

## INTRODUCTION

Arctic environments present unique challenges to tundra plant species. Growing seasons are short, temperatures low, and nutrients scarce, yet a surprisingly morphological diversity of plants coexist across Arctic ecosystems today. Tundra shrubs — woody, often deciduous species, including dwarf birch (*Betula nana*), dwarf willows (*Salix sp.*) and green alder (*Alnus viridis*) — are key players in permafrost ecosystems, with direct impacts on surface energy balances, nutrients, and carbon cycling. While these shrubs may be dwarfed in comparison with trees in other systems, collectively tundra plant communities are the gatekeepers of vast amounts of belowground carbon currently locked in Arctic permafrost.

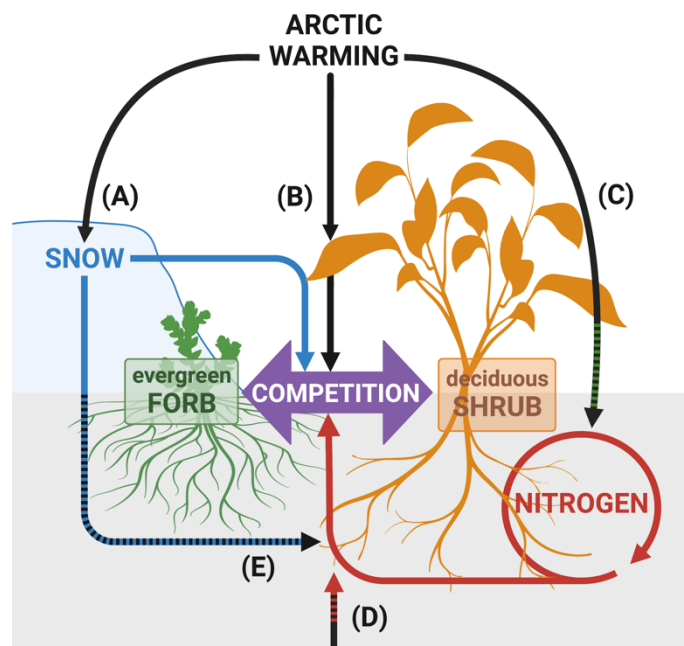
In the coming decades, tundra species must adapt to a rapidly changing world. Mean air temperature is increasing three times faster in the Arctic than the rest of the globe, already +3.1°C since the 1970's (AMAP 2021). Multiple lines of evidence from plot-level observations to satellite remote sensing suggest that anthropogenically-caused Arctic warming is already reshuffling plant communities. Tundra shrubs are responding particularly rapidly, increasing in range, size, and abundance across the Arctic and outcompeting other species, a pattern termed “Arctic shrubification”. Quantifying and testing the mechanisms controlling these competitive shifts in tundra plant communities is critical to our understanding of the current and future Arctic carbon cycle with urgent global consequences. **The proposed research builds on our ongoing work to clarify the mechanisms of Arctic shrubification through the synthesis of historical and contemporary data on plant community change, and the implementation of new experiments informed by trait-based mathematical models of plant competition.**

In the last decade, researchers have made great progress in describing patterns of Arctic shrubification. Satellite remote sensing, drone surveys, repeat photography, and field-based sampling all indicate widespread greening, isolated browning events, and shifts in species composition (Mekonnen et al. 2021, Myers-Smith et al. 2020). Plot-based observational studies and warming experiments indicate an increase in shrubs with increasing temperatures (Elmendorf et al. 2012a,b). Dendrochronological surveys from Canada and Alaska show that willows and alders are recruiting to higher elevations and expanding upslope (Myers-Smith & Hik, 2018, Salmon et al. 2019). Evidence for shrubification includes growth of existing shrubs, infilling of shrub patches, and expansion of species-ranges — all of which indicate shrubs are outcompeting other tundra species (Bret-Harte et al. 2002, Gough et al. 2019, Myers-Smith et al. 2011). A recent synthesis of plot-based data and remote sensing patterns demonstrates widespread increases in deciduous shrubs across Alaska (Macander et al. 2022). In fact, “shrubification” is the most common pattern of vegetation change observed across the Arctic (Heijmans et al. 2022).

Our understanding of the drivers and mechanisms controlling patterns of Arctic shrubification remains much less clear. This task is complicated by the fact that while temperature is the ultimate driver of this process, there are multiple pathways through which Arctic warming impacts competitive hierarchies (Fig. 1). First, increases in air temperature can directly alter plant ecophysiological function, favoring deciduous shrubs (Fig.1, arrow B). Second, warming accelerates snowmelt and lengthens the growing season, and might thereby favor deciduous shrubs that benefit from longer snow-free periods (Fig.1, arrow A). Finally, warming increases nitrogen availability by enhancing microbial activity and accelerating nitrogen mineralization, and releasing new nitrogen from thawing permafrost (Fig. 1, arrows C, D). Although the relative contribution of these various pathways is uncertain, past experiments suggest that warming impacts on snow regimes and nutrient cycling may overwhelm the direct effect of warming (Shaver & Chapin 1980, Sturm et al. 2001, Addis et al. 2019, Prager et al. 2020).

Disentangling these various drivers is made even more complex by ecosystem feedbacks. Shifts towards deciduous shrubs increase the rate of litter input and decomposition, creating a positive feedback mediated through increasing nutrient availability (McLaren et al. 2017, Wookey et al. 2009). Shrubs also

alter snow regimes: tall branches can trap snow, increasing winter snowpack depth and insulating plants and soils (Sturm et al. 2001 & 2005, Schimel et al. 2004). These interrelated pathways limit the power of statistical approaches that correlate patterns of shrubification with environmental drivers. Moreover, understanding how environmental change in the Arctic propagates through multiple pathways to restructure tundra communities requires identifying and quantifying salient mechanisms controlling competitive outcomes between functionally different plant species (Fig. 1). Investigating Arctic shrubification as a competitive process remains a significant challenge and research priority in the field (Martin et al. 2017, Mekonnen et al. 2021).



**Figure 1. Climate drivers act through multiple pathways to structure plant competition in Arctic ecosystems.**

Warmer air temperatures impact plant competition by altering ecophysiological rates (B), snow regimes (A, E), and nutrient cycling (C,D). Warming increases microbially-mediated nitrogen mineralization in soils (C) and unlocks new sources of nitrogen from thawing permafrost (D). Warming may accelerate snowmelt and delay accumulation, extending the length of the growing season (A). More snowpack insulates the soil, increasing winter mineralization of nitrogen (E). Negative feedbacks between warming and shrubs could occur due to canopy shading in summer (C,D), while positive feedbacks may arise from shrub-trapping of snow in the winter (A,E). Changes in snow (A,E) with warming may intensify or weaken competitive interactions between deciduous tundra shrubs and evergreen competitors (purple arrow).

