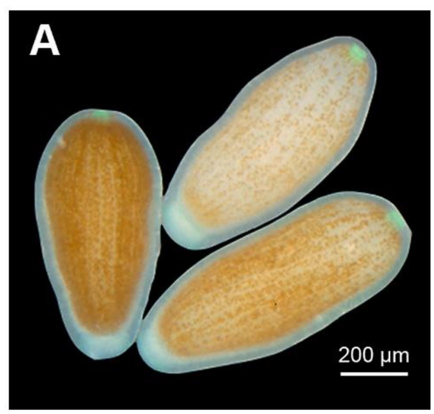

Figure 4. Coral planula observed at 100X

<u>Importance of Coral Reefs</u>

Corals have a complex 3D structure with high **rugosity**, or roughness, to provide nooks and crannies that are used by marine animals as homes. This habitat is important for all types of small creatures that we rarely see ("cryptofauna") including shrimp, crabs and worms, providing them a safe place to reproduce in addition to hiding from predators. Coral reefs are also important to the people of Hawaiʻi. The reefs in Hawaiʻi contribute $800 million per year to the state's economy, mostly from the tourist dollars that they attract to the islands (Davidson et al. 2003). Coral reefs are important in the dispersal of wave energy making coral reefs important to homeland security during storms and tsunamis. There is also a strong cultural tie to coral in Hawaiian legends and coastal resource management.

Other uses for corals include certain chemicals corals use to defend themselves from other corals. These chemicals have been found to have human application in fighting bacterial infections and cancer. Within this century, scientists have isolated chemicals secreted by corals to fight cancer (Rangell & Falkenberg 2015). On a global scale, reefs have provided the optimal foundation for ports and harbors on all coasts. But impacts to reef health and structure from overuse by humans is taking its toll - as reefs die, shores are prone to storms, erosion, and wave action, in addition to straining the relationships corals have with other animals that depend on them for survival. All around the world, corals have adapted to their environments and their ecosystems, but a changing environment (which includes rising sea levels, increasing ocean temperatures, and ocean acidification) threatens these fragile relationships.

Figure 5. A coral "spat" (settled coral after metamorphosis, left) and coral planula (right).

Research at the Hawai‘i Institute of Marine Biology

Healthy coral colonies make up the foundation of any health reef ecosystem. Since these animals play such an important role to a wide variety of organisms, both marine and terrestrial, there are many labs at HIMB that are dedicated to furthering our understand of individual coral formations, their interactions within a reef ecosystem, the human applications that can be derived from corals, and the damage that humans have caused to these organisms. The Karl Lab looks at coral resilience on a small scale and investigates the factors that impact possible microhabitats present on an individual coral. The Toonen-Bowen Lab examines the larger scale relationships within a coral reef ecosystem by assessing a variety of factors that impact reef invertebrates and fish. The Hagedorn Lab investigates cooling rates of coral gametes, cooling rates of symbiotic algae and cryopreservation of coralline genetic material for future studies and conservation. . The Point Lab was instrumental in understanding the biochemical mechanisms behind ocean acidification and corals. The Point Lab designed flumes that were able to mimic future oceanic conditions and provide key insights into understanding the chemical processes and mechanics behind ocean acidification

Dr. Ruth Gates is the principal investigator of the Gates Lab and has been working with researchers and graduate students to investigate attributes in corals that determine differences and sensitivity in their ability to adapt to thermal stress, ocean acidification and anthropogenic pollution. The Gates Lab's main focus is on early coral resilience, settlement, and development and the breeding of so-called "super corals". "Super corals" are naturally occurring coral species that are more adaptable to environmental changes and better suited to a climate that has shifted towards more acidic conditions. By looking at larval coral responses to conditions that mimic future ocean conditions, the scientists in the Gates Lab are able to examine the genes that make corals most resilient in the face of a changing climate. Additionally, the Gates Lab studies the correlation between the abundance of zooxanthellae and their impact of coral recruit growth.

