



T.R.

NİĞDE ÖMER HALİSDEMİR UNIVERSITY

GRADUATE SCHOOL OF NATURAL AND APPLIED SCIENCES

DEPARTMENT OF AGRICULTURAL GENETIC ENGINEERING

COMPARISON OF DEVELOPMENT AND PHYSIOLOGY OF *Deschampsia*
antarctica ECOTYPE COLLECTION GROWN IN TWO DIFFERENT
TEMPERATURES

YKBAL AVEZBAYEVA

June 2024

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Master Thesis

Supervisor

Prof. Dr. Zahide Neslihan ÖZTÜRK

June 2024

The study titled “**Comparison of development and physiology of *Deschampsia antarctica* ecotype collection grown in two different temperatures**” is presented by **Ykbal AVEZBAYEVA** under the supervision of Prof. Dr. Zahide Neslihan ÖZTÜRK and accepted as a Master thesis by the jury at the Department of Agricultural Genetic Engineering of the Niğde Ömer Halisdemir University, Graduate School of Natural and Applied Sciences.

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THESIS DECLARATION

I hereby declare that thesis was written by myself. Every detail in this thesis document was gathered, organized and presented in accordance with the scientific and academic guiding principles as well as the thesis writing regulations of the institute of natural and applied sciences, Niğde Ömer Halisdemir University. All information sourced from other researchers have been duly referenced.



Ykbal AVEZBAYEVA

SUMMARY

COMPARISON OF DEVELOPMENT AND PHYSIOLOGY OF *Deschampsia antarctica* ECOTYPE COLLECTION GROWN IN TWO DIFFERENT TEMPERATURES

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Deschampsia antarctica E. Desv, is perennial plant belonging to the Poaceae family that thrives in terrestrial ecosystems of Antarctica. Its remarkable adaptability to harsh conditions has captured the interests of scientists. There are numerous studies focusing on the effect of temperature on plant development, these studies are often limited to collections from a single site. Our study focused on investigating how different temperatures (12°C and 22°C) impact the ecotypes of *D. antarctica* which were collected from diverse ecosystems of Antarctica: Ardley, Galindez, King George, Lagotellerie (200 m), Lagotellerie (5 m), Livingston and Ridge islands. Results showed various growth performance among ecotypes. Samples collected from Livingston island consistently exhibited robust growth, while samples collected from Galindez, King George and Ridge islands showed significant height increases at 22°C. The study highlights how habitat variations shape plant development and resilience to temperature fluctuations. Understanding these temperature responses is crucial for informing conservation efforts in Antarctica, as it provides insights into how different ecotypes may adapt and survive under changing environmental conditions.

Keywords: *D. antarctica*, development, physiology, different ecotypes, influence of temperature

ÖZET

İKİ FARKLI SICAKLIKTA BÜYÜTÜLEN *Deschampsia antarctica* EKOTİP KOLEKSİYONUNUN GELİŞİMSEL VE FİZYOLOJİK KARŞILAŞTIRILMASI

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Deschampsia antarctica E. Desv, Poaceae familyasına ait, Antarktika'nın karasal ekosistemlerinde yetişen çok yıllık bir bitkidir. Zorlu koşullara uyum sağlama yeteneği bilim insanlarının ilgisini çekmiştir. Bitki gelişimine sıcaklığın etkisi üzerine yapılan çalışmalar genellikle tek bir yerden alınan örneklerle sınırlıdır. Çalışmada, Antarktika'nın Ardley, Galindez, King George, Lagotellerie (200 m), Lagotellerie (5 m), Livingston ve Ridge adaları gibi çeşitli ekosistemlerinden toplanan yedi farklı *D. antarctica* ekotipinin 12°C ve 22°C gibi farklı sıcaklıkların etkilerini araştırılmıştır. Sonuçlar, ekotipler arasında çeşitli bir büyüme performansı sergilemiştir. Livingston adasından toplanan örnekler tutarlı bir şekilde sağlam büyüme gösterirken, Galindez, King George ve Ridge adalarından toplanan örnekler ise 22°C de belirgin artış göstermiştir. Çalışma, habitat farklılıklarının bitki gelişimini ve sıcaklık dalgalanmalarına karşı dayanıklılığının asıl şekillendirdiğini vurgulamaktadır. Bu sıcaklık tepkilerini anlamak, farklı ekotiplerin değişen çevresel koşullara uyum sağlayabileceği ve hayatta kalabileceği konusunda fikir sunduğundan Antarktika'daki koruma faaliyetlerini doğrulamak için son derece önemlidir.

Anahtar Sözcükler: *D. antarctica*, gelişim, fizyoloji, farklı ekotipler, sıcaklığın etkisi

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SYMBOLS AND ABBREVIATIONS

Symbols	Descriptions
cm	Centimeter
°C	Degree Celcius
g	Gram
h	Hour
L μ L	Liter Microliter
M	Molar
mL	Milliliter
mm	millimeter
min	Minute
nm	nanometer
%	Percentage
rpm	Revolutions per minute
V	Volume
W	Weight

Abbreviations	Descriptions
ANOVA	Analysis of Variance
ADF	Actin-depolymerizing factor
CV	Coefficient Variance
cDNA	Complementary Deoxyribonucleic Acid
chl	Chlorophyll
DaADF	Daucus carota Actin-depolymerizing factor
EST	Expressed Sequence Tag

GLM	General Linear Model
H ₂ O	Water
rDNA	ribosomal Deoxyribonucleic Acid
UV	Ultraviolet
UV-vis	Ultraviolet visible



CHAPTER I

INTRODUCTION

Deschampsia antarctica E. Desv, commonly known as Antarctic hairgrass, is a remarkable plant species native to the extreme environment of Antarctica (Smith, 1980). Its adaptations to the harsh conditions such as low temperature, high light intensity and increasing UV radiation as a result of the ozone layer thinning of the Antarctic continent make it a fascinating subject of scientific inquiry (Sánchez-Venegas et al., 2009). The survival of *D. antarctica* was enhanced when growing associated with *Usnea antarctica* (Du Rietz) cushions, highlighting the complex ecological relationship between lichens in the Antarctic region. Lichens, such as *U. antarctica* play a crucial role in polar ecosystems as they offer microhabitats and nutrient sources for other organisms (Øvstedal et al., 2001; Molina-Montenegro et al., 2013; Casanova et al., 2014). There are already some indications that climatic changes have been implicated in affecting the local population density and distribution of *D. antarctica* and *Colobanthus quitensis* (Kunth) Bartl., the two vascular plant species that adapted along the Antarctic Peninsula (Bokhorst et al., 2007; Cavieres et al., 2016). *D. antarctica* is the most abundant and widely distributed throughout maritime Antarctica compared to *C. quitensis* (Smith et al., 2003; Sáez et al., 2018). Since *D. antarctica* species has unique adaptive abilities, they can survive and reproduce in the extreme climatic conditions of Antarctica. It can grow from 0.5 to 25 cm in height. Young stems are enclosed in a shell, and the leaves are sessile with linear lead blades. *D. antarctica* can form siblings of the correct shape with a lifespan of up to 35-40 years. Aging and death of old plants are often observed in the central part of the sibling, and the cycle of formation of a new sibling begins when the plant takes root in a new place (Alberdi et al., 1991; Smith et al., 2003).

Antarctica is located in the Southern Hemisphere around the geographical South Pole, which occupies 10% of the Earth's surface. It is isolated from other continents due to geographical remoteness, the presence of an oceanic polar frontal zone and the fact that it is surrounded by the Antarctic Circumpolar Current (Smith, 1984; Convey, 1996). More than 95% of this region is blanked by a massive ice sheet that

can reach up to 3,000 meters in thickness. This continent is largely devoid of any complex life forms, with only prokaryotes (simple microorganisms) and fungi, along with some basic aquatic creatures residing in subglacial reservoirs, managing to survive in such extreme conditions. The majority of terrestrial life in this area is concentrated in the northern coastal regions and on nearby islands.

The Antarctic biome is typically divided into three distinct biogeographical zones. These zones are the 1) Subantarctic, 2) Maritime and 3) continental Antarctic zones (Figure 1.2, Díaz et al., 2019). However, when it comes to terrestrial vegetation, it is primarily found in the Subantarctic and maritime Antarctic zones which offer the most favorable climate and growing season, because it is situated closest to a biologically diverse continental landmass, southern South America, which could potentially serve as a vital source of viable immigrant propagules (Longdon, 1979; Smith, 1984; Smith, 1991).

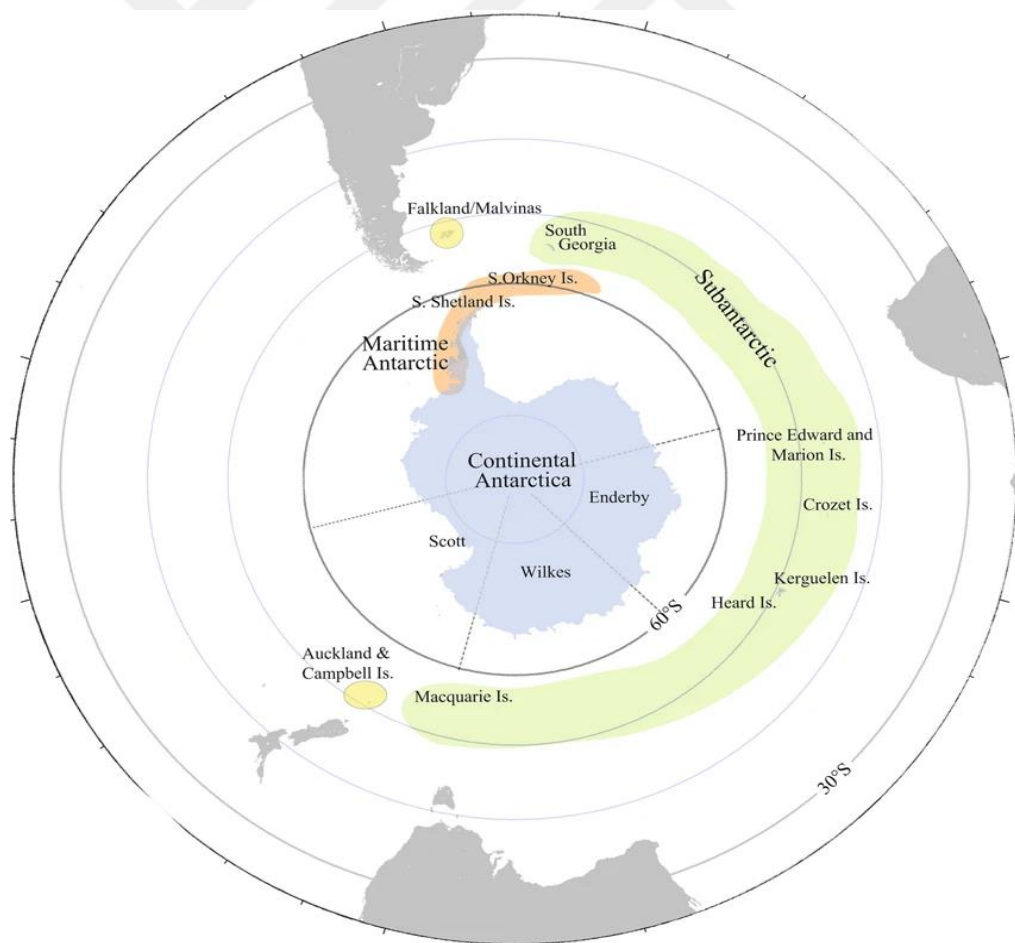


Figure 1. 1. Map of Antarctica. Subantarctic zone (green), Maritime zone (orange) and Continental Antarctic zone (in blue color) (Díaz et al., 2019)

These ecosystems are notable for their exceptional adaptability to some of the most severe environmental conditions. These challenges include intense winds that desiccate the soil and vegetation, rapid temperature fluctuations, frequent, freeze-thaw cycles, and an average daily temperature during the growing season that drops below freezing, necessitating robust cryoprotective mechanisms for plant survival. Furthermore, these areas experience high levels of ultraviolet radiation adding to the environmental stressors that life must contend with (Kyryachenko et al. 2005; Parnikoza et al. 2011; Matvieieva 2013; Cavieres et al., 2016). Nevertheless *D. antarctica* exhibits remarkable adaptations to Antarctica's extreme conditions, including cold tolerance, and UV radiation resistance (Robinson et al., 2003). Besides surviving within the Antarctic ecosystem, *D. antarctica* plays a crucial ecological role. It serves as a food source for herbivores, influences soil structure, and impacts cycling (Convey et al., 2006). The fascinating adaptations of this plant to extreme environmental conditions have galvanized scientific curiosity worldwide. During the third National Antarctic Scientific Expedition, Prof. Dr. Sedat Serçe collected *D. antarctica* samples and brought alive plants back to Türkiye. Plant samplings were performed from six islands representing two different climatic conditions observed in Antarctica (Antarctic tundra and Antarctic seaside) as categorized in Table 1.1. *D. antarctica* plant samples were directly transferred to soil in Niğde Ömer Halisdemir University, Ayhan Şahenk Faculty of Agricultural Sciences and Technologies, Agricultural Genetic Engineering Department. Plant samples were kept at growth-chambers at 12°C from there on and multiplied depending on the tillering frequency. This pivotal achievement not only represents a significant task in its own right, but also serves as a vital foundation that will enable us to conduct a much broader and more detailed examination related to *D. antarctica* and Antarctic continent in the future.

The primary objective of this study is to compare the development and physiology of these seven ecotypes and shed light on the effects of two different temperatures on the growth, physiology, and morphology of *D. antarctica* plants. Notably, this study marks the first-ever study conducted on unique collection of ecotypes from Antarctic regions carried out in Niğde Ömer Halisdemir University, Ayhan Şahenk Faculty of Agricultural Sciences and Technologies, Agricultural Genetic Engineering Department.

Table 1. 1. Sites where plant samples were collected by Prof. Dr. Sedat Serçe during the 3. National Antarctic Scientific Expedition

	Name of Island	The altitude of Sampling	Attribute of the Ecosystem	
1	King George	15 meter	Antarctic seaside	
2	Ardley	5 meter	Antarctic seaside	
3	Livingston	5 meter	Antarctic seaside	
4	Galindez	10 meter	Antarctic seaside	
5	Ridge	100 meter	Antarctic tundra	
6	Lagotellerie	5 meter	Antarctic seaside	
7	Lagotellerie	200 meter	Antarctic tundra	

CHAPTER II

LITERATURE REVIEW

2.1 *Deschampsia antarctica* E. Desv.

Deschampsia antarctica E. Desv., commonly known as Antarctic hairgrass, is a remarkable perennial plant belonging to the Poaceae family that survives in the challenging terrestrial ecosystems of Antarctica (Smith, 1980). This Antarctic hairgrass, exhibit distinct morphological features that vary depending on environmental conditions. Under favorable conditions it has dark green and well-developed leaves and study and upright stalks, but under adverse environmental conditions, the plant has a much smaller size and loose and olive-colored locks (Convey, 1996). The taxonomic classification of *D. antarctica* is outlined below (Anonymous, 2024).