Recent advances in plant ecology that integrate functional traits into mechanistic models of plant competition offer a promising way forward in understanding the mechanisms of shrubification (Dybzinski et al. 2011, Levine et al. 2022, Detto et al. 2022). These physiologically-based models can predict outcomes of competition across gradients in environmental drivers, and analytically determine optimal, competitively stable plant functional strategies. This approach is an advance beyond traditional models of competition between two plant functional types, in that the most competitive strategies are determined from a true continuum of plant trait values represented in tundra ecosystems (Thomas et al. 2019, Saccone et al. 2017). Furthermore, mechanistic modeling approaches provide more predictive understanding than statistical fits of trait-environment correlations, which might break down in an era of global change. By scaling plant traits to whole-plant carbon balances across environmental gradients, this framework can help clarify the mechanisms controlling plant competition at a species level, and thus shifts towards shrubification.

Although this novel quantitative approach offers promising insight, any explanation of shrubification must account for observed heterogeneity across natural systems. A further challenge in my research is thus to clarify mechanisms of shrubification acting across heterogeneous landscapes. Evidence for shrubification is not ubiquitous, with a recent synthesis indicating no change or shifts towards non-shrub dominated systems in 20% of surveyed tundra communities (Heijmans et al. 2022). Even across the North Slope of Alaska, a region with some of the first and strongest evidence for shrubification, patterns of

shrub expansion are variable (Tape et al. 2006, Bret-Harte et al. 2008, Villarreal et al. 2012, Hollingsworth et al. 2021). In fact, even the same patterns of shrubification may be the end-result of shifts in very different environmental drivers in different locations (e.g. warming, nutrient, or snow pathways, Fig. 1). Fortunately, mechanistic models of plant competition should be able to predict community-level outcomes as a function of the spatial environmental context. However, this has not yet been empirically tested across tundra ecosystems. While understanding the full mosaic of pan-Arctic landscapes is a challenge, when leveraged with direct experimental manipulations of climate drivers, natural heterogeneity offers an opportunity to test the generality of our process-based understanding.

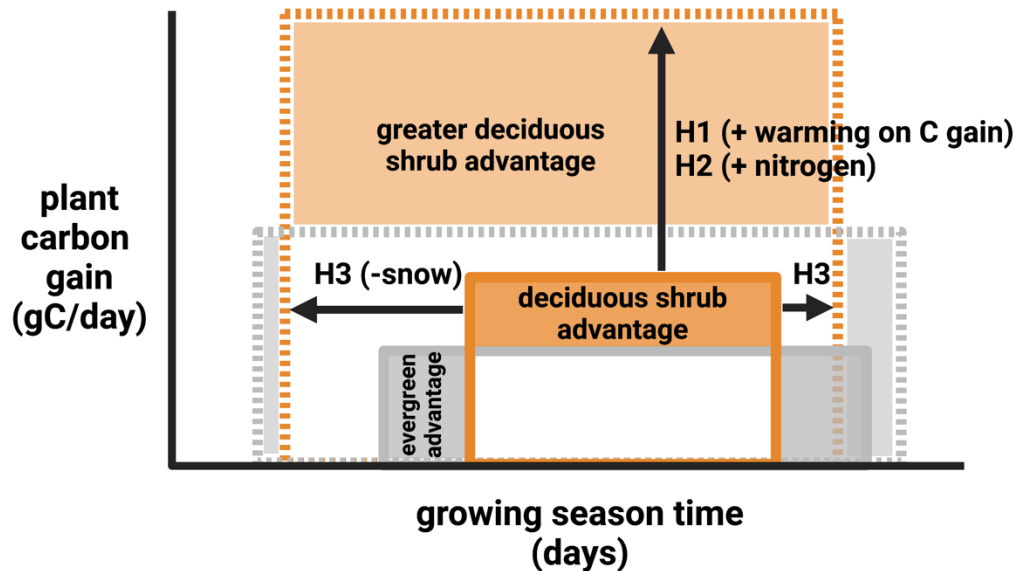
Through the research proposed here, I will address the following questions:

- **Question 1 (Describe Mechanisms):** *How do functional trait differences regulate the mechanisms of shrubification in Arctic ecosystems?*
- **Question 2 (Untangle Drivers):** *How do altered temperature, nutrient availability, and growing season length in a changing Arctic regulate shrub expansion?*
- **Question 3 (Test Generality):** *How do the mechanisms controlling Arctic shrubification vary across landscape scales?*

My approach to address these questions combines mathematical models, field manipulations of environmental drivers, and observations over natural landscapes to predict future trajectories of tundra shrubification. In particular, I test the following non-mutually exclusive hypotheses encapsulated in Figure 1:

- **Hypothesis 1 (Photosynthetic Advantage):** *Warming temperatures grant shrubs a disproportionate photosynthetic advantage over other tundra species (Fig. 1, arrow B)*
- **Hypothesis 2 (Nutrient-mediated Dominance):** *Increasing nutrient availability resulting from Arctic warming disproportionately benefits nitrogen-demanding shrubs (Fig. 1, arrow C, D).*
- **Hypothesis 3 (More Time to Win):** *Earlier snowmelt favors shrub expansion by extending the period over which deciduous shrubs outperform their competitors (Fig. 1, arrow A).*

To test these hypotheses, I will develop and parameterize a trait-based model of plant competition in tundra ecosystems. This model allows me to test whether between species trade-offs emerging from functional trait differences accurately describe mechanisms controlling shrubification in a warming Arctic (Q1). To empirically untangle the multiple pathways through which Arctic warming may cause shrubification (Q2), I have already established a three-year shrub transplant experiment that directly manipulates temperature, nutrient availability, and snowmelt date / growing season length. **Funding from the DDRIG will allow me to test the generality of mechanisms controlling shrubification (Q3)** by enabling the following efforts: (1) I will quantify shrub growth and the extent of shrubification across natural gradients in snowmelt date / growing season length in the Toolik region, and (2) I will conduct an additional experiment examining how shrub establishment is limited by temperature and nutrient availability at different locations within the heterogeneous landscape of the region. Proposed work funded by the DDRIG also includes the collection of two datasets on plant photosynthesis and nutrient availability complementary to Q1 and Q2. In all, the proposed DDRIG research builds on my ongoing work to clarify the mechanisms of Arctic shrubification through the synthesis of historical data with new models, additional observations across natural gradients, and new experiments informed by mathematical models of plant competition.