Classroom Laboratory:

Background Research

Researchers do a considerable amount of preliminary investigation by reading previously published results in order to become more informed of the latest information on a particular topic area. There is already a considerable amount of information available and accessible on the internet. Before their field trip to HIMB, students should do their own background research and become more familiar with the key concepts of reproduction mechanisms in coral, how corals choose suitable places for settling, and how changing environmental conditions affect a polyp's ability to survive. The following links can provide a starting point for students' research:
http://www.icriforum.org/about-coral-reefs/what-are-corals
http://oceanservice.noaa.gov/education/kits/corals/coral06_reproduction.html
http://oceanservice.noaa.gov/facts/coralreef-climate.html

Classroom activities

Below is a classroom activity designed to help students become familiar with the complex ways corals reproduce and form new reef structures. One important theme to keep in mind is how human changes to the environment affect coral's ability to settle and undergo metamorphosis into the conventional coral form we are familiar with. The exercise will try to instill in students the difficulty of a coral larva's journey from planula to adult coral.

1. One recommended classroom activity is for the students to research and illustrate the different modes of coral reproduction. The diagrams drawn by students should include depictions or descriptions of **budding**, **brooding**, **fragmentation**, and **broadcast spawning**. Students may also outline the differences between sexual reproduction and asexual reproduction and list any specific species of coral that use a certain mode of reproduction.
    a. Items needed:
        i. Research materials (computers, books, journal articles)
        ii. Posterboard and markers

Part II: Field trip day at HIMB

Introduction

In this activity, students will be scanning terracotta tiles to collect qualitative and quantitative data on coral recruitment and growth in different environments. These tiles will be retrieved from different spots around Moku o Lo‘e that each have unique environmental conditions varying in temperature, turbidity, human activity, pH, and salinity. Instructors will also collect water quality data at these tile sites before students arrive.

Once the students arrive in the classroom at the Marine Science Research Learning Center at HIMB, they will separate into groups of four, be assigned two coral settlement tiles, and make predictions on coral polyp health based on previous data and water quality. Each group will measure coral larval health, settlement, and growth by recording the number of recruits found on the tile that day and by taking pictures of coral colonies on digital microscopes. The pictures taken will be analyzed on open-source, digital imaging software (ImageJ, NIH) in which the exact area and number of polyps comprising each coral colony can be determined. Once the data is collected, students will add their data to a collaborative database documenting coral health in Hawaii‘s marine ecosystems.

Tools Available

- Coral tiles
- Pipette(s)
- Graduated beakers
- Petri dishes
- Filtered sea water
- Digital microscopes (Celestron, MicroCapture Pro)
- Micro-Scaling tool
- Laptops w/ImageJ software

Materials to bring to HIMB

- Writing utensil
- Paper/Notebook

HIMB Classroom Laboratory Activity

Procedure

The instructors will retrieve the coral settlement tiles and record water quality data using field pH meters to measure acidity or basicity and refractometers to measure salinity. They will also record any other observations about the tile sites (e.g., whether runoff from previous heavy raining is present at tile collection site) to provide environmental information for the students. Each pair of students will record the number of coral recruits on one tile using a Celestron

Digital Microscope and an orthographic chart of a tile (See Appendix A for orthographic chart). To collect data, students should do the following:

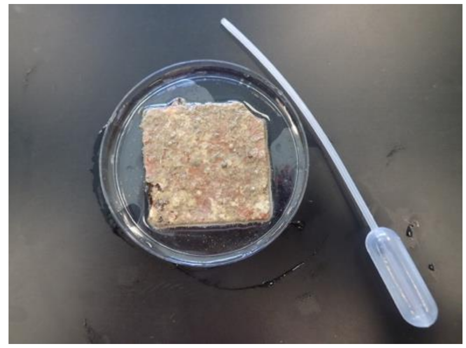

Figure 6. Coral settlement tile in water and bulb pipette. Notice the tile orientation with the "chip" off the tile in the upper right hand corner.