Kindom	: Plantae
Phylum	: Angiosperms
Class	: Monocots
Order	: Poales
Family	: Poaceae
Genus	: <i>Deschampsia</i>
Species	: <i>Deschampsia antarctica</i>

The plant is characterized by noticeable variability in the size of the tuft and the lengths of the length of the culm. As a rule, it forms compact tufts, but under ideal conditions these tufts can grow into dense tussocks. The culms of plant are short, thin, straight and smooth, while the leaf sheaths on the vegetative shoots are smooths, fused together and straw-like, with pronounced coarse veins on the upper side. The ligule can reach a length of up to 1.5 mm. The number of inflorescences is less than the vegetative shoots, but they are longer. Depending on the soil fertility of the soil, the length of the shoots can vary significantly. The panicles have a noticeable curve

at the base of the inflorescence axis, and they disintegrate as they mature. Spikelets are relatively large, measuring around 6 mm in length, and typically contain two flowers. The callus decorated with short hairs, and mature flowers usually remain closed, indicating self-fertilization inside the flower. The leaves of *D. antarctica* are leathery and tightly folded in a bud. Cross-section reveal three primary ribs, occasionally four or five. The outer walls of the leaf epidermal cells are protected by a thick cuticle, which enhances the plant's resilience (Barcikowski et al., 2001).

D. antarctica is a diploid ($2n=26$) species that has been well adapted to the extreme conditions of Antarctica. It can perform various metabolic processes at very low temperatures and high levels of ultraviolet radiation and drying, which allows plant to develop, including growth and flowering in harsh conditions (Bravo et al., 2001). In 2020, Díaz, Serçe et al. presented the first genome size analysis focused on samples collected from the Antarctic Peninsula and surrounding islands for both *C. quitensis* and *D. antarctica* species (Figure 2.1, Díaz, Serçe et al., 2020). This study investigated the genetic diversity and evolutionary dynamics of *C. quitensis* and *D. antarctica* (Table 2.1, Díaz, Serçe et al., 2020). The observed diploid and tetraploid ploidy levels of these species, respectively, highlight their adaptive strategies to the extreme Antarctic conditions. Although minor genome size deviations hint at B-chromosomes in *D. antarctica*, overall genome size consistency across populations reflects a recent colonization history of the continent (Díaz, Serçe et al., 2020). B-chromosomes, known for their smaller size and variable presence among individuals and populations, defu Mendelian inheritance patterns and can impact traits such as fertility, development, or behavior due to their non-essential genetic content. Studying the deviations of this chromosome in *D. antarctica* provides valuable insights into its genetic composition and potential adaptive strategies.

Table 2. 1. Average estimations of 2C genome size (pg), standard deviation (SD) and average percentage of coefficient of variation (CV) for the *Deschampsia antarctica* and *Colobanthus quitensis*

Species	Population	2C (SD)	CV (%)	Standard
<i>Deschampsia antarctica</i>	Ardley	10.65 (0.03)	3.17	<i>Pisum sativum</i>
	Galindez	10.61 (0.02)	2.95	
	King George	10.63 (0.03)	3.34	
	Lagotellerie (200m)	10.62 (0.04)	3.13	
	Lagotellerie (5 m)	10.71 (0.14)	3.17	
	Livingston	10.59 (0.02)	3.63	
	Ridge	10.63 (0.04)	2.60	
<i>Colobanthus quitensis</i>	Galindez	2.08 (0.02)	4.13	<i>Petunia hybrida</i>
	King George	1.99 (0.02)	3.54	
	Lagotellerie (200 m)	2.02 (0.05)	3.73	
	Lagotellerie (5 m)	2.02 (0.01)	3.69	
	Livingston	2.02 (0.02)	3.56	
	Ridge	2.01 (0.01)	3.64	

(Díaz, Serçe et al., 2020)

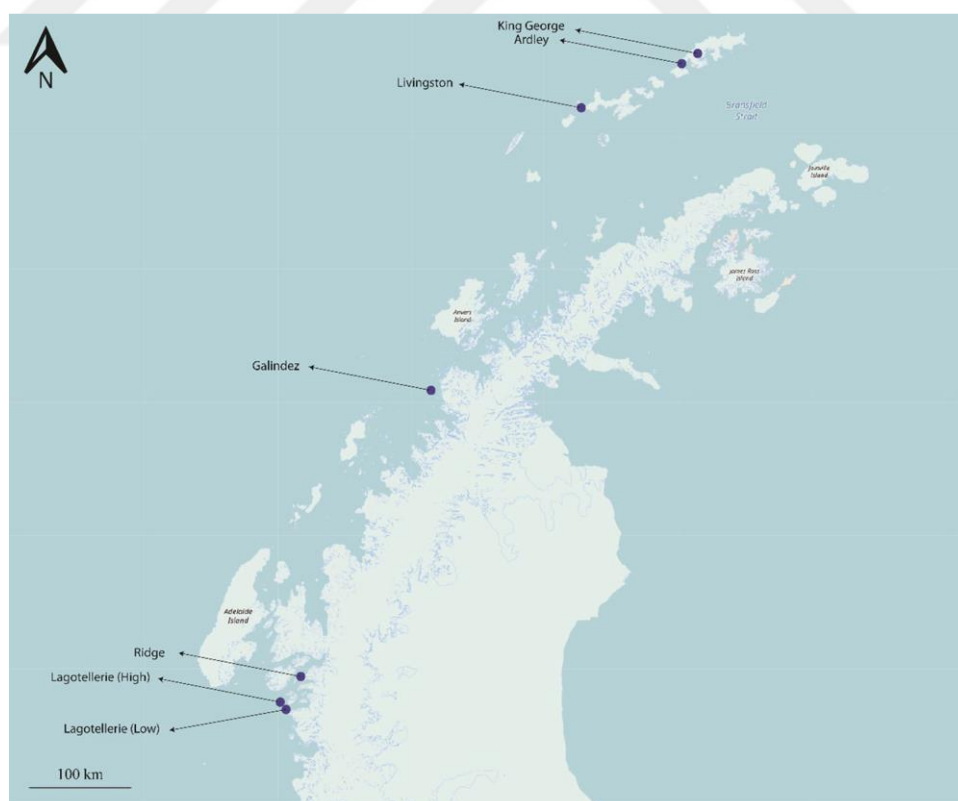


Figure 2. 1. Map of the Antarctic Peninsula indicating where the samples were collected (created with OpenStreetMap©) (Díaz, Serçe et al., 2020)

In addition, this plant can carry out photosynthesis at freezing temperature (Bravo et al., 2001). The morphology and growth density of this plant are influenced by two main factors: the distance from the coast and local topography. These factors play a key role in the formation of different habitats and microclimates within the same ice-free territory. Interestingly, variations in microclimates can even be observed within the same population of *D. antarctica*, highlighting the indicate relationship between the plant's characteristics and immediate surroundings (Bölter et al., 1989; Zwolska et al., 2002). This species has garnered significant attention from scientists across various fields of biology due to its unique adaptive abilities, which enable it to survive and reproduce in extreme climatic conditions of the Antarctic continent.

2.2 Ecological role of *D. antarctica* in Antarctic continent

Making a significant contribution to the sustainability and functioning of the local ecosystem, *D. antarctica* certainly plays a crucial role in the ecology of Antarctica.

D. antarctica, with its root system and interaction with the soil, supports microbial diversity in its environment. Thanks to the potential symbiotic relationship it can maintain with fungi or bacteria, the plant could promote nutrient cycling and improve the overall condition of the soil in the harsh conditions in which it thrives (Convey et al., 2006; Schultz et al., 2019).

Only a few flowering plants grow in Antarctica, *D. antarctica* is one of them and plays a special role in stimulating the growth and development of plants. This species plays a vital role in the complex process of creating other vegetation, as it can stabilize the soil and change the microclimate, paving the way for successful growth and expansion (Schultz et al., 2019).

Survival in Antarctic conditions largely depends on the ability to withstand harsh ultraviolet radiation and extremely low temperatures - qualities that this herb possesses to an exceptional extent.

2.3 Adaptation of *D. antarctica* to the Antarctic extreme conditions and climate change

The Antarctic continent is known for being the coldest, highest, driest, windiest and most isolated landmass on the planet. Plants exist at physiological limits because of incredibly harsh conditions of this continent. Vegetations are restricted in the ice-free zones which is covered less than 2% of continent's total area. Antarctic growth conditions are characterized by a number of unique features: low summer temperatures, a strongly seasonal climate, an extremely short growing season, continuous daylight in the middle of summer and the frequent strong winds (Robinson et al., 2003). The limited number of flowering plants in Antarctica is restricted to specific extreme conditions, resulting in the presence of only two vascular plants: *D. antarctica* and *C. quitensis* (Lee et al., 2013).

Antarctica played a pivotal role in the identification of the stratospheric ozone hole, an occurrence resulting from human-induced atmospheric pollution. Consequently, this discovery ignited worldwide apprehension regarding the potential the harmful impacts of ultraviolet (UV)-B radiation on various biological systems, including humans (Bornman et al., 2019).

In their study, Callaghan et al. (1992) revealed that polar vegetation and ecosystems play crucial role in shaping the climate system through feedback mechanisms. These mechanisms include positive loops, such as reduced reflectivity and increased carbon emissions from warming soils. But in Antarctica, the responses to environmental changes are expected to be less compared to the Arctic due to factors such as dominance of less adaptable cryptogams, the constrained availability of Antarctic soils, and geographical barriers hindering the movement of plants and invertebrates. In addition, the vegetation of this continent holds significant value for climate change research because of its geographical isolation. Antarctica remains relatively free from the impacts of human activities such as land use changes, invasive species, and pollution (Callaghan et al., 1992).

There are already some indications that climatic changes have been implicated in affecting the local population density and distribution of *D. antarctica* and *C.*

quitensis, the two vascular plant species that occur along the Antarctic Peninsula (Bokhorst et al., 2007). The population size of *D. antarctica* has shown a more rapid increase in response to global warming and harsh conditions compared to bryophytes and *C. quitensis* (Lewis, 1994; Smith, 2003; Sáez et al., 2018). Several studies have conducted the physiological and biochemical changes in *D. antarctica*, because of its successful adaptation to the harsh conditions of Antarctica and climate change. This plant exhibits unique morphological characteristics such as specific leaf anatomy and chloroplast ultrastructure, which play a pivotal role in enhancing its photosynthetic efficiency (Giełwanowska 2005; Sáez et al., 2019).

According to Ruhland's study, it was found that warming improves growth and sexual reproduction in Antarctic plants (*D. antarctica*, *C. quitensis*), with *C. quitensis* showing increased leaf production while *D. antarctica* showing reduced growth and sexual reproduction (Ruhland et al., 1999). Later in 2010 studies of the influence of temperature and salinity on the results of observation by Ruhland et al. (2010) revealed a positive correlation between temperature and plant growth. The study showed that higher temperatures improve the growth of *D. antarctica* plant and the length of roots, while salinity reduces the length of root and increase the concentration of free proline, which suggests that proline can act as an osmoregulatory in this plant (Ruhland et al., 2010).

Numerous studies, including the research conducted by Byun et al. (2021), have confirmed that *D. antarctica* demonstrates a uniform mechanism to tolerate various abiotic stressors. Byun et al. (2021) conducted a study to investigate the molecular mechanisms that enable Antarctic plant *D. antarctica* to thrive in harsh environments. In study, they focused on a gene called actin depolymerizing factor (ADF) and identified eight ADF genes in the *D. antarctica* transcriptome. Among these genes, significant induction of *DaADF3* transcription has been demonstrated in response to dehydration, cold, and Antarctic harsh conditions. Using promoter analysis, they found many elements in *DaADF3* that are sensitive to drought and low temperatures, indicating its involvement in reactions to abiotic stress. To confirm its function, *DaADF3* was overexpressed in rice plants, resulting in increased resistance to low-temperature stress compared to wild-type plants. And this increased resistance was accompanied by changes in the organization of actin filaments at the tips of the

roots. These results highlight the crucial role of *DaADF2* in increasing the cold tolerance of transgenic rice plants and in facilitating the adaptation of *D. antarctica* to the Antarctic harsh conditions (Byun et al., 2021).

The study conducted by Lee et al. (2008), resulted with generation of 1,969 high-quality *D. antarctica* EST sequences. Among these sequences, 1,199 unique genes were identified. This discovery is important because it may shed light on the potential functions of these genes based on existing knowledge, providing valuable information for further studies.

Results from this study serves as a valuable resource for designing microarrays, allowing researchers to choose between cDNA microarrays or oligo-based platforms for their analyses. This flexibility enhances molecular studies, especially in the field of marker screening and gene expression profiling under different environmental conditions. Through cluster analysis, the genes with similar characteristics also were grouped together by Lee et al. (2008), facilitating the understanding of gene relationships and expression patterns. Redundancy information in the dataset helps refine the data by eliminating repetitive sequences, optimizing them for further studies.

In conclusion, cluster analysis and redundancy information obtained from the study by Lee et al. (2008) serve as a crucial resource in enhancing cDNA libraries through the elimination of duplicate transcripts and concentration on distinct sequences. This process optimizes the dataset, making it more adaptable and efficient for future functional genomics investigations. Through the elimination of multiple transcripts, researchers can better isolate and characterize genes of interest, enhancing the precision and effectiveness of new investigations. Consequently, these analytical instruments not only aid in a more profound comprehension of gene expression dynamics but also lay the way for more targeted and effective research on functional genomics in the future (Lee et al., 2008).

In the study conducted by Amosova et al. (2015) showed that the variability in the karyotypes of *D. antarctica* specimens collected from four islands: Galindez, Skua, Great Jalour and Darboux in the Maritime Antarctica zone is probably the result of

genome instability caused by environmental stressors. The presence of telomere repeats at intercalary sites indirectly supports the hypothesis that chromosome fusion events may have played a role in the unusual chromosome number observed in this species, particularly in comparison to other cereals. Furthermore, differences in C-banding patterns, rDNA loci, and highly repeated DNA sequences between *D. antarctica* and its close relative, *D. caespitosa* or known as tufted hairgrass (which is found in temperate regions) suggest significant reorganization during the evolutionary divergence of these species. This molecular cytogenetic analysis is not only important for future comparative studies of *Deschampsia* species (refer to Table 2.1, Cardone et al., 2009), but also provides a basic information for genetic and biotechnological applications, potentially helping in the breeding of cold-tolerant crops. Overall, this research enhances in understanding of evolutionary dynamics and adaptation mechanisms in Antarctic environment and offers practical solutions to improve agriculture (Amosova et al., 2015).