**Figure 2. Conceptual Diagram of Hypotheses (H1-H3).** Plant carbon gain is impacted by temperature, nutrient availability, and growing season length. In current environmental conditions (*solid lines*), the relative advantages of deciduous shrubs and evergreen competitors balance out (compare area of orange vs. grey shaded regions within *solid lines*). In future conditions (*dashed lines*), shifts in environmental factors expected with climate change (black arrows) cause disproportionate increases in deciduous shrub carbon gain relative to evergreen competitors as described in H1-H3 (compare area of orange vs. grey shaded regions within *dashed lines*).

## RESEARCH PLAN

### Study System

The proposed work will be conducted across a diverse mosaic of tundra plant communities surrounding Toolik Lake. Located in the low-Arctic in the foothills of the Brooks Range, these plant communities span wet-to-dry hillslope gradients characteristic of the vegetation types found across the North Slope of Alaska (Walker et al. 2016). This landscape is underlain by continuous permafrost with active-layer depths reaching a maximum of 0.5-1.0m at the end of August. The potential growing season — when air temperature daily averages are above 0°C — lasts around 120 days from about May 15 to September 15. Mean summer air temperatures are around 9°C (Cherry et al. 2014). Nutrient availability is highest in the spring during the thaw period. Nutrients are rapidly recycled and estimates of N-mineralization in the field are low, indicated limited nutrient availability in summer months (Schimel et al. 2004, Shaver et al. 2014). The winter snowpack begins accumulating in mid-to-late September and spring snowmelt typically occurs between mid-May and early-June. The timing of snowmelt varies considerably across years and landscapes; some late lying snowbeds melt 2-3 weeks after surrounding tundra. Toolik Field Station is the home site of the Arctic Long Term Ecological Research (ARC-LTER) established in 1980 and part of the International Tundra Experiment (ITEX) network since the 1990s.

### **Question 1: How do functional trait differences regulate the mechanisms of shrubification in Arctic ecosystems?**

### Background

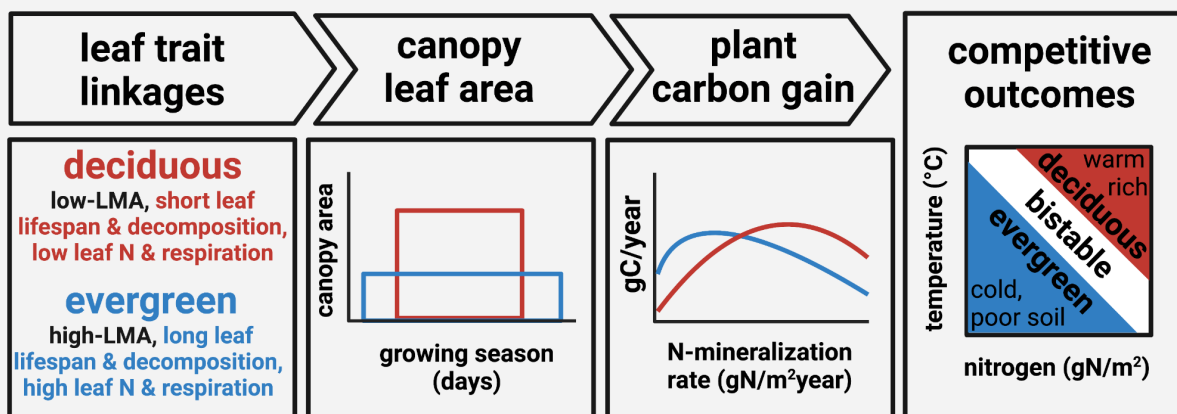
Functional trait approaches to plant communities offer a powerful way to gain insight into strategy variation across species and to predict species responses to global change (Kraft, Godoy, and Levine

2015). In theory, functional traits reflect the adaptation of species to their environment. Correlations between functional traits (e.g. leaf mass per area and leaf lifespan) reflect inherent constraints and tradeoffs in how plants acquire resources and construct their tissues. And in fact, global correlations in plant traits are maintained in Arctic ecosystems (Thomas et al. 2020).

In this proposal, I adapt an existing model of competition between plants differing in their functional traits (Weng et al. 2017) to explore the mechanisms controlling shrubification in the Arctic. In the model I expand upon here (Weng et al. 2017), plants consist of correlated suites of functional traits linked to leaf mass per area (LMA,  $\text{g/cm}^2$ ). Among these correlated traits is leaf longevity — the critical trait for understanding the dominance of deciduous Arctic shrubs over effectively evergreen competitors. This and other traits in the model determine canopy leaf area and plant carbon gain. Ultimately, the trait linkages enforce a tradeoff between carbon-use efficiency (CUE) and nitrogen-use-efficiency (NUE). Environmental conditions determine the winning strategy along the CUE — NUE trade-off. Specifically, the model predicts the winning “evolutionary stable strategy (ESS)” in a game-theoretic framework and is simple enough to analytically solve for transitions in competitive outcome with changing environmental variables including temperature and soil nutrients. Prior work successfully used this approach to provide a mechanistic explanation for patterns of deciduous and evergreen forest dominance across latitudinal gradients (Weng et al. 2017).

#### Box 1: LMA-ESS — a functional trait-based model of plant competition across climate gradients

**Leaf trait linkages.** In the model, leaf lifespan (evergreen to deciduous), nitrogen content per area, respiration, and decomposition time are modeled as increasing functions of leaf mass per area (LMA), following commonly observed empirical relationships (Wright et al. 2004, Lloyd et al. 2013, Osnas et al. 2013). For example, evergreen leaves must be thick and durable to last multiple seasons (high-LMA, longer leaf lifespan) but these high-LMA leaves will also have higher respiratory costs, take more N to construct per area, and take longer to decompose. In contrast, low-LMA deciduous leaves decompose faster and have lower respiration costs per area, but end up requiring much more N overall as leaves must be reconstructed every year (low-LMA, short leaf lifespan).



A species' leaf traits, along with the nutrient availability in soil (which fuels leaf production) determine its **canopy leaf area** per plant. Low-LMA deciduous species can build larger canopies within a single growing season, but including shoulder seasons, while evergreen species can maintain greater canopy area. Canopy leaf area in turn determines **plant species carbon gain**. Net carbon gain for a given canopy leaf area depends on the difference between species' photosynthesis and respiration. Photosynthesis is modeled using a maximum rate of photosynthesis in full sun ( $V_{max}$ ) that saturates with increasing canopy area due to self shading. Respiration is the sum of leaf respiration and canopy construction costs. The leaf traits of the dominant plants determine a **nutrient-mediated litter**

**feedback.** N-mineralization depends on total ecosystem N and the residence time of nitrogen in leaves and soils, both of which depend on leaf traits, with decomposition accelerated by higher temperatures.