1. Gently place a settlement tile in a petri dish with sea water. Use the bulb pipette to occasionally rehydrate the surfaces that are not submerged in seawater.
2. Record the placement of each coral colony of the tile on the orthographic chart. You can use the digital scope or hand magnifying glass to help you zoom on areas.
3. After a coral colony has been found, take a picture of the colony on the laptop using the digital scope with the micro-scale tool in the picture. **Make sure that the micro-scale tool is in the image, the colony/spat is in the center of the image, and take one picture for each colony/spat.** This step is crucial in being able to determine the correct scale of the colony in ImageJ.
4. Save the picture to the desktop **with the file name generated on your Excel sheet** (e.g., 20170315-PL201-C6.png). These tiles can be difficult to handle while taking pictures so make sure to work in pairs, where one student handles the tiles and the other student handles the computer.
5. Count the number of polyps you see for the colony. Note the health condition of the polyps (healthy, bleached, dead, or other). Next, we will measure the area of the colony.

Once you have recorded all the colony locations on the orthographic chart and have saved the digital pictures taken on the desktop, you are going to analyze the pictures with ImageJ.

A. Click on the ImageJ icon (). You will see a small ImageJ toolbar appear.
B. Go to *File>Open* (or, command+O) and then select the first picture to be analyzed from the desktop.
C. Select the *Straight* line tool in the toolbar (). Click, drag and release to draw a straight line between two of the numbered ticks (e.g., the 0 (zero) and 1 millimeter notches) on the ruler in the picture.
D. Go to *Analyze>Set Scale...*
    a. In the pop-up box, in the *Known Distance* box enter "1" (because the line is 1mm long).
    b. In the *Unit of Length* box, enter in unit used (mm), then hit "OK"
E. Go to *Analyze>Set Measurements...*
    a. In the popup-box, check off Area, then hit "OK"

F. Select the Freehand selections tool () and carefully trace the perimeter of the coral colony
G. Go to *Analyze>Measure* (command+M)
H. A data window will be displayed with your coral area measurement in the unit you specified (mm^2). Record the area of the colony on your datasheet.
I. Repeat steps B-I for each additional coral colony/spat picture.

Once each picture has been analyzed, students will add their data to previous students' data using the provided Excel data sheet. After all the students have added their data to the database, the instructor will share the class's results and compare it with previously collected data.

After recording their observations, the students will discuss and make predictions on the coral colony health based on certain water quality conditions. This laboratory exercise is an example of descriptive research, used by scientists to generate hypotheses. After collecting growth data from their coral tiles, the students will be encouraged to generate their own research questions/hypotheses. Below are some questions to help guide students towards the important factors that might affect coral settlement and survival:

Guiding Questions

1. How is climate change and human interaction affecting Hawai'i's marine environments?
2. What factors influence where a coral will settle?
3. What types of resources does a coral need to compete for? Who are coral's main competitors for resources on the reef?
4. Are coral and zooxanthellae (the algae that live in coral) both affected equally by environmental changes? What types of changes affect them the most?
5. What environmental factors affect how quickly a coral can grow? Can the same species have different growth rates in different environments?
6. Do "natural" environmental changes (e.g., seasons, storms, animal migrations) affect coral reefs more than anthropogenic changes (e.g., pollution, boat activity, ocean acidification)?

Part III: Post-activities back in the classroom

A critical part of the scientific process is being able to analyze and interpret your data, and then communicate your findings with both the scientific and general public communities. Once back in your home classroom, you will discuss your data with your labmates and can further examine the larger coral growth data set available online to compare with your findings. After a research project, scientists work hard to publish a written report and/or present their experiment at a regional, national, or international conference meeting. Being able to effectively communicate research to a broad audience allows you to successfully contribute to our current understanding of the natural processes of our world.