Table 2. 2. Chromosome numbers in *Deschampsia* species

Species	2n	References	Species	2n	References
<i>D. alpina</i> (ssp) ^{a, b}	52	Löve and Löve (1975)	<i>D. koelerioides</i>	26	Tzvelev (1976)
	50	Engelskjon (1979)	<i>D. komarovi</i>	52	Petrovsky and Zhukova (1981)
	39, 49, 52	Alberts (1980)		26	Zhukova (1980)
<i>D. antarctica</i> ^b	26	Present work	<i>D. liebmanniana</i>	26, 52	Taken from Fedorov (1969)
<i>D. argentea—maderensis</i>	26	Dalgaard (1991)	<i>D. mackenzieana</i> ^a	52	Purdy and Bayer (1995)
<i>D. atropurpurea</i>	14	Alberts (1972)	<i>D. macrothyrsa</i> ^a	26	Probatova (1984)
<i>D. beringensis</i> ^a	26, 42	Sokolovskaya and Probatova (1975)	<i>D. media</i>	26	Alberts (1980)
<i>D. borealis</i> ^a	26, 28	Tzvelev (1976)		28	Kerguelen (1975)
<i>D. bottnica</i> ^{a, b}	26	Alberts (1980)	<i>D. mildbraedii</i>	52	Morton (1993)
<i>D. brevifolia</i> ^a	52	Krogulevich (1976)	<i>D. obensis</i>	52	Zhukova and Petrovsky (1980)
	52	Petrovsky and Zhukova (1981)	<i>D. orientalis</i> ^a	52	Petrovsky and Zhukova (1981)
<i>D. caespitosa</i> ^b	26 (0-2B)	Pashuk (1980)		26	Löve and Löve (1981)
	26, 52	Stoeva (1982)	<i>D. parviflora</i> ^a	26, 28	Kerguelén (1993)
	26	Beuzenberg and Hair (1983)	<i>D. pamirica</i> ^a	26	
	26	Strid and Franzén (1983)	<i>D. pumila</i>	26	Dalgaard (1989)
	26, 52	Rhotera and Davy 1986	<i>D. refracta</i>	26	Alberts (1980)
<i>D. chapmanii</i> ^b	26	Edgar (1993)	<i>D. rhenana</i> ^a	49–52	Alberts (1980)
<i>D. danthonioides</i> ^b	26	Taken from Fedorov (1969)	<i>D. setaceae</i>	14	Hubbard (1984)
<i>D. elongata</i> ^b	26	Taken from Fedorov (1969)		14	Lövkvist and Hultgard (1999)
<i>D. festucaeifolia</i> ^a	27, 28	Osada (1993)	<i>D. sukatschewii</i>	52	Zhukova (1980)
<i>D. flexuosa</i>	28	Stoeva; Arohonka (1982)	<i>D. tenella</i> ^b	26	Edgar and Connor (2000)
	28	Strid and Franzen 1983	<i>D. wibeliana</i>	26	Alberts (1980)
	26	Druskovic and Lovka (1995)		26 (05B)	Mesíček (1992)
	28	Petrova and Stoyanova (1998)			
<i>D. glauca</i> ^a	26, 52	Zhukova and Petrovsky (1975), (1976)			

(Cardone et al., 2009)

Understanding the effects of recent climate change on the Antarctic Peninsula is crucial for assessing the sustainability and long-term viability of *D. antarctica* plant population. Changes in temperature, precipitation patterns, and other climatic factors have a direct impact on the development, density, and distribution of plant species in the area, potentially resulting in modified habitats, shifts in species composition, and survival challenges for plant (Turner et al., 2005; Gieldanowska et al., 2011). There are several studies have investigated the effects of temperature on various aspects of *D. antarctica*, including plant physiology, height and chlorophyll levels. However, these studies have focused on specimens obtained from only one site of Antarctic zones, rather than comparing samples from different Antarctic environments, such as Antarctic seaside and tundra. Comparative studies between different Antarctic habitats could provide valuable insights into how the species reacts to temperature fluctuations and its broader ecological and evolutionary patterns within this area having desirable features.

CHAPTER III

MATERIALS AND METHODS

This study was conducted in the laboratories of the Department of Agricultural Genetic Engineering at Niğde Ömer Halisdemir University between May and December of the year 2023. The purpose of study was to compare the effect of two different temperatures on *D. antarctica* plants' development, physiology and morphology.

3.1 Plant Material

The *D. antarctica* samples collected during the 3rd National Antarctica Scientific Expedition by Prof. Dr. Sedat Serçe were bought back to Türkiye and are currently kept at Niğde Ömer Halisdemir University, Ayhan Şahenk Faculty of Agricultural Sciences and Technologies, Agricultural Genetic Engineering Department. These samples were utilized for this study (Figure 3.1).



Figure 3. 1. Plant material of *D. antarctica* used in thesis research

3.2 Method

Two separate trials were conducted in two different growing condition, 12°C and

22°C, with the aim of determining the response of *D. antarctica* ecotypes collected from Ardley, Galindez, King George, Lagotellerie (200 m), Lagotellerie (5 m), Livingston and Ridge islands to two different temperature conditions 12°C and 22°C. In preparation for planting, a suitable substrate was crafted. Turf and perlite were combined in a ratio of 3:1, creating an optimal mixture. This optimal mixture was then filled into pots designed for transplanting and sowing plants. To ensure accuracy, each pot was labeled with the respective names of the *D. antarctica* ecotypes (Figure 3.1).

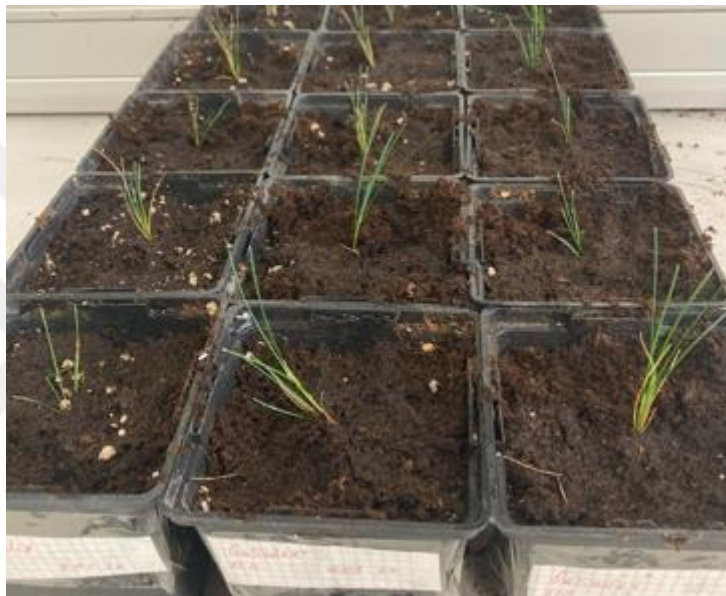


Figure 3. 2. Transplanted *D. antarctica* plants into individual labeled pots

3.2.1 Comparison of *D. antarctica* ecotypes' response to two different temperature conditions (12°C and 22°C)

In the study *D. antarctica* siblings were taken carefully from each ecotype to transplant them into new labelled pots. The prepared plants were transplanted into new pots, and single sibling was placed in each pot. The experiment was set up in a completely randomized design with 5 replications arranged in randomized complete blocks, with five pots. The plants were then grown under a 16/8-hour (day/night) photoperiod in 12°C and 22°C for 6 months (Figure 3.2).



Figure 3.3. *D. antarctica* ecotypes grown in two different temperature conditions (a: 12°C; b: 22°C)

In the growth period the following measurements and analyses have been conducted on plants.

Plant Height: At three-weeks intervals, the height of plant culm from the soil to the tip were measured in centimeters using a ruler for each plant in each pot.

Plant Sibling Number: At three-weeks intervals, the enumeration and recording of each newly established sibling were meticulously performed.

After six months the plants were removed from the pots, and root length, number of siblings, and fresh and dry weights of roots were measured.

Plant Root Length: The roots of plants in each pot were cleaned with distilled water, then their lengths were measured in centimeters using a ruler to determine their overall lengths (Figure 3.3).

Plant Leaf-Root Fresh Weight: The weights of plant leaves and roots in each pot were measured separately after cutting and cleaning with distilled water, and after removing excess water from the plants using a sensitive balance.

Plant Leaf-Root Dry Weight: The weights of plant leaves and roots in each pot were measured separately after drying at a constant temperature of 65°C for duration of 48 hours using a sensitive balance.

Plant Chlorophyll Content: The chlorophyll content of the plants was measured using a UV-Vis spectrophotometer (UV-1800, Shimadzu) at 646 and 663 nm. The chlorophyll extraction procedure employed from the original methodology introduced by McKinney (1941), with modification based on the refinements proposed by Starnes and Hadley (1965).

Leaf chl *a* and *b* content were determined by 0.5 g of fresh *D. antarctica* ecotype leaves which were grinded using mortar and pestle with adding 80% of 10 mL acetone solution to obtain homogenous sample (Figure 3.4).



Figure 3. 4. Homogenization of the *D. antarctica* leaves for chlorophyll content analysis

The homogenized solution was kept at 4°C overnight for chlorophyll extraction from plant tissues. After overnight the solution was brought up with acetone to the volume 25 mL. The mixture was centrifuged at 6000 rpm for 15 min at 4°C. The resulting solution was then transferred to a new test tube (Figure 3.5).

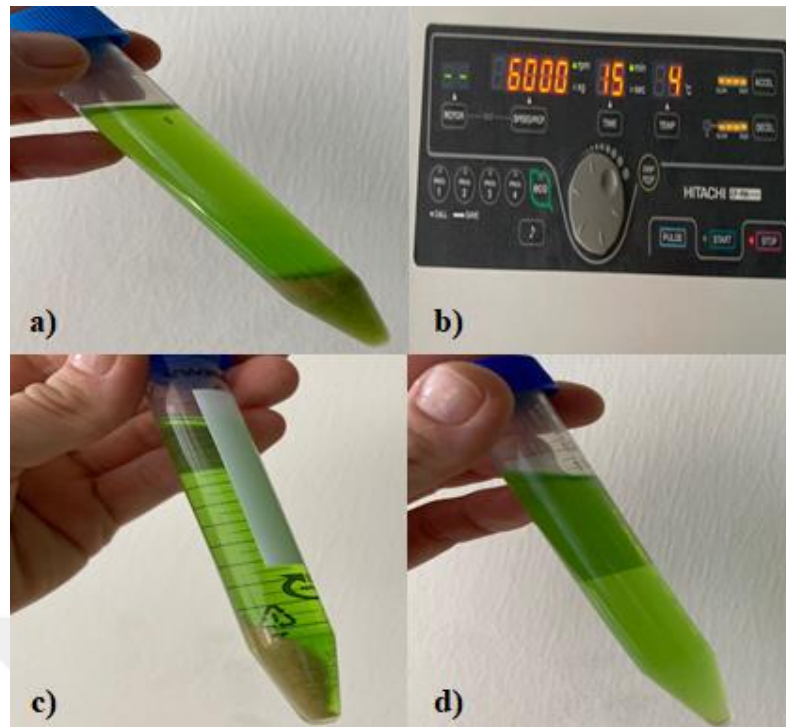


Figure 3. 5. Preparation of the solution for chlorophyll content measurement (a, b, c, d)

To determine the absorption of the supernatant, a UV-Vis spectrophotometer (UV-1800, Shimadzu) was utilized at 646 and 663 nm. To ensure accurate results, the cuvettes underwent a thorough cleansing process using 70% ethanol and were then meticulously dried. Following this, a volume of 2 mL of 80% acetone was introduced into the cuvettes, which were subsequently positioned in the spectrophotometer for the purpose of calibration, resulting in zero readings. Once the machine was blanked, 2 mL of the solution was added to the cuvettes and placed in the machine for the measurement of chlorophyll content (Figure 3.6).



Figure 3. 6. Chlorophyll content measurement using a UV-Vis spectrophotometer

For calculation of chlorophyll content were determined by formula defined by Arnon (1949) (Arnon, 1949).

$$\text{Chl } a \text{ (}\mu\text{g/mL)} = (12.7 * A_{663} - 2.69 * A_{646}) * V/W$$

$$\text{Chl } b \text{ (}\mu\text{g/mL)} = (22.9 * A_{646} - 4.68 * A_{663}) * V/W$$

$$\text{Total chlorophyll (}\mu\text{g/mL)} = (20.2 * A_{646} + 8.02 * A_{663}) * V/W$$

Here:

A_{663} – Absorbance at 663 nm

A_{646} – Absorbance at 646 nm

V- Final volume of extract in ml

W- Weight of sample

3.2.2 Statistical Analysis

All statistical analyses were conducted using the procedures of SAS (SAS, 1990). All ANOVAs were constructed using the GLM procedure employing the completely randomized design used (SAS, 1990).

CHAPTER IV

RESULTS AND DISCUSSION

4.1 Experiment Results Conducted Under Two Different Temperature Conditions

In the study, the response of seven different ecotypes of *D. antarctica* to the two different (12°C and 22°C) temperature was investigated in terms of physiological and morphological characteristics under *in vivo* conditions. Under the mentioned conditions, the behavior of different *D. antarctica* ecotypes upon two different temperature conditions was determined by measuring plant height, sibling number, root length, plant leaf-root fresh weight, plant leaf-root dry weight, chlorophyll content of plants developed in growth chambers. Plant developed from single sibling completed their entire 6-months growth under two different conditions are given in Figure 4.1.

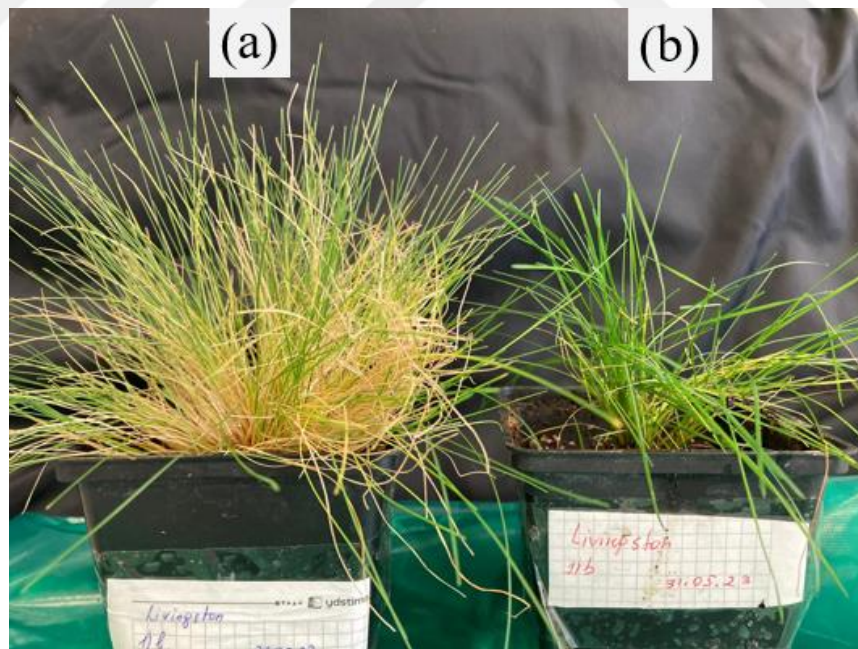


Figure 4. 1. An image of plants belonging to the Livingston ecotype grown under two different temperature conditions (a: 22°C, b: 12°C)

4.1.1 Plant Height

The plant heights were monitored by measuring them every three weeks after sowing for a period of six months. The analysis of variance (ANOVA) results are presented in Table 4.1 and 4.2, showing the effect of two different temperature conditions, 12°C and 22°C, on plant height across all seven *D. antarctica* ecotypes. According to the results, a significant interaction was observed between ecotypes and temperature.

Table 4. 1. ANOVA table showing measured plant height at three-week interval during 6-month development of *D. antarctica* ecotypes grown in 12°C

Source	DF	1 st	2 nd	3 rd	4 th	5 th **	6 th **	7 th **
Ecotype	6	6.645	7.083	8.820	14.607	13.505	14.398	11.253
Error	14	4.380	7.602	5.809	5.640	1.340	1.595	0.573
Total	20							
CV		36.500	30.677	29.237	27.790	14.323	14.551	8.618

*p≤0.05, **p≤0.01

Table 4. 2. ANOVA table showing measured plant height at three-week interval during 6-month development of *D. antarctica* ecotypes grown in 22°C

Source	DF	1 st **	2 nd **	3 rd **	4 th **	5 th **	6 th **	7 th **
Ecotype	6	12.96	35.29	43.08	42.39	37.67	27.23	27.51
Error	14	0.338	0.726	0.537	0.538	0.730	1.144	0.796
Total	20							
CV		7.508	6.918	6.367	6.401	8.538	10.653	9.676

*p≤0.05, **p≤0.01

The results presented in Tables 4.3 and 4.4 highlight distinct effects on the average plant heights of *D. antarctica* ecotypes under two different temperature conditions during 6-month period. These results allow for a comparative analysis how the growth responses of ecotypes vary distinct temperature conditions.