**Competitive outcomes** are determined by how plants deplete light and nutrients. The competitive winner is determined as the plant that accumulates more carbon following an “evolutionary stable strategy (ESS)” in a game-theoretic framework. Environmental conditions (temperature, nitrogen availability, growing season length) determine the ESS value of LMA, and therefore the dominance of deciduous versus evergreen species. The model has analytical solutions which predict that in cold, poor soils, high-LMA evergreen species dominate. However, as soils become warmer and richer, there is a zone of bistability where one might expect patchy distributions of plants based on initial community composition. With even further warming, in the richest soils, deciduous plants dominate ubiquitously.

For more details on the model structure, see Weng et al. 2017.

The model I develop here and parameterize with the traits of Arctic plants will provide a quantitative framework in which to evaluate the mechanisms behind shrubification as outlined in Hypotheses 1-3 above (Fig. 2). Already, a generic version of the model predicts that under warming and nutrient addition, the competitive balance shifts towards carbon-efficient, nitrogen-demanding species, as exemplified by deciduous shrubs (H2, Box 1). While our proposed work will confirm that this prediction holds when the model is informed by measurements of tundra plant traits, the generic prediction is supported by observations of plant community change across Alaska and experimental evidence from Toolik Field Station. For example, observational records show that with recent warming deciduous shrubs are increasing across Alaska more than other species, and specifically more than evergreen shrubs (Macander et al. 2022). Meanwhile, in ARC-LTER experiments, deciduous species such as *Betula nana* and *Salix pulchra* are rising to dominance while evergreen species including *Rhododendron tomentosum* and *Vaccinium vitis-idea* diminish in response to long-term nutrient addition (Fig. 4).

Although the evergreen — deciduous leaf trait-axis does not encompass all trait variation distinguishing tundra shrubs from their competitors, I focus on leaf strategy as a starting point that may be expanded in further work to include woody allocation, defense traits, and microbial symbiosis (Bret-Harte et al. 2002, Christie et al. 2015, Vowles et al. 2019). For the rest of this proposal, however, I will consider the interaction between deciduous shrubs and evergreen plant competitors as a useful simplification of the competitive dynamic underlying shrubification in the system (see Fig. 2).

#### Approach:

*Model parameterization of plant trait linkages.* In order to understand how functional traits structure plant competition, I will first parameterize the LMA-ESS model using plant trait data from the Tundra Trait Team database, a synthesis of plant-trait datasets from the Arctic Data Center and Arctic LTER (including Bjorkman et al. 2018, Heskell et al. 2013, Jespersen et al. 2018, Gough et al. 2021). I will use these existing data to tailor the model to the functional relationships between LMA and other leaf traits for tundra plants including the deciduous shrubs *Betula nana*, *Salix pulchra*, and *Alnus viridis*, and their evergreen competitors including *Rhododendron tomentosum*

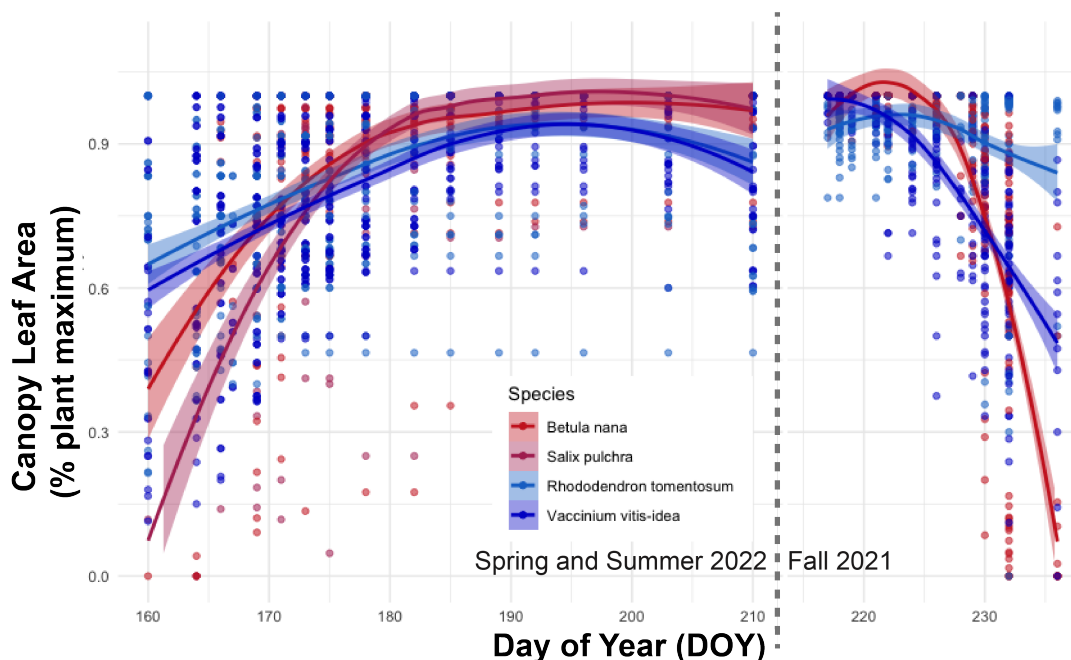
**Table 1.** Example trait values for a deciduous and an evergreen tundra species.

Plant Trait*	<i>Betula nana</i> (deciduous)	<i>Rhododendron tomentosum</i> (evergreen)
leaf lifespan (days)	80	750 (2 years)
LMA (mg/cm <sup>2</sup> )	8.12	18.1
leaf N (mgN/cm <sup>2</sup> )	0.168	0.234
leaf respiration (mgCO <sub>2</sub> /g h <sup>-1</sup> )	1.12	1.34
leaf decomposition (% mass lost in year)	30%	20%

\*Data from Bjorkman et al. Shaver et al. 1981, Bjorkman et al. 2018, Sullivan 2012, and McLaren et al. 2019.

and *Vaccinium vitis-idea*, among others (Table 1). I will first assess if an LMA-based framework of physiological trait linkages applies in tundra ecosystems (for example, if leaf N is a predictable function of LMA) by leveraging the Tundra Trait Team database. Based on prior work, I expect that established relationships will hold, although it is important to check with data specific to tundra plants. These species-level profiles of plant traits will allow us to test whether deciduous vs. evergreen species display the expected tradeoffs between growth and nutrient efficiency described by the model (Box 1, Table 1).