Begin class with a general discussion of your experience at HIMB. Were you prepared for the lab exercise and able to complete the activity? Did you enjoy your time there? Now begin to discuss the subject matter:

A) Did your group discussions before doing the lab reflect what was seen on each of your tiles? If not, why do you think your presumptions did not align with what was seen at the tile site?
B) Did you notice if there were any environmental conditions or combinations of conditions that killed the coral colonies? Did coral planulae settle on certain parts of the tile more so than other sides?
C) Did you notice other organisms growing on the tiles?
D) Do your findings influence the way you view the ocean? Will it change your behavior in terms of reducing your carbon footprint?

<u>Lab Report</u>

Written or oral lab reports are a good way to interpret your data, and share the story of your research. For your laboratory exercise at HIMB, you will be expected to eventually provide an in-depth laboratory report that should include the following independent sections:

- **Title**: summarize the entire experiment in several words.
- **Introduction**: in one half to three-quarters of a page describe the lifecycle of coral polyps, how coral settle to form new reef structures, as well as how humans and climate change are affecting coral larvae, and why we should care.
- **Materials and Methods**: develop and describe, in detail, the observations you conducted; include all of the materials you used to complete it as well.
- **Results**: compile your data and express them visually and where appropriate in graphs, tables, or figures.
- **Discussion** (including **generated hypotheses/research questions**): analyze your data in essay form; discuss the results, emphasize what did and didn't work, and propose a new experiment or changes to your original experiment which may help explain the results; discuss the research questions or hypotheses generated by your observations.

- **Conclusion**: in a paragraph or so, summarize your results and make concise conclusions about them; also include a sentence or two conveying your general conclusions about your results

NOAA Data in the Classroom: Investigating Coral Bleaching

For further discussion about factors affecting coral reef health and survival, the National Oceanic and Atmospheric Administration (NOAA) offers a free, interactive data analysis module focusing on coral bleaching (https://dataintheclassroom.noaa.gov/content/coral-bleaching). The module, which includes additional slide presentations, data, lesson plans, and worksheets, describes in greater detail the threats faced by corals worldwide.

Beyond the Classroom

Scientists need to recognize the importance of their work outside of the laboratory. Although gaining knowledge just for knowledge's sake can be fun, research should have a practical application to humanity and the world at large. After completing these laboratory exercises, we hope your students are more aware of some of the issues facing their community in light of global climate change. Hawai‘i boasts a unique and biodiverse environment that is particularly threatened by increasing temperatures and rising sea levels.

Students should brainstorm ways in which they might be able to raise awareness about the issues facing corals, or observe or monitor changes in their local environment. Students could host an activity week or produce a video or presentation on the importance of corals in Hawaiian culture. For science fair projects, students could monitor changes in coral bleaching over time, human use and activity at different beaches, or compare bleaching at different sites around their island. There are also volunteer opportunities such as beach cleanups, invasive algae removals, or watershed restoration projects. Even at home, students could be encouraged to turn off lights or use less water. Interviews with local kupuna to learn about traditional practices in caring for the marine environment could also help students strengthen their sense of responsibility to their community.

For more information on Hawaiian community coastal resource management, the Department of Land and Natural Resources offers a community guidebook.

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Appendix A: Coral Orthographic Growth Chart