Differential Growth Responses to Temperature: Across the measurement intervals, ecotypes showed different responses in plant height under 12°C and 22°C. Ecotypes such as Galindez, Livingston and Ridge showed higher average height at 22°C in

compared to 12°C in 2nd measured plant height interval. Conversely, ecotypes like Ardley and Lagotellerie (5 m) exhibited minimal differences in average height between the two temperature conditions, suggesting a lesser impact of temperature variations on their development. Each ecotype demonstrated specific reactions to temperature fluctuations. For instance, Galindez displayed a significant increase in average height from 10.7 cm at 12°C and maintained consistent growth trends across intervals at 22°C, resulting in higher average heights compared to 12°C conditions. Ecotype King George showed relatively stable heights with minor fluctuations at 12°C, while exhibited consistent growth patterns with slightly elevated heights at 22°C. Livingston showed consistently high heights across all intervals, displaying the highest heights overall. Ridge showed moderate growth with minor fluctuations at 12°C, and at 22°C displayed similar growth patterns as under 12°C but with marginally higher average height.

Temperature Sensitivity of Specific Ecotypes: Ecotypes such as Ardley and Lagotellerie (200 m) exhibited minimal differences in average height between 12°C and 22°C, suggesting a lesser impact of temperature variations on their development. However, observing the 3rd measured plant height interval in the Table 4.4, revealed a decrease in height for ecotypes such as Ardley and Lagotellerie (200 m) when comparing the 2nd and 3rd measurement intervals. Specifically, the height of Ardley decreased from 10.7 cm to 9.3 when exposed to a temperature 22°C. Similarly, the height of Lagotellerie (200 m) decreased from 9.6 cm to 8.1 cm under the same temperature condition as detailed in Figure 4.5. These findings show the sensitivity of certain ecotypes to temperature changes. The Lagotellerie (200 m) ecotype predominantly exhibited a decrease in plant height after 6-month of growth at 22°C compared to other ecotypes (refer to Figure 4.3 and 4.4).

Table 4. 3. Average of measured plant height at three-week interval during 6-month development of *D. antarctica* ecotypes developed under 12°C

Ecotype	1st	2nd	3rd	4th	5th	6th	7th	Mean
Ardley	7.6 ^a	8.8 ^d	7.3 ^d	7.4 ^d	5.4 ^f	7.0 ^f	8.3 ^c	7.4
Galindez	5.8 ^b	10.7 ^b	9.3 ^b	9.3 ^{b-c}	7.5 ^d	8.4 ^d	7.3 ^e	8.3
King George	5.2 ^c	7.5 ^e	5.8 ^f	5.8 ^e	6.0 ^e	6.3 ^g	6.7 ^f	6.2
Lagotellerie (200m)	5.2 ^c	9.4 ^c	9.1 ^{b-c}	9.9 ^b	8.4 ^c	7.9 ^e	7.9 ^d	8.3
Lagotellerie (5m)	5.7 ^{b-c}	8.8 ^d	8.3 ^c	7.9 ^c	9.3 ^b	9.2 ^b	9.5 ^b	8.4
Livingston	7.4 ^b	11.0 ^a	10.9 ^a	12.5 ^a	11.8 ^a	13.1 ^a	12.6 ^a	11.3
Ridge	3.2 ^d	6.8 ^f	7.0 ^e	7.1 ^{d-e}	8.2 ^{c-d}	8.8 ^c	9.2 ^{b-c}	7.2
Mean	5.7	9.0	8.2	8.5	8.1	8.7	8.8	8.2

Different letters indicate significant differences in by Tukey test at 0.05

Table 4. 4. Average of measured plant height at three-week interval during 6-month development of *D. antarctica* ecotypes developed under 22°C

Ecotype	1st	2nd	3rd	4th	5th	6th	7th	Mean
Ardley	5.8 ^e	10.7 ^d	9.3 ^f	9.3 ^e	7.5 ^e	8.4 ^d	7.3 ^e	8.3
Galindez	9.2 ^b	15.5 ^b	14.5 ^b	14.4 ^b	12.9 ^b	11.6 ^c	9.6 ^c	12.5
King George	7.0 ^{c-d}	10.5 ^{d-e}	10.5 ^e	10.4 ^d	8.6 ^d	8.4 ^d	7.7 ^d	9
Lagotellerie (200m)	6.3 ^d	9.6 ^e	8.1 ^g	8.1 ^f	6.0 ^g	6.0 ^f	6.1 ^f	7.2
Lagotellerie (5m)	7.0 ^{c-d}	8.4 ^f	7.3 ^h	7.2 ^h	7.2 ^f	8.4 ^d	7.2 ^{e-f}	7.5
Livingston	11.8 ^a	17.8 ^a	17.7 ^a	17.6 ^a	15.3 ^a	14.4 ^a	14.2 ^a	15.5
Ridge	7.2 ^c	13.6 ^c	13.2 ^c	13.2 ^c	12.6 ^c	13.0 ^b	12.4 ^b	12.2
Mean	7.7	12.3	11.5	11.5	10.0	10.0	9.2	10.3

Different letters indicate significant differences in by Tukey test at 0.05

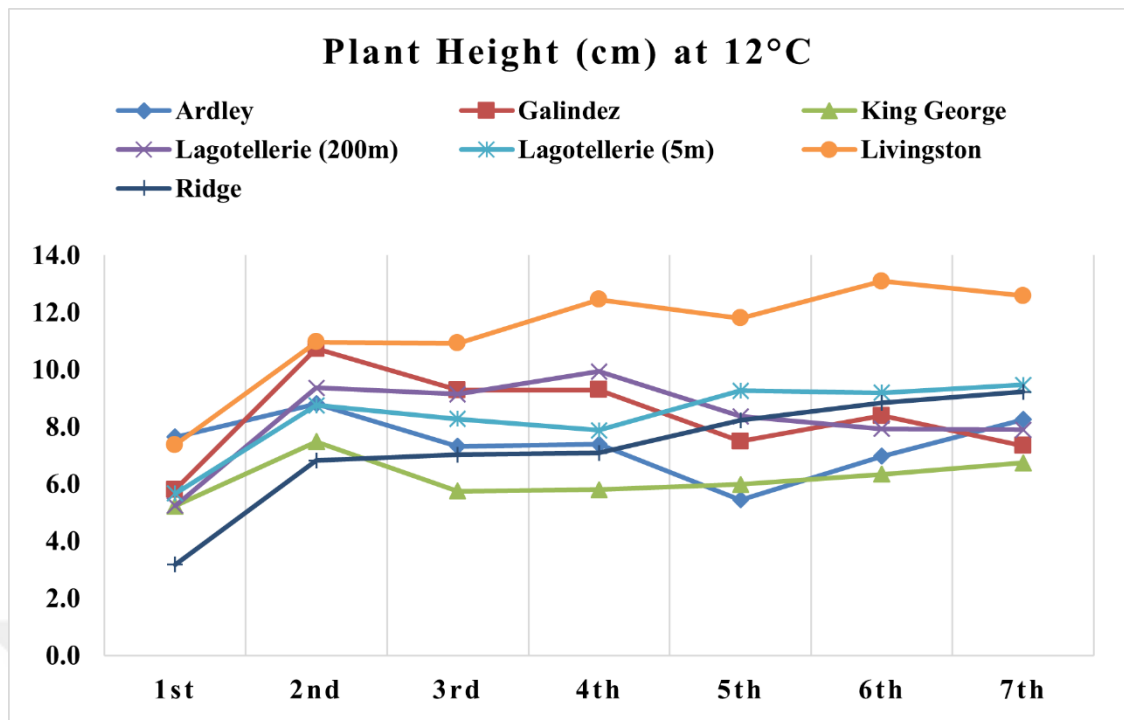


Figure 4. 2. Comparison of the plant height at three-week interval during 6-month development of *D. antarctica* ecotypes grown in 12°C

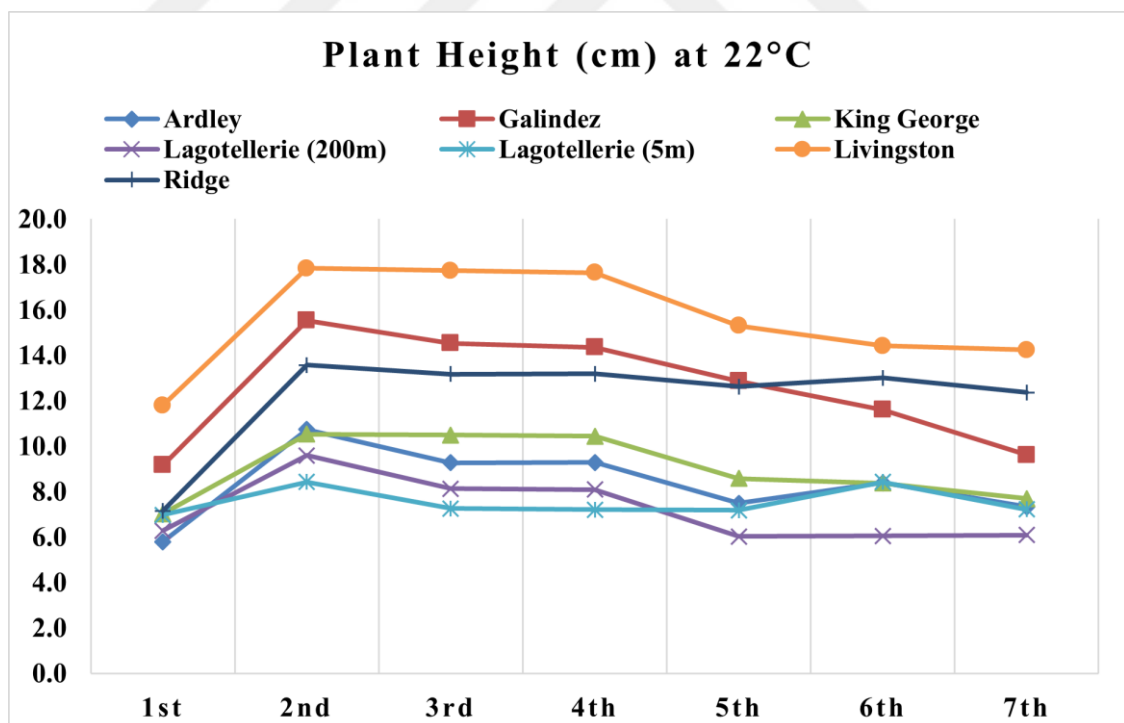


Figure 4. 3. Comparison of the plant height at three-week interval during 6-month development of *D. antarctica* ecotypes grown in 22°C

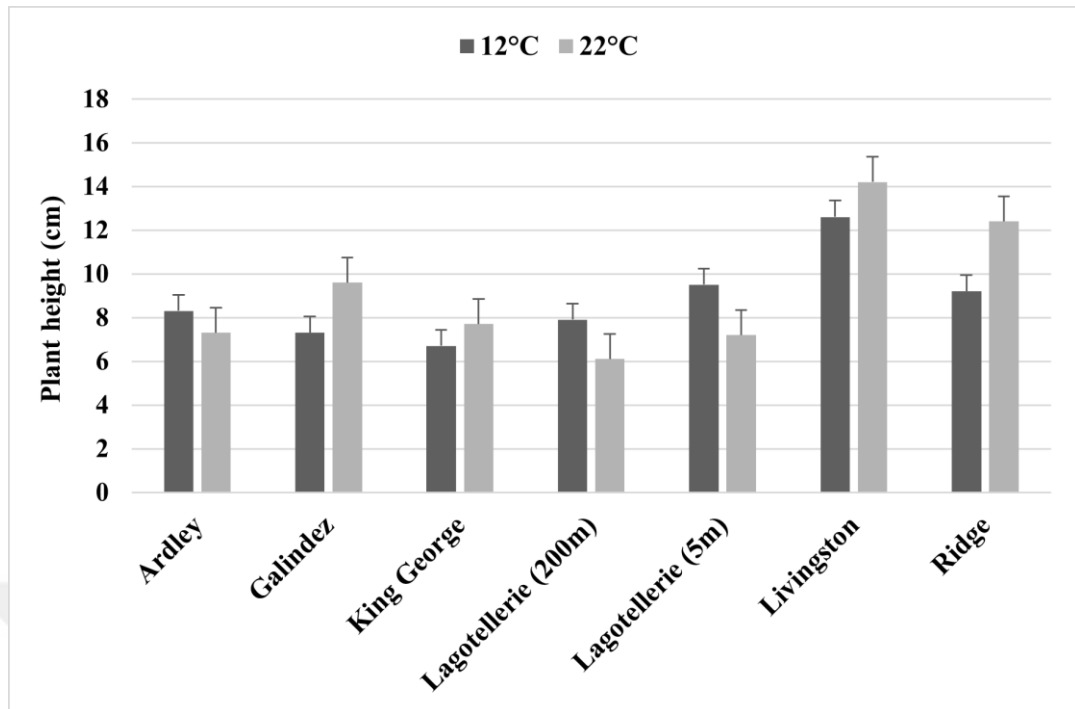


Figure 4. 4. Comparison of the final measured plant height at three-week interval during 6-month development of *D. antarctica* ecotypes grown under two different temperature conditions



Figure 4. 5. An image depicting tip drying and subsequent breakage of plant leaves in Lagotellerie (200 m) ecotype, grown in 22°C

According to the results showed in Figure 4.4, it revealed sensitivity in the Lagotellerie (200 m) ecotype compared to the Lagotellerie (5 m) as illustrated in Figures 4.6, 4.7, 4.8 and 4.9, and as well as compared to other ecotypes under similar temperature conditions.

