Morphology refers to the physical characteristics of an organ or organism. In most organisms, including plants, morphological characteristics result from three primary processes: (i) natural selection, (ii) phenotypic plasticity, and (iii) developmental programs and pathways. In each of these cases, environmental conditions exert some limit on physiological processes, and these limitations result in specific morphological characteristics. For example, endothermic animals that live in colder environments often have larger bodies and a smaller surface area to volume ratio than endotherms that live in warmer environments, which allows easier maintenance of body temperature within their range of thermal tolerance (homeostasis). Because morphology and underlying physiology are strongly influenced by the environment, organisms living in different environments can exhibit great variation in morphology.

Plants also have an optimal range of temperatures in which they can survive, and therefore have evolved a variety of morphological characteristics to help maintain homeostasis. Plant temperature requirements are primarily related to photosynthesis. At low temperatures, plant cell contents freeze and metabolic activity is halted, and at high temperatures, proteins associated with photosynthesis and other physiological processes are denatured. Leaves are the primary organ associated with photosynthesis and gas exchange, and are thus also the site of most temperature regulation. Leaves capture heat via solar radiation and ambient heat and lose heat primarily via evaporative water loss (transpiration). In order to maintain ideal temperatures for photosynthesis, plants have evolved a variety of leaf traits that influence their ability to capture heat and lose heat. Some of these leaf morphological traits include leaf surface area, leaf margin characteristics, stomatal density, and leaf shape. For example, we might expect that a plant in a hot environment might benefit from high stomatal density, which allows for greater evaporative water loss, and low leaf surface area, which reduces sunlight capture. However, if the hot environment is also dry, a plant with high stomatal density might be at risk of dehydration, and low leaf surface area could contribute to a reduced rate of photosynthesis. These evolutionary trade-offs add complexity to expected leaf morphology-environment relationships.

In this research project, you will collect leaves from woody dicot plants across a range of environments that are characterized by different mean annual temperatures (MAT) and investigate the relationship between various leaf characteristics and temperature to answer the following research question:

**Research Question: What leaf morphological characteristics are most strongly associated with mean annual temperature?**

Over the course of the semester, you will address this question by accomplishing the following:

1. Collect at least 150 woody dicot leaf samples across sampling sites characterized by a broad range of mean annual temperatures. As a class, we will take five field trips to different locations to collect samples. **You will be responsible for sampling at two additional sites outside of class/lab.**

2. Analyze leaf samples by collecting data on leaf shape and margin characteristics, stomatal density, and leaf surface area. You will use visual assessment, microscopy, and computational analysis to develop your leaf morphology data sets.
3. Preserve leaf samples according to herbarium standards to contribute to the University herbarium collection.
4. Extract mean annual temperature data for each of your sampling sites from an online database (climatewna.com).
5. Quantify the relationships between individual leaf morphological characteristics and mean annual temperature by fitting simple linear regression models to your data in R.
6. Synthesize and communicate your process and findings in a formal research paper.

This research project is designed to address a variety of research skills that are important for practicing biologists across a range of disciplines, as well as several skills and competencies that are specifically related to plant biology. Your performance will be assessed by formative assessments and summative assessments according to these learning outcomes.

Learning Outcomes:
1. Follow the process of science to develop an evidence-based hypothesis grounded solidly in scientific literature, collect data according to a sampling design that ensures sufficient representation, and generate meaningful conclusions from data that directly address the hypothesis and research question and reflect a mechanistic understanding of observed trends in data.
2. Use a species identification guide and other resources to accurately identify deciduous tree and shrub species.
3. Prepare plant herbarium specimen that are carefully and correctly preserved.
4. Effectively and accurately use computational tools including ImageJ and R to collect and analyze data.
5. Access and utilize publicly-available data from an online database.
6. Design and conduct a proper regression analysis that directly addresses the research question, tests the hypothesis, and is correctly interpreted and presented in the context of the research question and hypothesis.
7. Clearly articulate how morphology-climate relationships reflect underlying environmental constraints on physiology.
8. Understand and articulate how variation in plant characteristics reflect evolutionary responses to the environment.
9. Correctly communicate the motivation, process, results, and implications of the research in a format and style that are consistent with scientific standards.

Formative Assessments:
1. Pre-lab for week 1 (5 points)
2. Hypothesis and Introduction draft (15 points)
3. Data Analysis I Homework (10 points)
4. Data Analysis II Homework (10 points)
5. Research team meeting and inference table discussion with professor (10 points)

Summative Assessments:
1. Final research paper (100 points)
2. Final herbarium collection with at least 150 samples (150 points)
Pre-Lab Activity

Prior to the first lab of the semester, read through the research project overview and complete the following exercises. Bring this completed page with you to lab.

1. Vocabulary
Provide a definition for each of the following terms from the research project overview. Be sure to look up the definition in a reliable source and ensure that the definition you are using is consistent with the context in which the term is used in the overview.

Phenotypic plasticity:

Homeostasis:

Transpiration:

Evolutionary trade-offs:

Woody dicot plant:

2. Climate-morphology relationships
The table below lists several of the leaf morphological characteristics that you will measure in this research project. Fill in the blank boxes in the table with a prediction for what variation of that trait might be found in a warm environment versus a cool environment.

| Morphological trait                                      | Warm environment | Cool environment |
|----------------------------------------------------------|------------------|------------------|
| Leaf surface area                                         |                  |                  |
| (large, small, intermediate)                              |                  |                  |
| Stomatal density                                          |                  |                  |
| (high, low, intermediate)                                 |                  |                  |
| Leaf margin teeth size                                    |                  |                  |
| (large, small, intermediate, no teeth; see pp. X for examples) |                  |                  |
| Length to width ratio                                     |                  |                  |
| (>1:1, <1:1, =1:1)                                        |                  |                  |
| Leaf shape                                                |                  |                  |
| (obovate, elliptic, ovate; see pp. XX for examples)       |                  |                  |