*Validation of plant-level model predictions.* In order to validate that model assumptions concerning leaf traits produce canopy leaf area measures in line with observations of the species in the field, I will compare the predicted canopy leaf area for each species to measurements of this variable made in summer 2022. In addition, to include species differences in the duration of their growth, I will incorporate species-specific growing season lengths based on phenological data I collected in 2021 and 2022 (Fig. 3). These data reveal the relative seasonal advantage of evergreen vs. deciduous species in “time to grow” (H3, Fig. 3). I will also validate model predictions of plant carbon gain, the key fitness variable in the LMA-ESS model (Box 1). I will use measurements of leaf-level carbon flux in conjunction with canopy phenology to estimate whole plant carbon gain on an annual basis. In particular, **with funding from the DDRIG, I will collect leaf-level carbon fluxes, particularly late in the season, when warming is most dramatic, and ecophysiological measurements are sparse.** I will measure carbon exchange for six species (3 focal deciduous shrubs, 3 most common neighbor species) over the course of the 2023 growing season to fully-establish a seasonal time-series of leaf-level carbon-exchange. Measurements will be made using the LiCOR 6400/XT portable photosynthesis chamber across tagged plants used to collect existing phenology data (Fig. 3). I will also measure the temperature dependence of photosynthesis using the LiCOR 6400-88 Expanded Temperature Control Kit, building off work by Heskell et al. 2013.

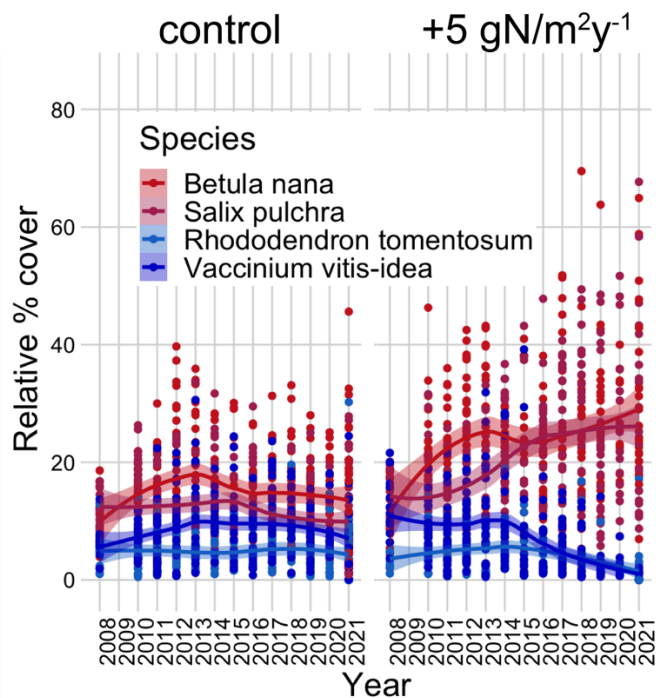


**Figure 3. Seasonal development of canopy leaf area** of four common tundra species. Points show measurements on 30 individuals per species. Lines show species-level means with 95% confidence intervals shaded. Evergreen shrubs maintain high canopy area throughout the summer and into the shoulder seasons, whereas deciduous shrubs both gain and lose leaves rapidly in the spring and fall (evergreen blue: *Rhododendron tomentosum*, *Vaccinium vitis-idea*; deciduous red: *Betula nana*, *Salix pulchra*). These data were collected in spring & summer 2022 and fall 2021 (excluding *Salix pulchra*).



*Model prediction of mechanisms of shrubification:* To test Hypotheses 1 (photosynthetic advantage), I will simulate competition between deciduous shrubs and evergreen competitors using the tundra-validated LMA-ESS model under a scenario with increasing temperature, but unchanged nutrient availability and snowmelt date. This warming-alone simulation will incorporate temperature-dependent maximum photosynthetic rates ( $V_{max}$ ) parameterized to individual species using the ecophysiological data collected above. Competitive dominance of deciduous shrubs in this scenario would indicate that photosynthetic advantage with warming could be an important mechanism of shrubification.

To directly test Hypotheses 2 (nutrient-mediated dominance), I will run model simulations under a scenario with increasing nutrient availability, but unchanged temperature and snowmelt date. Fortunately, I can validate N-addition simulations with data from ARC-LTER nutrient addition experiments (Gough et al. 2021). Specifically, I will compare model simulations of deciduous and evergreen distributions with a 15 year dataset of relative species cover from a fertilization gradient experiment with five levels of nutrient addition (control, +0.5, +1, +2, +5, and +10 gN/m<sup>2</sup>y<sup>-1</sup>). The data from this experiment show



**Figure 4. Changes in plant community composition over 2 decades of ARC-LTER experimental nutrient addition.** Points show species relative % cover in a 1 m<sup>2</sup> quadrat, with 8 quadrats measured per treatment plot. Lines show species-level means with 95% confidence intervals shaded. Data from two out of six treatments are shown here. Across treatments, deciduous shrubs rise to dominance at increasing levels of nutrient addition. Data were collected by ARC-LTER technicians over 16 years (Gough et al. 2021).

deciduous shrub abundance increases with progressively higher levels of nutrient addition, supporting N-mediated dominance as an important mechanism of shrubification (Fig. 4).

Similarly, to test Hypotheses 3 (more time to win), I will simulate the model with early and delayed snowmelt dates, holding temperature and nutrient availability constant. Model simulations of delayed snowmelt can be checked with plant community composition data from ITEX-AON snow fence experiments established in 1994 and 2005 (Schimel et al. 2004, DeMarco et al. 2011).

The simulation scenarios described above independently change a single factor, when in fact we expect all three factors synergistically mediate mechanisms of shrubification through multiple pathways (Fig. 1). Once the model is parameterized and validated for Arctic tundra, I can easily run simulations that alter multiple drivers in factorial combination and quantify their direct and interactive contributions to “shrubification” outcomes. In addition, a powerful advantage of this modeling approach is that I can analytically solve for transitions in competitive outcome at threshold-values of changing

environmental variables. These approaches allow me to both disentangle the multiple pathways through which Arctic warming mediates mechanisms of shrubification and quantify which shrubification mechanisms may be the most impactful in different scenarios of Arctic change.

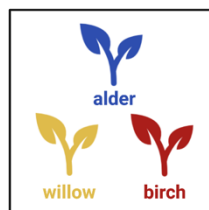


## Question 2: How do altered temperature, nutrient availability, and growing season length in a changing Arctic regulate shrub expansion?