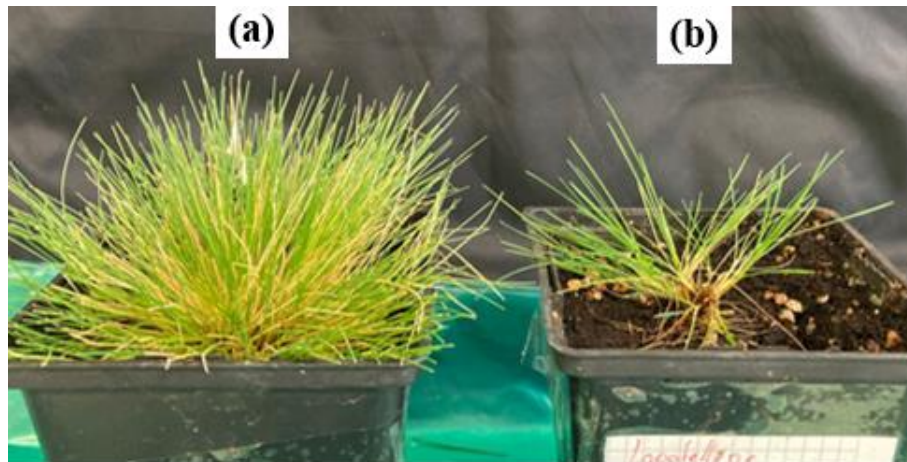


Figure 4. 6. An image belonging to the Lagotellerie (5 m) grown under two different temperature conditions (a: 22°C; b: 12°C)

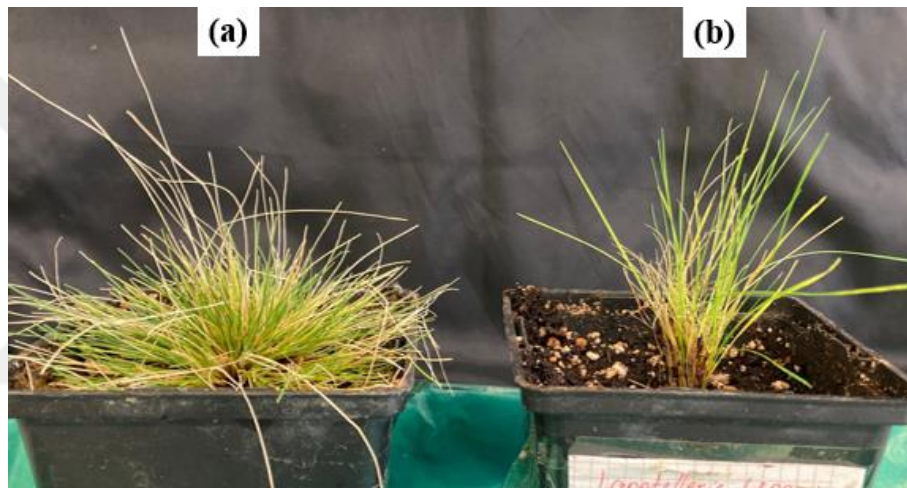


Figure 4. 7. An image belonging to the Lagotellerie (200 m) ecotype grown under two different temperature conditions (a: 22°C; b: 12°C)

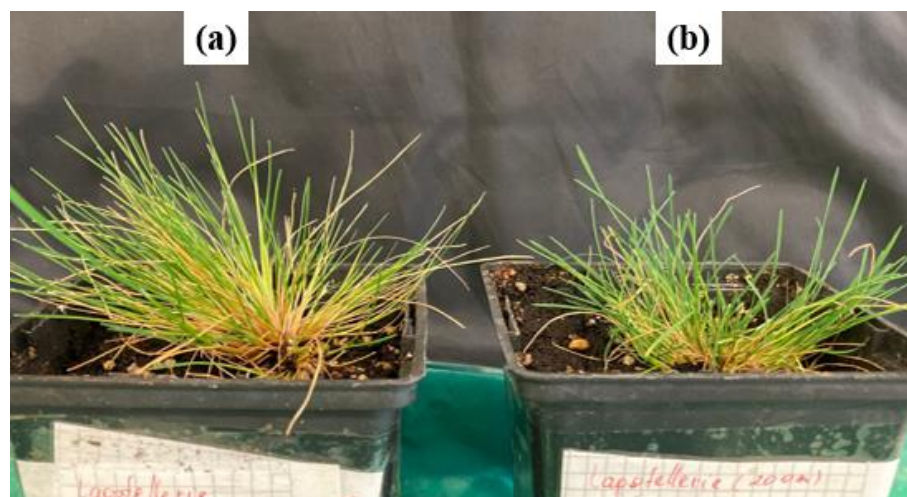


Figure 4. 8. An image belonging to the ecotypes grown under 12°C (a: Lagotellerie (5 m); b: Lagotellerie (200 m))

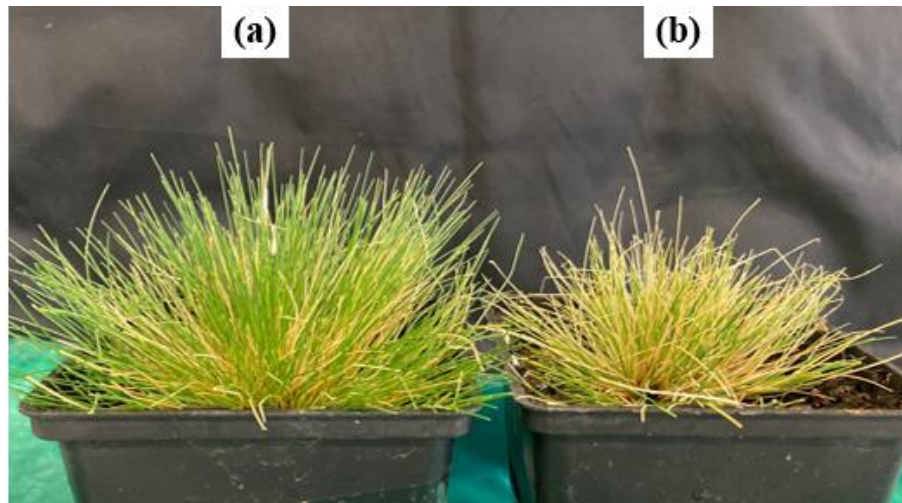


Figure 4. 9. An image belonging to the ecotypes grown under 22°C (a: Lagotellerie (5 m); b: Lagotellerie (200 m))

The study by Gielwanowska et al. (2005) examined *D. antarctica* plants across different environments, including the Antarctic tundra, a greenhouse, and the seashore. Their findings revealed that plants from the tundra and greenhouse exhibited stronger characteristics to withstand dry conditions compared to those from the seashore. The research delved into the alterations in the internal structure of under stress. The findings indicate a strong correlation between the leaf structure and the plant's habitat, suggesting that the plant adapts its leaf anatomy according to its specific growth location (Gielwanowska et al., 2005).

The results obtained from our study provide strong evidence to support the assumptions made. Furthermore, our research specifically highlights changes in leaf traits such as size, shape, and color, which further validate these assumptions. Notably, leaves from ecotypes grown in a 22°C environment showed a distinct trend towards greener coloration and a thinner structure compared to those cultivated in 12°C conditions, as showed in Figure 4.10.

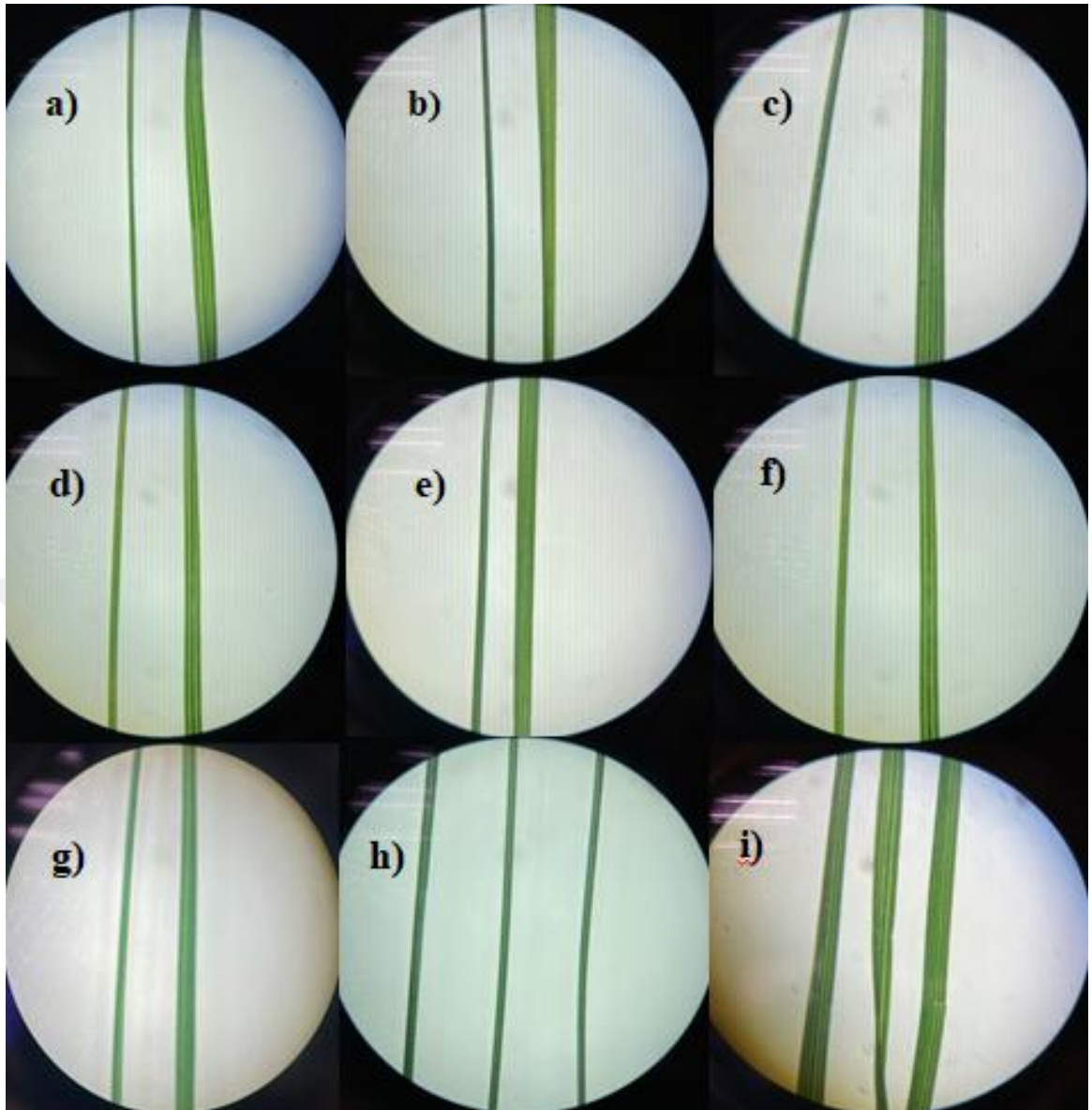


Figure 4. 10. Comparison of changes between leaves of plants after a period of 6 months under two different temperature conditions: 22°C on the left and 12°C on the right (a: Ridge; b: Galindez; c: Lagotellerie (5 m); d: Lagotellerie (200 m); e: Livingston; f: King George; g: Ardley; h: plant leaves grown in 22°C; i: plant leaves grown in 12°C)

The *D. antarctica* ecotype collection experienced significant effects on its growth, physiology, and structure due to a six-month interplay between two distinct temperature ranges. In a study conducted by Rodrigo Cortés-Antíquera et al. (2021) it was discovered that *D. antarctica* demonstrated remarkable resilience to heat shock treatments without any observable damage to its physical characteristics or oxidative state *in-vitro* conditions. This ability appeared to be supported by both enzymatic and non-enzymatic antioxidant systems, which acted as a defense

mechanism against heat-induced stress (Cortez-Antiquera et al., 2021). Interestingly, our study found that plants exposed to a temperature of 22°C exhibited noticeable differences in size, sibling numbers, and leaf color and shape. These findings align with the research conducted by Rodrigo Cortés-Antíquera et al. (2021).

In the study conducted by Xiong et al. (2000), the focus was on investigating the growth response and thermal acclimation of Antarctic vascular plants, specifically *C. quitensis* and *D. antarctica*, under different temperature conditions. The experiment spanned a period of 90 days, during which the plants were grown at three temperature settings: 7°C Day /7°C night, 12°C Day/7°C night, and 20°C Day/7°C night. These temperatures represented suboptimal, near-optimal, and supraoptimal conditions for photosynthesis (Xiong et al., 2000).

The key findings of the study revealed that plants grown at 20°C exhibited higher relative growth rates and produced significantly more biomass, leaves, and leaf area compared to those grown at lower temperatures (Xiong et al., 2000). Our study showed similar results between ecotypes grown in both 12°C and 22°C conditions, mirroring the patterns observed in the study carried out at 20°C. The findings of our study were remarkably similar to those observed in the 20°C study. We observed a consistent trend of increased growth and enhanced leaf production across different temperature conditions, as depicted in Figure 4.11.



Figure 4. 11. An image belonging to the *D. antarctica* plants grown in 22°C temperature condition

The alterations in leaf morphology, increased plant length in some ecotypes, and changes in sibling numbers are likely adaptive responses rather than indications of damage. Such changes are common strategies employed by organisms to adapt to varying environmental conditions. For instance, modifications in leaf shape can optimize light absorption or minimize water loss, thereby enhancing survival in fluctuating temperatures. Similarly, adjustments in plant length and sibling numbers may signify strategies aimed at optimizing growth and reproduction in different temperature settings. These adaptive responses highlight the organism's ability to thrive and reproduce in diverse environments, rather than suggesting any negative effects.

In addition, ecotype such as Ridge demonstrated a remarkable characteristic – it bloomed the flower at 22°C as showed in the Figure 4.12.

This intriguing pattern presents several potentials of explanations:

- **Temperature Resilience:** The Ridge ecotype may exhibit a narrower range of response to temperature fluctuations, maintaining a consistent performance

despite changes. However, it selectively initiates flowering as a stress response at 22°C (Figure 4.12).

- **Threshold Sensitivity:** It is possible that this ecotype has a specific threshold for flowering, particularly at higher temperatures like 22°C, perceiving this temperature as a signal to initiate reproduction when faced stressful conditions.
- **Strategic Resource Allocation:** Despite displaying less variation in performance across temperatures, the Ridge ecotype could strategically allocate resources, giving priority to reproduction (flowering) as survival tactic specifically in response to 22°C – induced stress.
- **Unique Adaptive Traits:** This ecotype may possess distinct genetic or physiological adaptations, enabling it to flower specifically in response to stress triggered by 22°C, demonstrating its specialized survival strategy under specific temperature stresses.

While the Ridge ecotype has maintained relatively stable performance across temperature changes, its stress-induced flowering at 22°C, indicating a specific sensitivity to this thermal threshold and thereby highlighting its unique adaptive mechanism in dealing with environmental stressors. This phenomenon is intricately tied to study by conducted Lee et al. (2008) that revealed distinct gene regulation patterns in response to environmental factors, highlighting the significance of genes in adapting to particular conditions (Lee et al., 2008). Our study supplements these finding by examining physiological and developmental changes, particularly in leaf morphology and growth patterns, which reflect underlying genetic adaptations to the existing environmental conditions. This ability to adapt allow plants to optimize their growth features based on the specific challenges presented by their habitats, ultimately improving their capacity to survive and flourish in various environmental settings.



Figure 4. 12. Stress-inducing flower of Ridge ecotype grown in 22°C

4.1.2 Plant Sibling

From the ANOVA tables, the effect of temperature conditions on the sibling number percentages of *D. antarctica* ecotypes that were grown under two different temperature condition, 12°C and 22°C were observed. In the Tables 4.5 and 4.6, significant differences were determined in terms of temperature and ecotype interaction under both temperature conditions.

Table 4. 5. ANOVA table showing plant sibling number at three-week interval during 6-month development of *D. antarctica* ecotypes grown in 12°C

Source	DF	1 st	2 nd	3 rd *	4 th *	5 th **	6 th **	7 th **
Ecotype	6	0.093	1.226	3.154	6.850	12.932	19.345	23.884
Error	14	0.047	0.638	0.944	1.632	2.560	2.645	3.415
Total	20							
CV		19.254	45.337	41.319	39.807	39.529	33.553	34.222

*p≤0.05, **p≤0.01

Table 4. 6. ANOVA table showing plant sibling number at three-week interval during 6-month development of *D. antarctica* ecotypes grown in 22°C

Source	DF	1 st	2 nd	3 rd	4 th	5 th	6 th	7 th
Ecotype	6	0.347	0.810	2.433	4.108	3.811	6.040	9.852
Error	14	0.238	0.492	0.0894	1.847	2.734	4.147	5.277
Total	20							
CV		15.238	13.167	12.393	13.057	12.809	13.537	13.592

*p≤0.05, **p≤0.01

Tables 4.7 and 4.8 displayed the sibling number percentage of *D. antarctica* ecotypes at three-week intervals under 12°C and 22°C temperature conditions, along with their mean values. Upon examining these results, significant insights emerge regarding the impact of temperature on sibling numbers.

When comparing the results of two tables, it was determined that the average of sibling numbers for all ecotypes grown under 22°C temperature conditions significantly increased compared to those under 12°C. This shift reflects the influence of a higher temperature environment on promoting greater sibling proliferation between *D. antarctica* ecotypes. In terms of specific ecotypes, under the 22°C conditions, Lagotellerie (200 m) and King George, exhibited the most robust development in terms of sibling numbers, while the weakest development was observed in the ecotype Ridge. This contrasts with observations from the 12°C conditions, where Ridge displayed the highest sibling numbers, and the weakest development was observed in the ecotypes King George and Ardley.