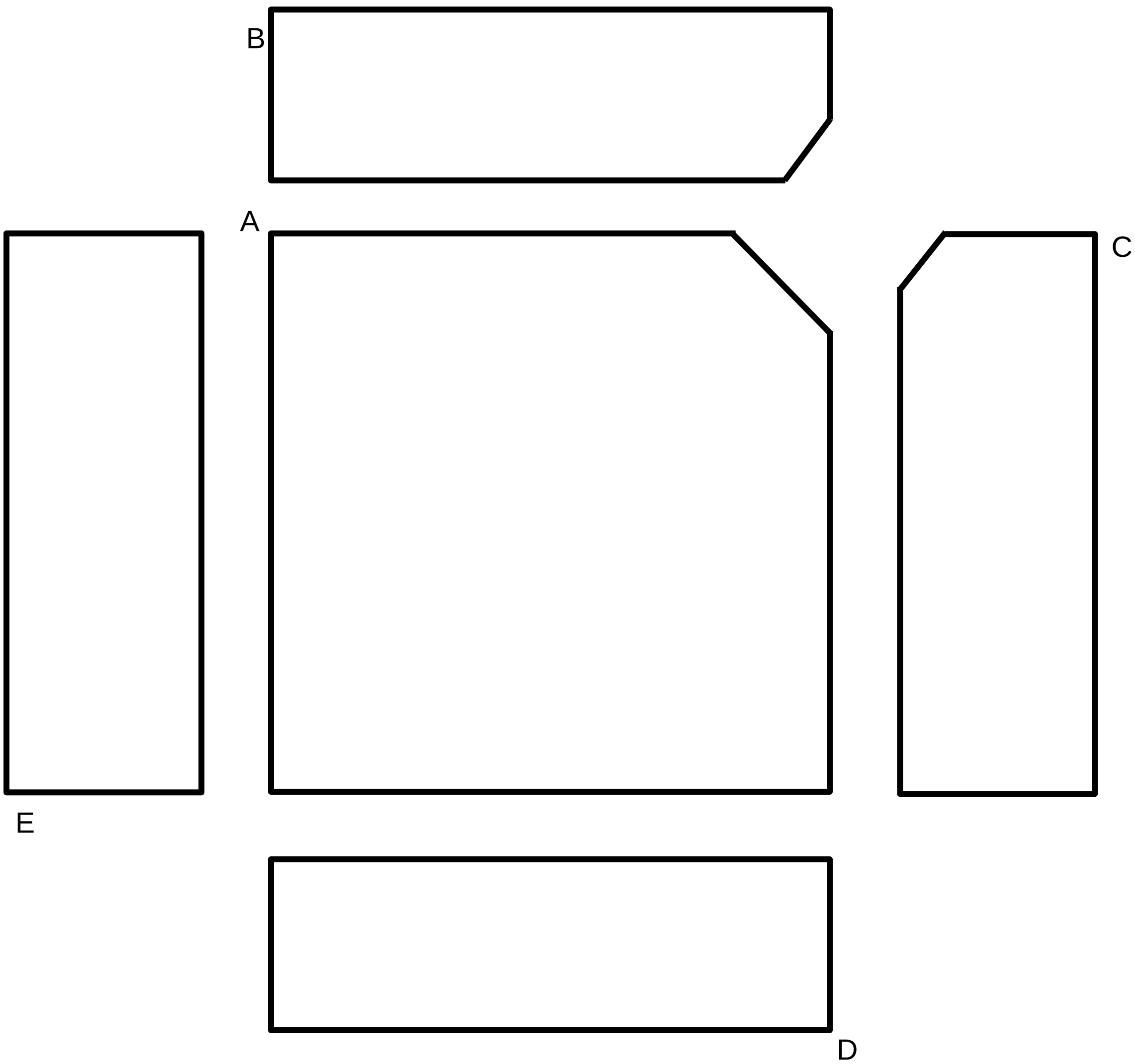
Date : ______________________________

School: ____

Tile ID: ______________________________

Environmental Data

pH: _________ **Temperature (C˚):** __________ **Salinity (ppt):** ___________



Appendix B: Relevant Next Generation Science Standards

Performance Expectations:
HS-LS2-6 Ecosystems: Interactions, Energy, and Dynamics
- Evaluate the claims, evidence, and reasoning that the complex interactions in ecosystems maintain relatively consistent numbers and types of organisms in stable conditions, but changing conditions may result in a new ecosystem.