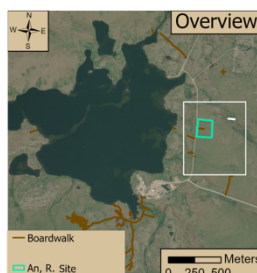
### Background

Work proposed under Question 1 will generate theoretical predictions of the primary mechanisms controlling shrub expansion in the Toolik region. Experimentally testing those predictions is the aim of work proposed under Question 2. To address this aim, I will directly manipulate temperature, nutrient availability, and snowmelt date, the three factors hypothesized in H1-H3 to regulate deciduous shrub expansion. More specifically, I will examine the individual and combined effects of these factors on a key life stage regulating shrub expansion (establishment) and do so for three shrub genera actively expanding across Alaska (Salmon et al. 2019, Tape et al. 2006). This experiment represents an advance over past manipulations of these factors at Toolik Field Station, which either lacked sufficient replication, manipulated different factors in different locations, and/or did not include the interactive effects of all factors studied here. Moreover, with the prior experiments relying on the response of extant shrub individuals varying in life stage and taxonomy, inferring a standardized response to treatment and comparing the response of different deciduous shrub species is difficult.

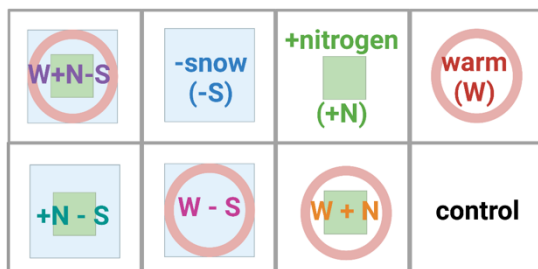
### Experimental Design



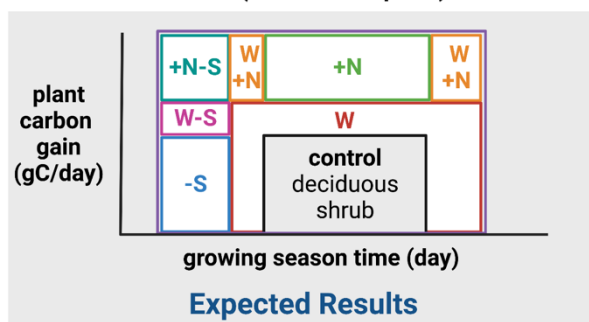
Plot (1.5m x 1.5m)



Site (10 blocks)



Block (8 treatment plots)



### Approach

In summer 2022, I designed and implemented a shrub establishment experiment that directly manipulates temperature, nutrient availability, and snowmelt. This experiment tests how these factors mediate mechanisms of shrubification — specifically the establishment of three northern deciduous shrub species (*Alnus viridis*, *Betula nana*, and *Salix pulchra*) known to be increasing across Alaska. Focusing on the establishment stage not only makes the experiment logistically feasible, but it minimizes the complexity introduced by feedbacks between established shrubs and these mediating factors. In this context, this experiment is specifically targeted towards the mechanisms regulating shrub expansion into currently unoccupied habitat.

**Figure 5. Untangling the multiple pathways through which Arctic warming may impact shrub expansion.** Treatments manipulate temperature, nitrogen, and snow. Within blocks, blue squares indicate snow removal (-S), green squares nitrogen addition (+N), and red circles warming chambers (W). Site location with respect to Toolik Lake is shown in light green in the top right. A conceptual diagram of expected experimental impacts on shrub carbon gain given hypothesized determinants of shrub growth (Fig 2. H1-H3) is shown in gray (bottom).

**Experimental Design.** In summer 2022, I selected 10 replicate blocks along a tundra hillslope in which I placed 8 treatment plots. In each block, half the plots were randomly assigned the snow removal (-S) treatment. Within each half-block, warming (W) and nutrient addition (+N) treatments were randomly assigned in factorial combination (Fig. 5). These treatment manipulations will be conducted from 2023-

2025 over three growing seasons. In snow removal plots, I will shovel snow off plots in late April/early May with a target advancement of snowmelt date by two weeks, equivalent to a 12% increase in current growing season length (Cherry et al. 2014). Exact timing will depend on annually variable snowpack conditions. In warming plots, I will deploy open top chambers (OTCs) each spring after snowmelt (natural or imposed). OTCs are clear, polycarbonate structures that increase air temperature by 1.5-2 °C through passive warming (Hollister et al. 2022). For all plots each year, OTCs will be removed post senescence in mid-September. In nitrogen addition plots, each spring following snowmelt I will add granular ammonium chloride (NH<sub>4</sub>Cl) and sodium nitrate (NaNO<sub>3</sub>) fertilizers for a combined rate of +5 gN/m<sup>2</sup>y<sup>-1</sup>. This rate is within estimates of increases in N-mineralization expected with Arctic warming from prior studies (Prager et al. 2020). In each plot, I planted focal individuals of three tundra shrub species (*Betula nana*, *Salix pulchra*, *Alnus viridis*) in summer 2022. These focal shrubs (20-40 cm height) were collected broadly across the Toolik landscape, with care taken to minimize disturbance to roots and stems.

*Response variables.* Beginning in summer 2023, I will collect plant trait data and nondestructively measure the biomass of the focal shrubs planted into the experimental matrix (Fig. 5). I will quantify allometric relationships between leaf number, leaf size, plant height, and biomass from individuals collected outside the experiment to infer the biomass growth of our transplanted shrubs. In order to assess shrub response to treatment relative to competitors, I will also measure the response of neighboring plant species in each plot. Within 20 cm of each focal shrub, I will identify the competitor species present, and for half of the plots, I will quantify relative cover using point-intercept methods. This experiment will run for three summers through Fall 2025, at which point all transplanted shrub individuals will be harvested and weighed. I will use mixed effect models to evaluate the main and interactive effects of each treatment on shrub growth and the growth of their neighbors. By measuring snowmelt date in all plots with timelapse cameras, I will have the opportunity to treat this factor as a continuous variable in further analyses.

Finally, by measuring the growth, phenology, and leaf area of the shrubs and background competitors in response to treatment in the same year and same general location, I will confirm that the model parameters informing the model under Question 1 apply under this more controlled context. Specifically, I will make these measures on the three focal shrubs and the three most common neighbor species within the experimental set-up.

*Untangling Multiple Pathways: field data collection of nutrient availability.* Although the nutrient addition treatment allows me to evaluate how this factor regulates shrub establishment independent of other changing factors, the other manipulated drivers in this experiment snowmelt date and warming — may affect shrub establishment through their effects on nutrient supply to the plants. The experimental design positions me to evaluate these pathways. I have installed iButtons at 10 cm depth in all 8 treatments across 3 blocks to assess how snowmelt timing and warming affect soil temperature, a variable expected to control nitrogen mineralization. With funding from the DDRIG, I will directly measure nutrient availability across treatments using rhizons, metal needles installed in the soil to collect pore water over the course of two growing seasons. More specifically, I will collect soil pore water in half of the plots at three time points over the course of the growing season (spring post-thaw, peak season, end of season) each year. Concurrent with each porewater collection, I will record active layer depth to assess treatment impacts on permafrost thaw (Fig. 1, arrow E). Samples will be frozen and sent to an analytical laboratory (e.g. UC Davis Analytical Laboratory) to measure concentrations of NO<sub>3</sub> and NH<sub>4</sub>. Mixed effects models will evaluate how each treatment affects the availability of these nutrients.