These results showed that the reproductive abilities and success rates of *D. antarctica* siblings influenced differently by the changes in temperatures. The distinct temperature conditions not only influenced the absolute number of siblings but also led to varying reproductive outcomes among ecotypes. The comparison between two tables highlights clear evidence: ecotypes grown at 22°C displayed a greater number of siblings in comparison to those grown at 12°C. This significant differences in reproductive yields highlights the crucial role of temperature in shaping the reproductive dynamics and success rates of *D. antarctica* ecotypes.

Table 4. 7. Average of plant sibling number at three-week interval during 6-month development of *D. antarctica* ecotypes developed under 12°C

Ecotype	1st	2nd	3rd	4th	5th	6th	7th	Mean
Ardley	1.0 ^d	1.0 ^e	1.0 ^f	1.1 ^e	1.2 ^e	1.3 ^e	1.5 ^f	1.1
Galindez	1.0 ^d	1.9 ^c	3.1 ^c	4.1 ^{b-c}	4.8 ^{c-d}	6.1 ^c	6.8 ^c	4.0
King George	1.0 ^d	1.0 ^e	1.0 ^f	1.0 ^f	1.0 ^f	1.2 ^f	1.3 ^g	1.1
Lagotellerie(200m)	1.3 ^b	1.5 ^d	2.0 ^e	3.7 ^d	4.7 ^d	5.7 ^d	6.1 ^e	3.6
Lagotellerie (5m)	1.5 ^a	2.7 ^a	3.2 ^b	4.2 ^b	5.6 ^b	6.7 ^b	7.4 ^b	4.5
Livingston	1.1 ^c	1.9 ^c	2.8 ^d	3.9 ^c	4.9 ^c	5.7 ^d	6.5 ^d	3.8
Ridge	1.1 ^c	2.3 ^b	3.4 ^a	4.5 ^a	6.1 ^a	7.3 ^a	8.3 ^a	4.7
Mean	1.1	1.8	2.4	3.2	4.0	4.8	5.4	3.3

Different letters indicate significant differences in by Tukey test at 0.05

Table 4. 8. Average of plant sibling number at three-week interval during 6-month development of *D. antarctica* ecotypes developed under 22°C

Ecotype	1st	2nd	3rd	4th	5th	6th	7th	Mean
Ardley	3.7 ^a	5.7 ^b	8.3 ^{b-c}	10.2 ^{c-d}	12.9 ^d	15.1 ^d	16.8 ^d	10.4
Galindez	2.7 ^f	5.1 ^d	7.4 ^c	10.3 ^c	12.4 ^e	14.5 ^e	16.2 ^e	9.8
King George	3.3 ^c	4.9 ^e	7.3 ^{c-d}	11.7 ^a	13.6 ^c	16.3 ^b	18.3 ^b	10.8
Lagotellerie (200m)	3.3 ^c	5.5 ^c	8.4 ^b	11.2 ^b	14.4 ^a	17.0 ^a	19.9 ^a	11.4
Lagotellerie (5m)	3.5 ^b	5.7 ^b	7.2 ^d	9.0 ^d	12.1 ^f	13.9 ^f	15.1 ^f	9.5
Livingston	3.1 ^d	6.0 ^a	8.7 ^a	11.7 ^a	13.9 ^b	15.7 ^c	17.3 ^c	10.9
Ridge	2.9 ^e	4.5 ^f	6.1 ^e	8.9 ^{d-e}	11.1 ^g	12.9 ^g	14.7 ^g	8.7
Mean	3.2	5.3	7.6	10.4	12.9	15.0	16.9	6.5

Different letters indicate significant differences in by Tukey test at 0.05

The growth of plants under varying temperature conditions for a period of six months showed a significant impact on reproductive abilities of siblings within ecotypes. Figure 4.13 and 4.14 showed the different responses of seven different *D. antarctica* ecotypes to the two different 12°C and 22°C temperature conditions.

In the Figure 4.15 the Lagotellerie (200 m) ecotype showed the greatest increase in the number of siblings when growing in 22°C. However, the Ridge ecotype exhibits a noteworthy rise in sibling quantity upon exposure to 12°C temperature (refer to the

Figure 4.13). It is noting that certain individuals within this ecotype may have fewer siblings compared to pots with a higher sibling count (Figure 4.16). This pattern highlights the sensitivity of different ecotypes to temperature, as seen in their sibling production dynamics as showed in the Figure 4.13 and 4.14.

The Lagotellerie (200 m) ecotype exhibits a greater proliferation of siblings under higher temperatures (22°C), while the Ridge ecotype tends to have higher sibling counts under lower temperatures (12°C). However, it is important to note that the Ridge ecotype has lower sibling counts compared to other ecotypes at 22°C, indicating variability across different growth conditions (Figure 4.15).

Climatic conditions have a significant impact on the distribution of *D. antarctica*, as highlighted by Parnikoza et al. (2011). The reproductive success of *D. antarctica* in warmer conditions could potentially be attributed to certain factors. One possible explanation is that high temperatures in the future might lead to morphological and anatomical alterations in Antarctic plants, allowing them to optimize light absorption and enhance growth, as proposed by Cavieres et al. (Cavieres et al., 2016).

During the period from 1964 to 1990, a study conducted by Fowbert et al (1994) focused on *D. antarctica*. The study revealed a significant increase in the population of *D. antarctica*, with its numbers growing by almost 25 times. This notable expansion in population was accompanied by an increase in the number of observed colonies. The researchers attribute this accelerated growth to the rising summer air temperatures in the maritime Antarctic. These warmer temperatures likely facilitated improved reproductive success for *D. antarctica*, potentially due to longer and more favorable growing seasons (Fowbert et al., 1994).

We observed the rise in sibling numbers at a temperature of 22°C corresponds with the findings of Fowbert's et al (1994). research. This correlation indicates a consistent pattern in the reproductive behavior of *D. antarctica*, highlighting its tendency for increased proliferation in response to higher temperatures, irrespective of the particular temperature range.

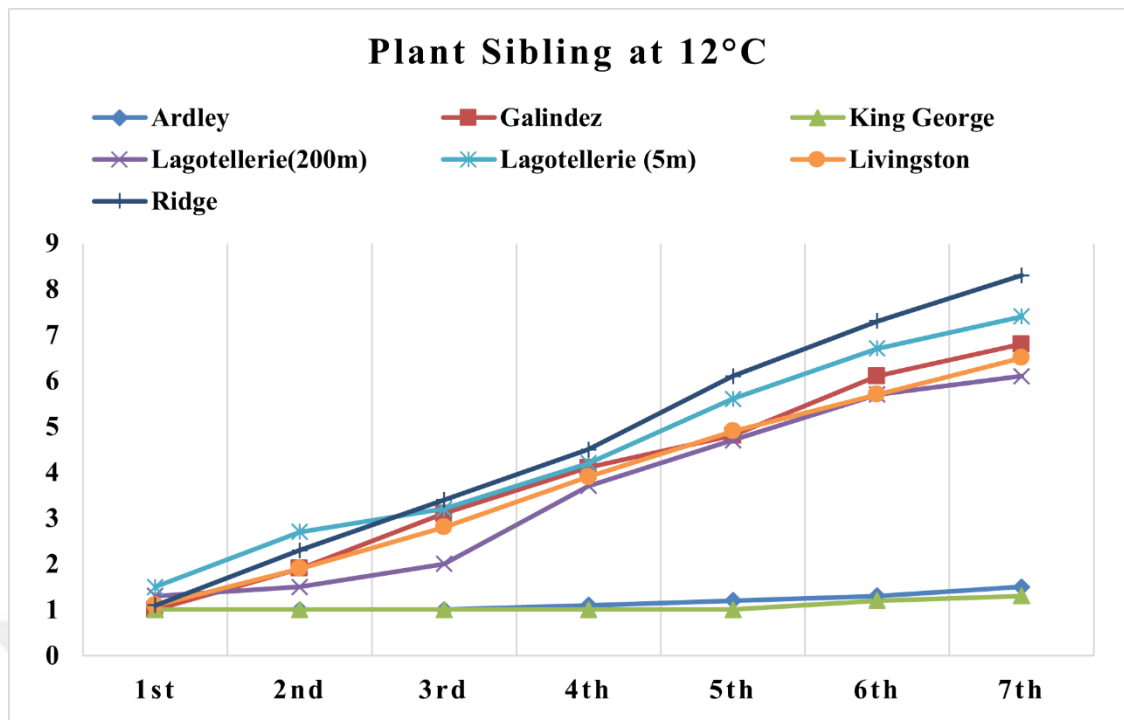


Figure 4. 13. Comparison of the plant sibling number at three-week interval during 6-month development of *D. antarctica* ecotypes grown in 12°C

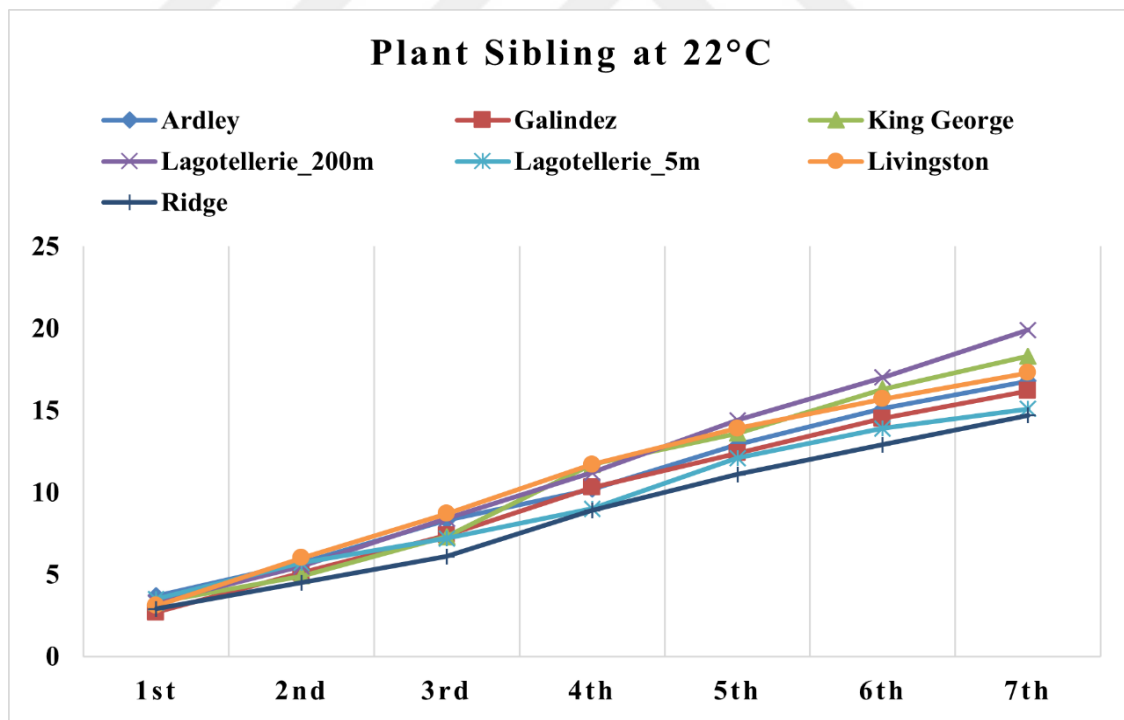


Figure 4. 14. Comparison of the plant sibling number at three-week interval during 6-month development of *D. antarctica* ecotypes grown in 22°C

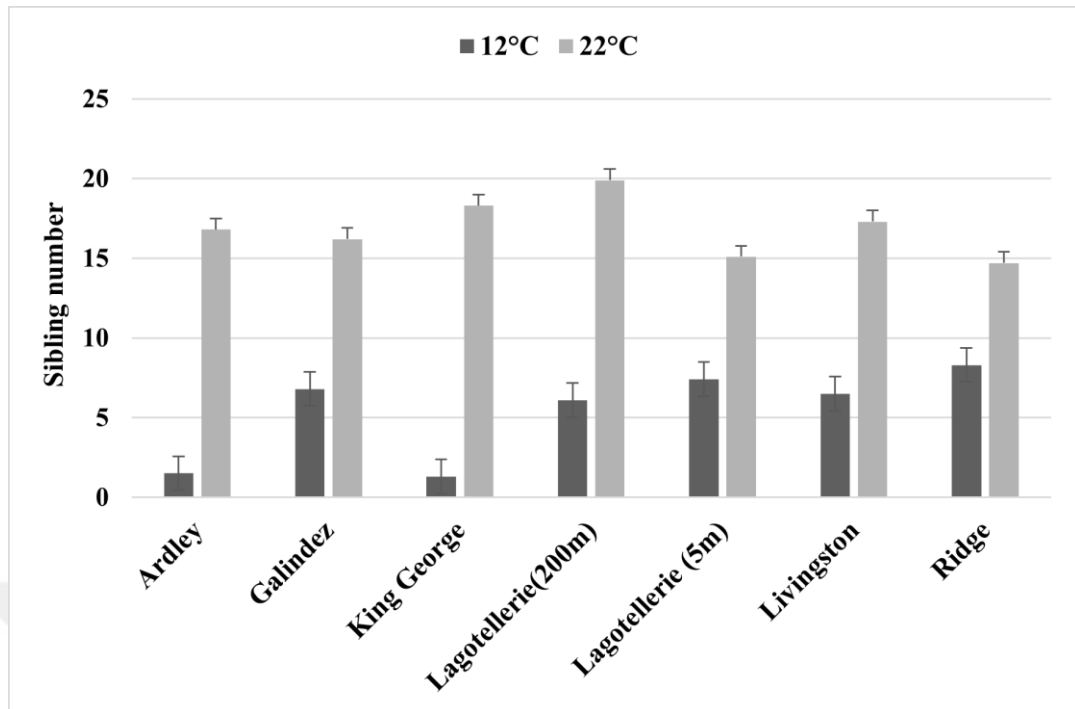


Figure 4. 15. Comparison of the final plant sibling number of *D. antarctica* ecotypes grown in two different temperature conditions

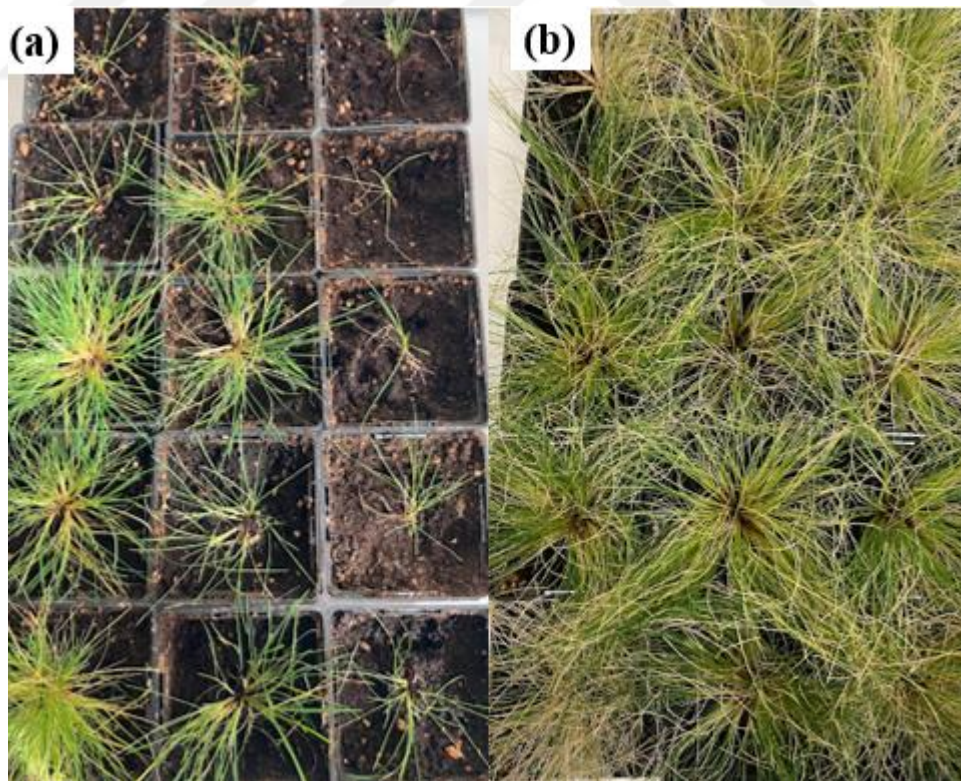


Figure 4. 16. An image of plants belonging to the Ridge ecotype grown under two different temperature condition (a: 12°C, b: 22°C)

The investigation conducted in our study regarding the differential sibling proliferation among distinct ecotypes under varying temperature conditions detailed in the Figure 4.13 and 4.14 aligns with the research conducted by Vera et al (2011). Their study focused on the demographic structure and colonization patterns of *D. antarctica* along altitudinal gradients (Vera et al., 2011). The results obtained from our study, specifically the contrasting sibling proliferation of the ecotypes under different temperature regimes, mirror the findings presented by Vera et al. (2011). Their study shed light on the dynamics of *D. antarctica* populations in environments characterized by specific altitudes and associated climate attributes, particularly the reduced snow cover during the early austral summers (Vera et al., 2011).