HS-LS2-7 Ecosystems: Interactions, Energy, and Dynamics
- Design, evaluate, and refine a solution for reducing the impacts of human activities on the environment and biodiversity.*

Science and Engineering Practices:
Scientific Knowledge is Open to Revision in Light of New Evidence
- Most scientific knowledge is quite durable, but is, in principle, subject to change based on new evidence and/or reinterpretation of existing evidence.

Engaging in Argument from Evidence
- Evaluate the claims, evidence, and reasoning behind currently accepted explanations or solutions to determine the merits of arguments.

Planning and Carrying Out Investigations
- Plan and conduct an investigation individually and collaboratively to produce data to serve as the basis for evidence, and in the design: decide on types, how much, and accuracy of data needed to produce reliable measurements and consider limitations on the precision of the data (e.g., number of trials, cost, risk, time), and refine the design accordingly.

Scientific Investigations Use a Variety of Methods (*Nature of Science)*
- Scientific inquiry is characterized by a common set of values that include: logical thinking, precision, open-mindedness, objectivity, skepticism, replicability of results, and honest and ethical reporting of findings.

Analyzing and Interpreting Data
- Apply concepts of statistics and probability (including determining function fits to data, slope, intercept, and correlation coefficient for linear fits) to scientific and engineering questions and problems, using digital tools when feasible.

Obtaining, Evaluating, and Communicating Information
- Communicate scientific information (e.g., about phenomena and/or the process of development and the design and performance of a proposed process or system) in multiple formats (including orally, graphically, textually, and mathematically).

Disciplinary Core Ideas:
LS2.C: Ecosystem Dynamics, Functioning, and Resilience
- A complex set of interactions within an ecosystem can keep its numbers and types of organisms relatively constant over long periods of time under stable conditions. If a modest biological or physical disturbance to an ecosystem occurs, it may return to its more or less original status (i.e., the ecosystem is resilient), as opposed to becoming a very different ecosystem. Extreme fluctuations in conditions or the size of any population, however, can challenge the functioning of ecosystems in terms of resources and habitat availability.

LS4.D: Biodiversity and Humans

- Biodiversity is increased by the formation of new species (speciation) and decreased by the loss of species (extinction).
- Humans depend on the living world for the resources and other benefits provided by biodiversity. But human activity is also having adverse impacts on biodiversity through overpopulation, overexploitation, habitat destruction, pollution, introduction of invasive species, and climate change. Thus sustaining biodiversity so that ecosystem functioning and productivity are maintained is essential to supporting and enhancing life on Earth. Sustaining biodiversity also aids humanity by preserving landscapes of recreational or inspirational value.

LS2.D: Social Interactions and Group Behavior

- Group behavior has evolved because membership can increase the chances of survival for individuals and their genetic relatives.

Crosscutting Concepts:

Cause and Effect

- Empirical evidence is required to differentiate between cause and correlation and make claims about specific causes and effects.

Common Core State Standards Connections:

ELA/Literacy

RST.11-12.1: Cite specific textual evidence to support analysis of science and technical texts, attending to important distinctions the author makes and to any gaps or inconsistencies in the account. (HS-LS2-2)

WHST.9-12.2: Write informative/explanatory texts, including the narration of historical events, scientific procedures/ experiments, or technical processes. (HS-LS2-2)

RST.11-12.7: Integrate and evaluate multiple sources of information presented in diverse formats and media (e.g., quantitative data, video, multimedia) in order to address a question or solve a problem. (HS-LS2-6)

WHST.9-12.7: Conduct short as well as more sustained research projects to answer a question (including a self-generated question) or solve a problem; narrow or broaden the inquiry when appropriate; synthesize multiple sources on the subject, demonstrating understanding of the subject under investigation. (HS-LS2-7)

WHST.9-12.9: Draw evidence from informational texts to support anafalysis, reflection, and research. (HS-LS4-4)

Mathematics

MP.2: Reason abstractly and quantitatively. (HS-LS2-2)

HSN.Q.A.3: Choose a level of accuracy appropriate to limitations on measurement when reporting quantities. (HS-LS2-2)