### Expectations

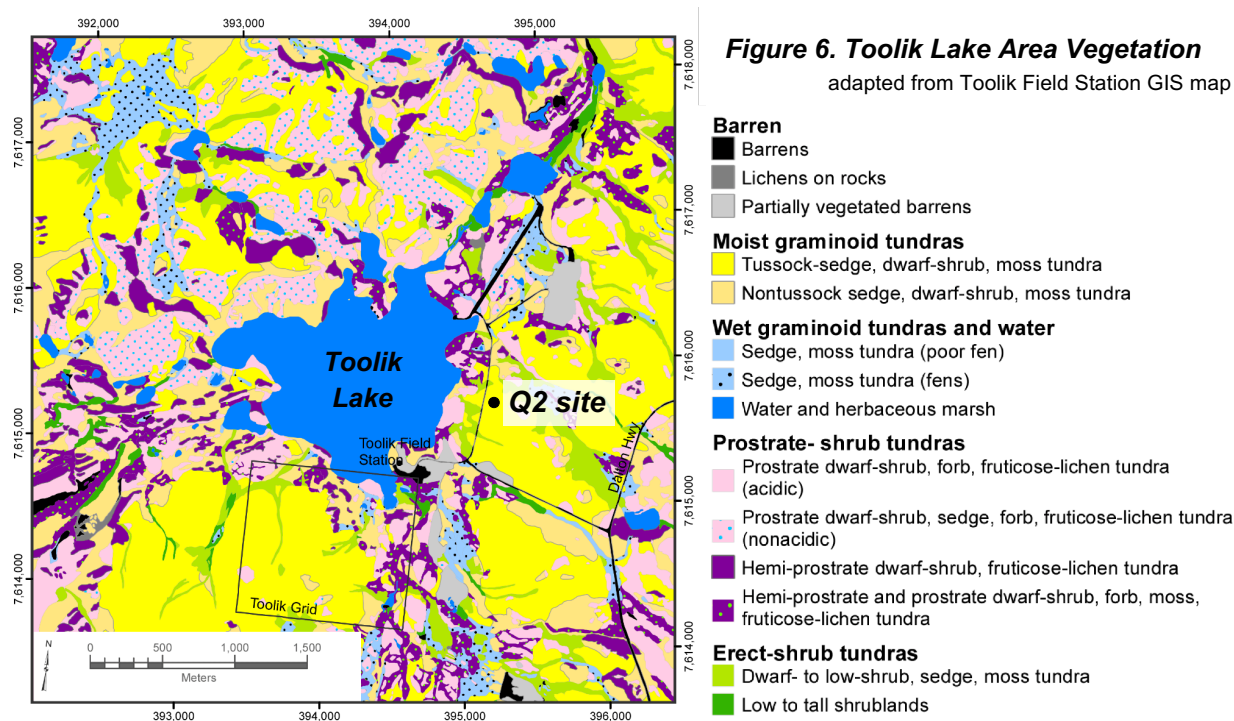
If direct physiological advantages with warming are paramount, I expect to see limited additional effects of snow and nutrient manipulations on shrub growth (H1; Fig. 1, arrow B). If the primary impact of

warming on shrubification is via nutrient-mediate dominance, we expect nutrient addition to be most effective at enhancing shrub establishment and growth (H2; Fig. 1, arrows C, D). Similarly, if the most important impact of Arctic warming on shrubification is via earlier snowmelt lengthening the growing season, then we expect experimental snow removal will be most effective at enhancing shrub establishment and growth (H3; Fig. 1, arrow A). Furthermore, if these are mechanisms that grant shrubs an advantage over other species, focal shrubs should respond more than neighboring species to the same manipulations. These data and analyses will help untangle the multiple pathways through which Arctic warming impacts shrubification processes.

### **Question 3: How do the mechanisms controlling Arctic shrubification vary across landscape scales?**

#### **Background**

Work proposed under Question 2 manipulates multiple environmental factors at a single site as a direct empirical test of mechanisms controlling the expansion of shrubs in the Toolik region. An important challenge, and the aim of work proposed in Question 3, and funded by the DDRIG, is to describe mechanisms of shrubification that have predictive power over heterogeneous Arctic landscapes. Arctic landscapes encompass substantial variation in snowmelt date and nutrient availability, reflected in significant spatial variation in tundra plant communities (Fig. 6). In the context of the full extent of environmental and biological variation across the Arctic and observed around Toolik Lake, we do not necessarily expect the same factors to limit shrubification across all locations. In fact, results from ARC-LTER and ITEX-AON manipulations of temperature and nitrogen availability sometimes favored herbaceous species, supporting the context dependence of shrubification (Bret-Harte et al. 2008, personal observation). Fortunately, the modeling framework developed under Question 1 predicts deciduous shrub dominance at different locations along axes of environmental variation (e.g. increasing temperature and nitrogen availability, Box 1 panel 4). Working with this framework, we are likely to find that different factors limit shrub growth and expansion in different environmental contexts across the landscape.



## Approach

The aim of work described under Question 3 is to test the generality across natural landscapes of the mechanisms of shrubification inferred by the modeling and experimental approaches described thus far. To provide context for this effort, I will survey snowmelt (with time lapse cameras) and nutrient availability (with soil extractions) across a mosaic of tundra landscapes in the Toolik region. These data will be used in combination with the environment-dependent model predictions to identify locations where we might expect different mechanisms to favor shrubs in response to warming. For example, if the parameterized model and experimental manipulation both suggest that longer growing seasons favor deciduous shrubs over evergreen competitors, at locations with relatively little spring snow we might expect further reductions in snow to have little effect in comparison to enhanced nutrient availability. I will use this information to identify the locations where we (1) expect to observe more or less shrubification in the natural landscape and (2) deploy an experiment mirroring that described under Question 2 testing the site-specific limitations on shrubification.