The connection between these studies implies a coherent relationship between temperature-induced responses in sibling proliferation, as observed in our study, and the ecotypic adaptations emphasized by Vera et al (2011). This correlation emphasizes the potential impact of temperature variations on the ecological dynamics and adaptive strategies of *D. antarctica* ecotypes across a range of habitats as illustrated in the Figures 4.13 and 4.14.

4.1.3 Plant Root Length

From the ANOVA table, the effect of two different 12°C and 22°C conditions, on the root length of *D. antarctica* ecotypes that were grown during 6-month period were observed. In the Tables 4.9 and 4.10, significant differences were observed in terms of ecotype and temperature interaction under both 12°C and 22°C conditions.

Table 4. 9. ANOVA table showing plant root length of *D. antarctica* ecotypes grown in 12°C

Source	DF	Mean Square	F value
Ecotype	6	8.434	3.24*
Error	14	2.599	
Total	20		
CV		18.292	

*p≤0.05, **p≤0.01

Table 4. 10. ANOVA table showing plant root length of *D. antarctica* ecotypes grown in 22°C

Source	DF	Mean Square	F value
Ecotype	6	15.096	3.84*
Error	14	3.930	
Total	20		
CV		16.905	

*p≤0.05, **p≤0.01

According to the results in Table 4.11, the root length percentage of *D. antarctica* ecotypes grown under two different 12°C and 22°C temperature conditions, during a 6-month period were examined. Table 4.11 provided the mean of root lengths of seven different *D. antarctica* ecotypes growth under two different temperature conditions, along with the overall mean root length for each ecotype. According to these results at 12°C, the Ardley ecotype had an average root length of 7.0 cm, while Galindez had a mean root length of 12.1 cm. At 22°C the Galindez ecotype showed the highest mean root length of 14.8 cm, while the King George ecotype exhibited the lowest root length at 9.6 cm.

Upon analyzing the overall mean root lengths between both temperature conditions, Galindez showed the highest mean root length, followed by Lagotellerie (200m). In contrast, Ridge had the lowest overall mean of root length.

Table 4. 11. Average root lengths (cm) values result of *D. antarctica* ecotypes developed under two different temperature conditions

Ecotype	12°C	22°C	Mean
Ardley	7.0 ^j	12.6 ^c	9.8
Galindez	12.1 ^{c-d}	14.8 ^a	13.4
King George	7.4 ⁱ	9.6 ^e	8.5
Lagotellerie (200m)	8.2 ^h	13.8 ^b	11.0
Lagotellerie (5m)	8.6 ^g	11.1 ^{d-e}	9.9
Livingston	9.4 ^{e-f}	11.8 ^d	10.6
Ridge	9.0 ^f	8.4 ^{g-h}	8.7
Mean	8.8	11.7	10.3

Different letters indicate significant differences in by Tukey test at 0.05

Figures 4.17, 4.18 and 4.19 show distinct responses of *D. antarctica* ecotypes to the temperatures 12°C and 22°C. The graphical images effectively showcasing how each of these seven ecotypes exhibited different reactions and growth patterns in direct dependence on specific temperature conditions.

The results indicate that root length is enhanced when exposed to higher temperature of 22°C compared to lower temperature of 12°C (Figure 4.19). This finding is consistent with a previous study conducted by Ruhland et al. (2010), which also observed a positive effect of elevated temperatures on the growth and root length of *D. antarctica* (Ruhland et al., 2010). The variations in growth were evident not just between plants that were cultivated under varying temperature conditions, but also among different ecotypes that were grown in the same controlled environment, as indicated in Figure 4.17 and 4.18.

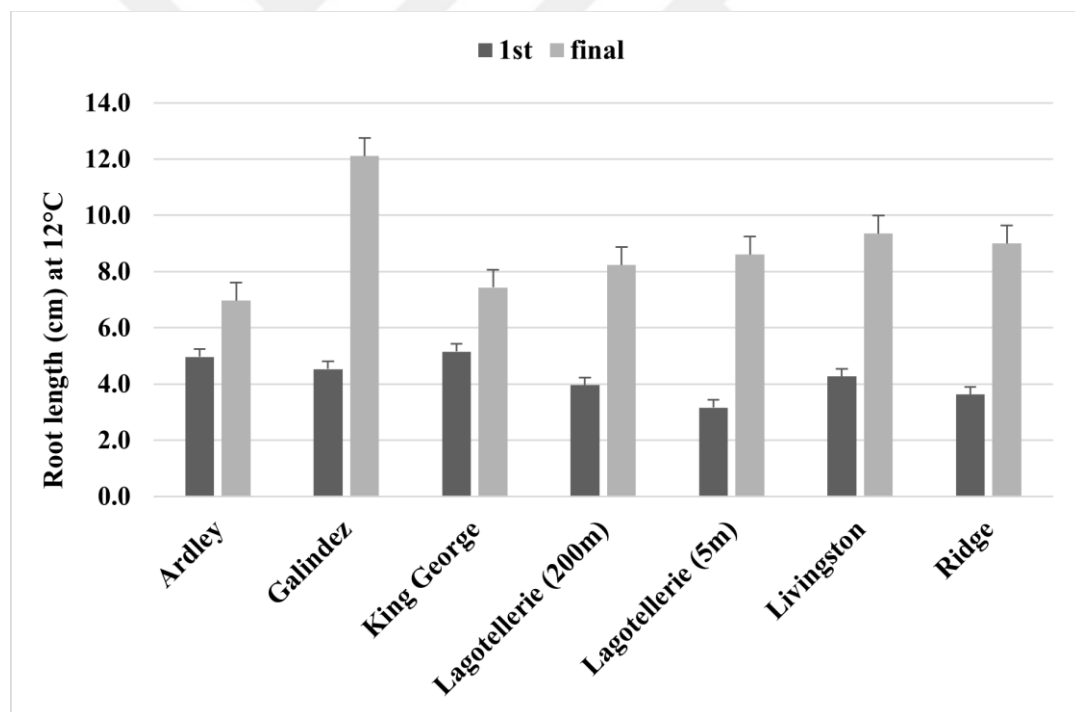


Figure 4. 17. The root length of *D. antarctica* ecotypes grown under 12°C

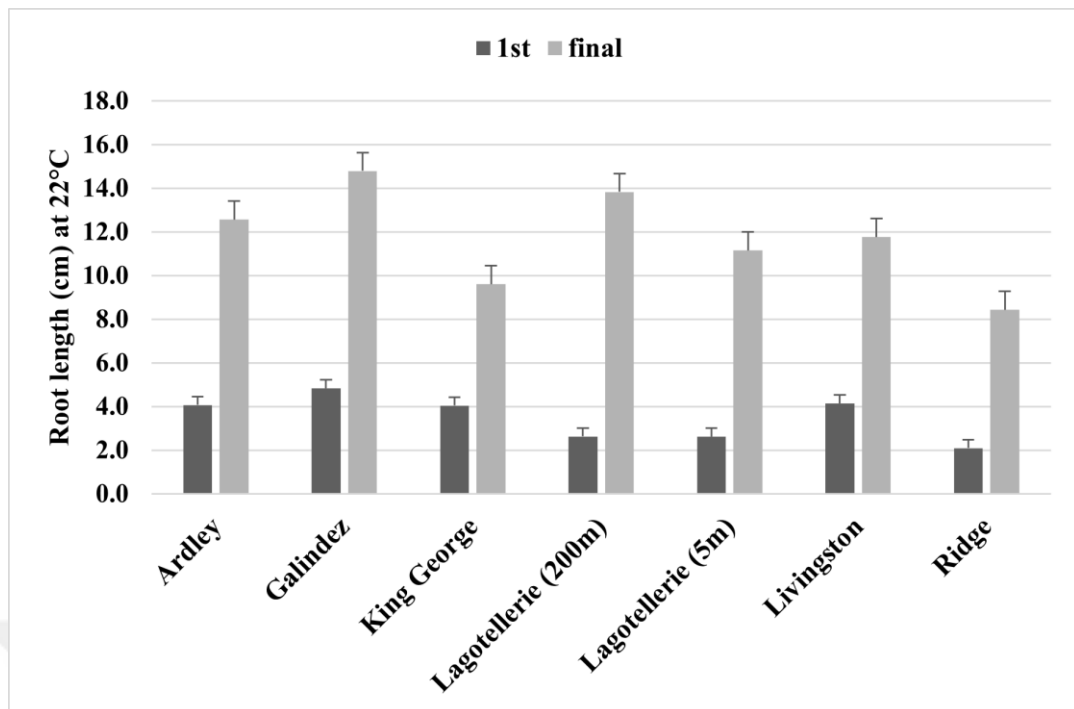


Figure 4. 18. The root length of *D. antarctica* ecotypes grown under 22°C

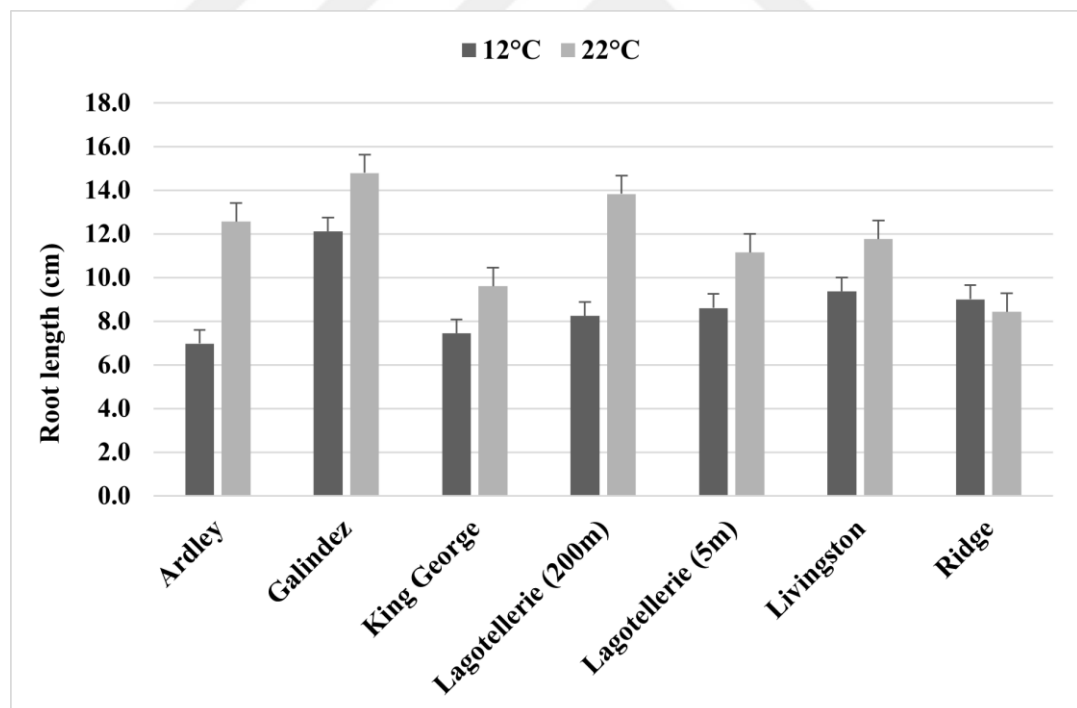


Figure 4. 19. Comparison of the root length of *D. antarctica* ecotypes grown under two different temperature conditions

Ecotypes grown at a temperature of 22°C showed almost the same height, size and length of the roots in each pot (Figure 4.20 a). In contrast, those that were grown at

12°C showed marked differences in growth rate and other features, as shown in Figure 4.21.

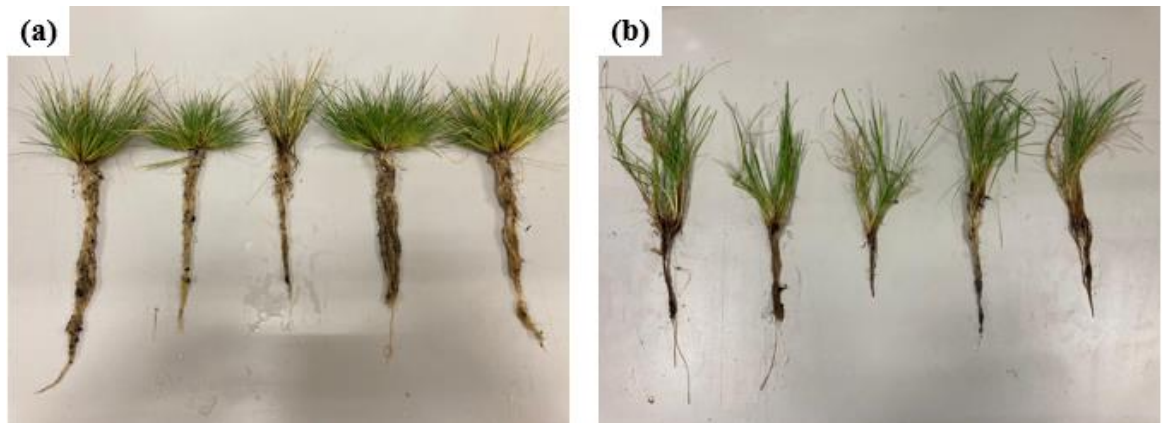


Figure 4. 20. An image of plants belonging to the Galindez ecotype grown under two different temperature conditions (a: 22°C, b: 12°C)



Figure 4. 21. An image of plants belonging to the Ridge ecotype grown under 12°C

4.1.4 Plant Leaf Fresh Weight

The results of this study as recorded in the ANOVA tables shows the effects of two different temperature conditions on leaf fresh weight of *D. antarctica* ecotypes. According to the results, the temperature and ecotype interaction were found to be significant for plants grown under 12°C and 22°C conditions.