*Natural observations of shrub populations.* I will quantify natural patterns of shrubification across the landscape gradients in snowmelt / growing season length and nutrient availability around the Toolik region. At each of 20 locations stratified across the community types shown in Fig. 6, I will measure environmental variables as described in the prior paragraph and characterize natural shrub populations with the following measures: maximum height and % cover along a 20 m transect, and seedling density in ten 1 m<sup>2</sup> plots spaced along the transect. These observations will be carried out in summer 2023. I will then regress the degree of shrubification against the snowmelt date (averaged over several years) and nutrient availability of the sites to test how these factors regulate shrub dominance. Relationships matching those predicted by the empirically parameterized plant trait-based model under Question 1 would support the broad applicability of the theoretical mechanisms to the habitat.

*Landscape extension of the shrub expansion experiment.* Of the 20 locations surveyed for environmental variables and shrubification, I will select 6 locations expected to differ in the degree to which temperature and/or nutrient availability regulate shrubification. At each of these 6 sites, I will establish a factorial manipulation of temperature and nutrient availability, in a blocked design replicated ten times. Snow removal is too logistically challenging to experimentally deploy at this scale, hence the smaller design, but I plan to include sites varying naturally in snowmelt date to also include this variable in the design. I will randomly assign the warming and nutrient addition treatments to plots within each block, and manipulate these factors during the 2024 and 2025 growing seasons following the same protocols described under Question 2. I will transplant three shrub species into each plot in fall 2023 before manipulations begin and measure their fate for two subsequent growing seasons, following the same protocols as described under Question 2. Last, I will conduct the same statistical analysis of shrub transplant growth as described under Question 2 but use site-level environmental data as predictors of the response to treatment.

## Expectations

The natural patterns of shrubification, and its variation across the landscape will point to factors limiting or promoting shrubification in nature, helping to validate the theoretical predictions and experimental results under Questions 1 and 2. For example, high rates of shrub growth and establishment in nutrient-rich sites would provide further evidence for nutrient based mechanisms as a current limit of shrub dominance (Hypothesis 2). With the experiment manipulating mechanisms of shrubification in different locations, we might find that the same factor regulates shrub establishment in all locations, supporting a general explanation for this process across the landscape. If alternatively, I find that different factors limit shrub establishment in different locations, I can test whether this context dependency is predicted by our theory. For example, I might find that nutrient addition most substantially benefits shrub establishment in

sites that already have longer growing seasons, a prediction that may emerge from the model. In this way, both the observational patterns and the experiment allow us to leverage natural heterogeneity to test the generality of controlling mechanisms of shrubification.

### **INTELLECTUAL MERIT**

While ecosystem scientists have long recognized that biotic and abiotic processes are inexorably linked in the Arctic, shrubification has not commonly been studied with an integrated theoretical and experimental approach to competition. This community ecology perspective offers new and necessary sets of quantitative and conceptual tools with which to leverage historical data, experimental infrastructure, and natural observations. The mechanistic modeling approaches based on trade-offs in plant traits developed here offer a powerful tool to quantify and predict mechanisms of shrubification from first principles. Meanwhile, our direct manipulation of the causal variables behind shrubification, coupled to observations of tundra plant community change along natural environmental gradients offers the opportunity to validate the mechanisms of shrubification inferred by models.

Furthermore, conducting research in rapidly shifting Arctic ecosystems directly tests the strengths and limitations of our current theoretical frameworks for generating insight into contemporary ecological change. The analysis and validation of physiologically-based models, as proposed here, can identify the thresholds in environmental factors that shift competitive dominance, an approach that can be applied to ecosystems undergoing rapid global change beyond the Arctic. In all, the proposed work stands to deepen our understanding and predictive capacity of how future climates shape plant community change.

### **BROADER IMPACTS**

**Climate change resilience.** At a global scale, understanding the physiological and environmental interactions driving ecological dynamics offers a pathway to better understand contemporary and future patterns of Arctic shrubification, a process with major implications for the global carbon cycle. Information on the mechanisms of shrubification has great potential to enhance the ability of Earth System Models, such as GFDL LM4, to predict current and future Arctic carbon budgets. At a more local scale, understanding future trajectories of vegetation change is critically important to the food security of many communities across Alaska. In this context, I will develop a public-facing R Shiny interface for model simulations that allows individuals to interactively examine how different “mechanisms of shrubification” influence maps of climate change impacts on plants important to traditional subsistence lifestyles. These include salmonberries (Iñupiaq/Latin name: *aqqiqutaq/Rubus chamaemorus*), crowberries (*paungaq/Empetrum nigrum*), blueberries (*asiavik/Vaccinium uliginosum*), and cranberries (*kimmigñaq/Vaccinium vitis-idaea*). Finally, the research infrastructure funded by this project will generate tools that can be an asset in starting respectful and trusting partnerships between scientists and local communities in Alaska.

**Publicly Available Data and Methods.** All data and code generated by this project will be contributed to publicly accessible pan-Arctic databases essential to my work — specifically the Tundra Trait Team and the Arctic Data Center. I will also develop a tutorial on running simulations of plant competition models in R that aims to be broadly accessible to the Arctic research community. My goal is for these modeling approaches to become accessible tools for all Arctic researchers tackling the complex challenge of understanding complex ecosystems in a rapidly changing world.

**Undergraduate Mentoring.** The High Meadows Environmental Institute (HMEI) administers an internship program that embeds Princeton undergraduate students in ongoing research projects. Already, I have mentored one HMEI intern in summer 2022, who collected data contributing to Fig. 3. I plan to mentor two HMEI interns in summer 2023 and subsequent field seasons (see Table 2).



**K-12 Educational Outreach.** The results from the research will be incorporated into an LTER Data Nugget distributed to K-12 schools and AKDaTUM module for college courses. These educational modules extend the reach of outreach efforts by leveraging existing networks and established infrastructure across the U.S. (LTER) and Alaska.

**Training and collaboration.** This project will support the training and mentoring of the PhD student leading the work, and grow collaborations between students at Princeton University and institutions across the Arctic LTER community. This project will support the training and mentoring of one research technician from Alaska.

**Table 2. Project Timeline.** Grey blocks denote field season work and blue blocks denote winter work.

Activity	Objective	Year 1 (2023)	Year 2 (2024)	Year 3 (2025)
Model Development & Validation	Q1	Winter	Winter	
Photosynthesis Measurements**	Q1	June, July, August		
Climate (W, +N, -S) Manipulations	Q2	May-September	May-September	May-September
Soil Nitrogen Sampling*	Q2	June, July, August	June, July, August	June, July, August
Shrub & Competitor Sampling	Q2-Q3	July	July	July, September
Natural Observations	Q3	June, July		
Landscape Extension of (W, +N) Manipulations*	Q3	September (transplanting)	May-September	May-September
Analysis & Writing	Q1-Q3		Winter	Winter
R Shiny & Outreach	Broader Impacts		Winter	Winter

\* Indicates effort by the research technician for which DDRIG funding is requested.