Table 4. 12. ANOVA table showing leaf fresh weight of *D. antarctica* ecotypes grown in 12°C

Source	DF	Mean Square	F value
Ecotype	6	0.262	3.30*
Error	14	0.079	
Total	20		
CV		68.967	

*p≤0.05, **p≤0.01

Table 4. 13. ANOVA table showing leaf fresh weight of *D. antarctica* ecotypes grown in 22°C

Source	DF	Mean Square	F value
Ecotype	6	536.040	286.32**
Error	14	1.872	
Total	20		
CV		13.853	

*p≤0.05, **p≤0.01

A representation of the mean values of leaf fresh weight percentage of *D. antarctica* ecotypes after 6-month growth under two different temperature conditions, 12°C and 22°C, are in Table 4.14. As can be understood from this table, plants showing robust growth at 22°C tend to exhibit diminished performance at 12°C. Upon examining the results, it was found that the average leaf fresh weight all ecotypes in the 22°C condition was 9.9 g, while the average for all ecotypes in 12°C condition was 0.4 g.

The data from the table highlight a clear pattern where in the Galindez ecotype consistently displayed the highest leaf fresh weight among all ecotypes when grown at 22°C (Figure 4.23). Conversely regardless of the temperature setting, the King George ecotypes displayed the lowest leaf fresh weight regardless of the temperature setting was 12°C or 22°C (Figure 4.24).

The Ridge ecotype stood out among the ecotypes grown at 12°C, demonstrating exceptional performance with highest leaf fresh weight as showed in the Figure 4.22.

This robust growth wasn't limited to just leaf fresh weight; the ecotype also showed significant sibling proliferation, highlighting its robust growth and reproductive vigor under the 12°C condition compared to other ecotypes in the same environment. These findings highlight the resilience and adaptability of the Ridge ecotype to lower temperatures, which could hold valuable insights for conservation and cultivation efforts involving this particular ecotype.

Table 4. 14. Average leaf-fresh weight values result of *D. antarctica* ecotypes developed under two different temperature conditions

Ecotype	12°C	22°C	Mean
Ardley	0.1 ⁱ	4.8 ^d	2.4
Galindez	0.3 ^h	40.1 ^a	20.2
King George	0.1 ⁱ	3.7 ^e	1.9
Lagotellerie (200m)	0.7 ^{g-h}	3.5 ^{e-f}	2.1
Lagotellerie (5m)	0.7 ^{g-h}	5.3 ^c	3.0
Livingston	0.3 ^h	4.7 ^{d-e}	2.5
Ridge	0.8 ^g	7.2 ^b	4.0
Mean	0.4	9.9	5.1

Different letters indicate significant differences in by Tukey test at 0.05

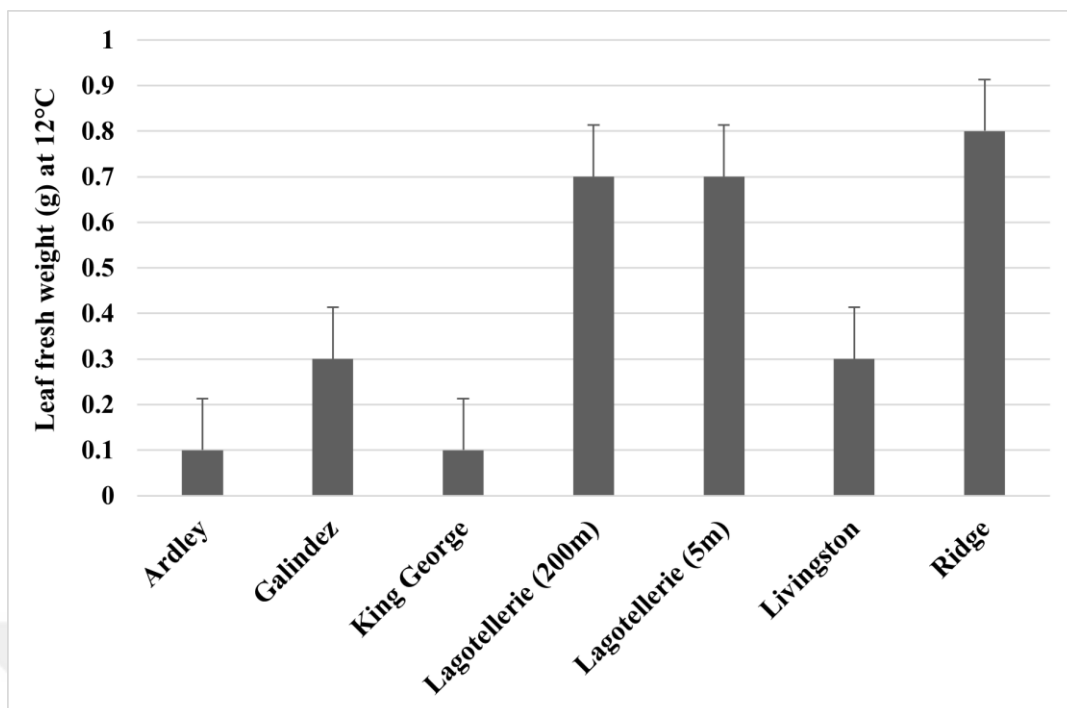


Figure 4. 22. Leaf-fresh weight of *D. antarctica* ecotypes grown under 12°C

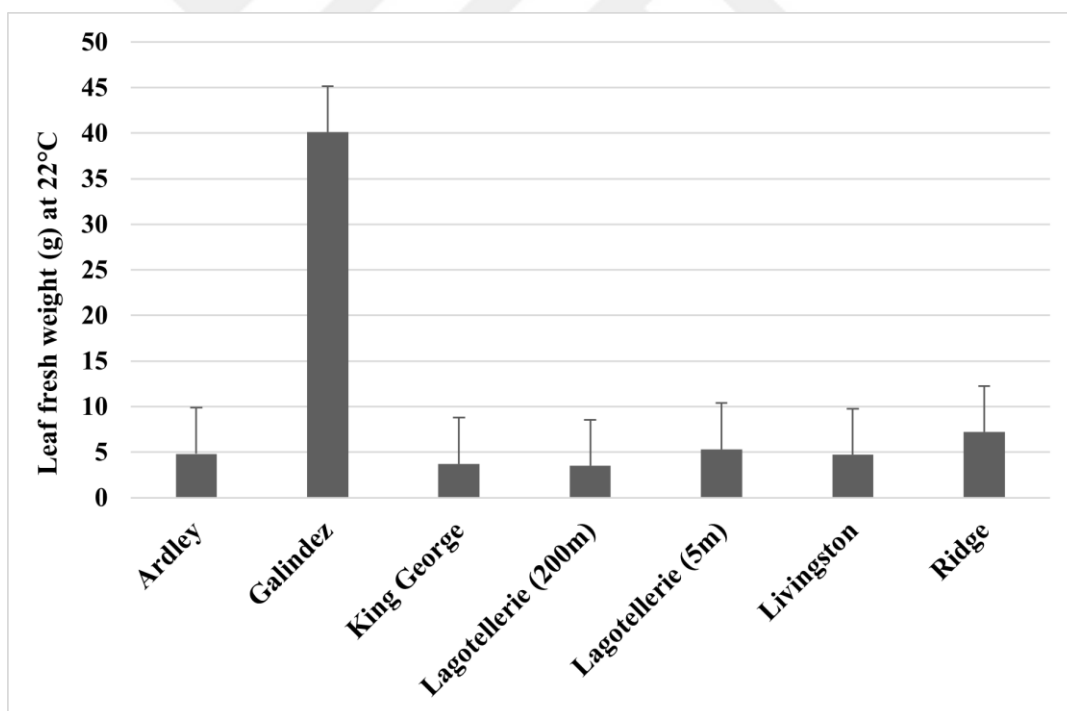


Figure 4. 23. Leaf-fresh weight of *D. antarctica* ecotypes grown under 22°C

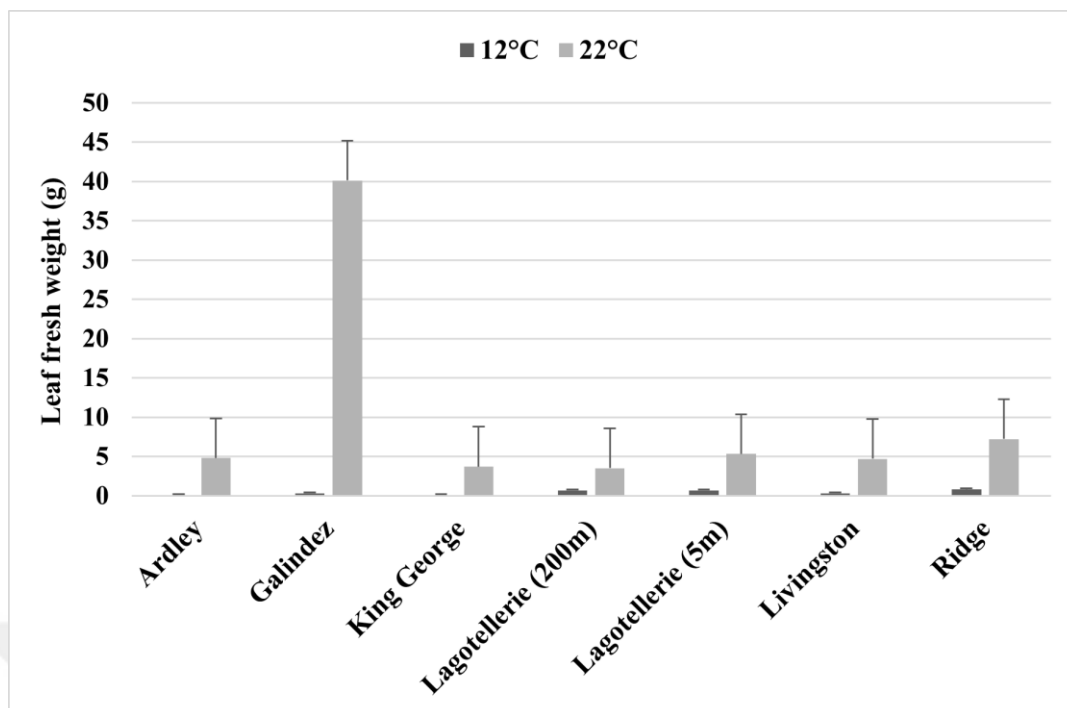


Figure 4. 24. Leaf-fresh weight of *D. antarctica* ecotypes grown under two different temperature conditions

4.1.5 Plant Leaf Dry Weight

The results of the ANOVA are documented in Table 4.15 and 4.16 showing the effects of two different temperature on leaf dry weight of *D. antarctica* ecotypes. Significant differences were observed in the temperature and ecotype interaction under both temperature conditions, 12°C and 22°C.

Table 4. 15. ANOVA table showing leaf dry weight of *D. antarctica* ecotypes grown in 12°C

Source	DF	Mean Square	F value
Ecotype	6	0.011	2.58
Error	14	0.004	
Total	20		
CV		82.512	

* $p \leq 0.05$, ** $p \leq 0.01$

Table 4. 16. ANOVA table showing leaf dry weight of *D. antarctica* ecotypes grown in 22°C

Source	DF	Mean Square	F value
Ecotype	6	0.221	3.81*
Error	14	0.058	
Corrected Total	20		
CV		23.299	

*p≤0.05, **p≤0.01

Table 4.17 represents the mean values of leaf dry weight percentage of *D. antarctica* ecotypes grown under two different temperature conditions. Each ecotype is listed along with its corresponding leaf dry weight (in grams) recorded under both temperature conditions.

At 12°C, ecotypes showed varying levels of development, with Ardley, Lagotellerie (200 m), Lagotellerie (5 m) and Galindez displayed leaf fresh weights ranging 0.0 to 0.1 g. Livingston and King George ecotypes showed no discernible increase in leaf dry weight at this temperature, while Ridge ecotype had a slightly higher average leaf dry weight of 0.2 g. Conversely, at 22°C, there was a general increase in leaf dry weight between ecotypes. Ardley, King George and Livingston ecotypes showed average leaf dry weights of 1.1 g, demonstrating notable growth compared to their counterparts at 12°C. Galindez and Lagotellerie (200 m) ecotypes also showed increased development, with average leaf dry weights of 0.7 g and 0.8 g, respectively. The Lagotellerie (5 m) ecotype exhibited the most significant growth at this temperature, with an average leaf dry weight of 1.1 g, along with Ridge ecotype exhibited a notable increase to 1.5 g.

The overall mean leaf dry weight between ecotypes at 12°C was 0.1 g, while at 22°C, it rose to 1.0 g. The results indicate a significant improvement in growth under the higher temperature conditions as illustrated in Figures 4.25, 4.26 and 4.27.

Table 4. 17. Average leaf-dry weight values result of *D. antarctica* ecotypes developed under two different temperature conditions

Ecotype	12°C	22°C	Mean
Ardley	0.0 ^g	1.1 ^b	0.5
Galindez	0.1 ^f	0.7 ^d	0.4
King George	0.0 ^g	0.9 ^c	0.4
Lagotellerie (200m)	0.1 ^f	0.8 ^{c-d}	0.5
Lagotellerie (5m)	0.1 ^f	1.1 ^b	0.6
Livingston	0.0 ^g	1.1 ^b	0.6
Ridge	0.2 ^e	1.5 ^a	0.9
Mean	0.1	1.0	0.55

Different letters indicate significant differences in by Tukey test at 0.05

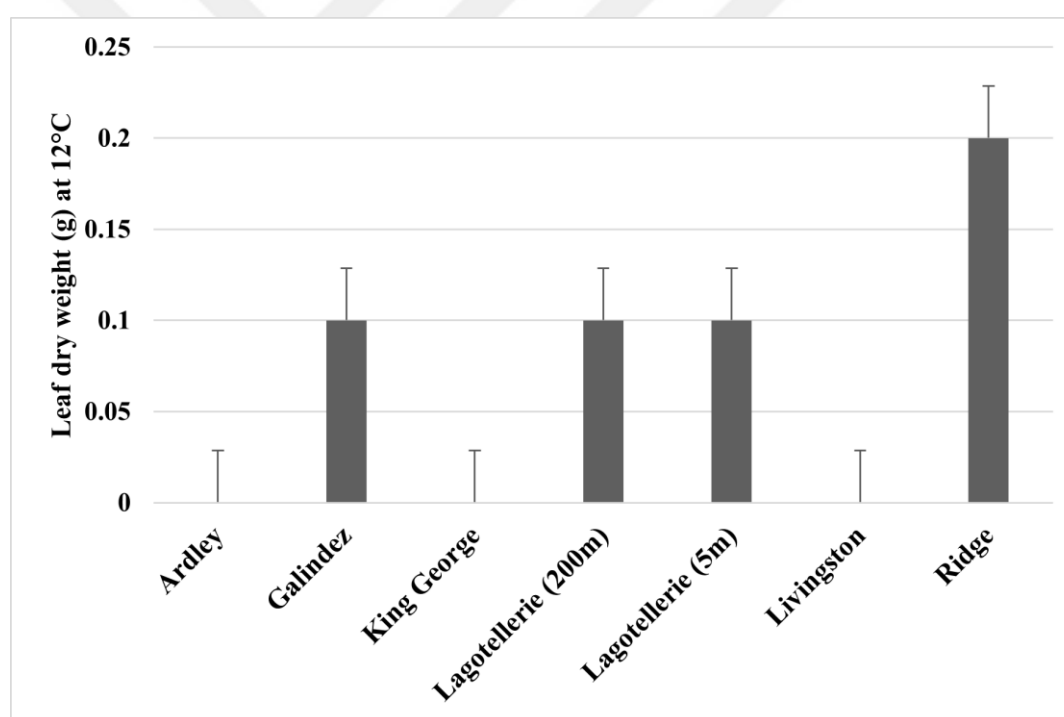


Figure 4. 25. Leaf-dry weight of *D. antarctica* ecotypes grown under 12°C

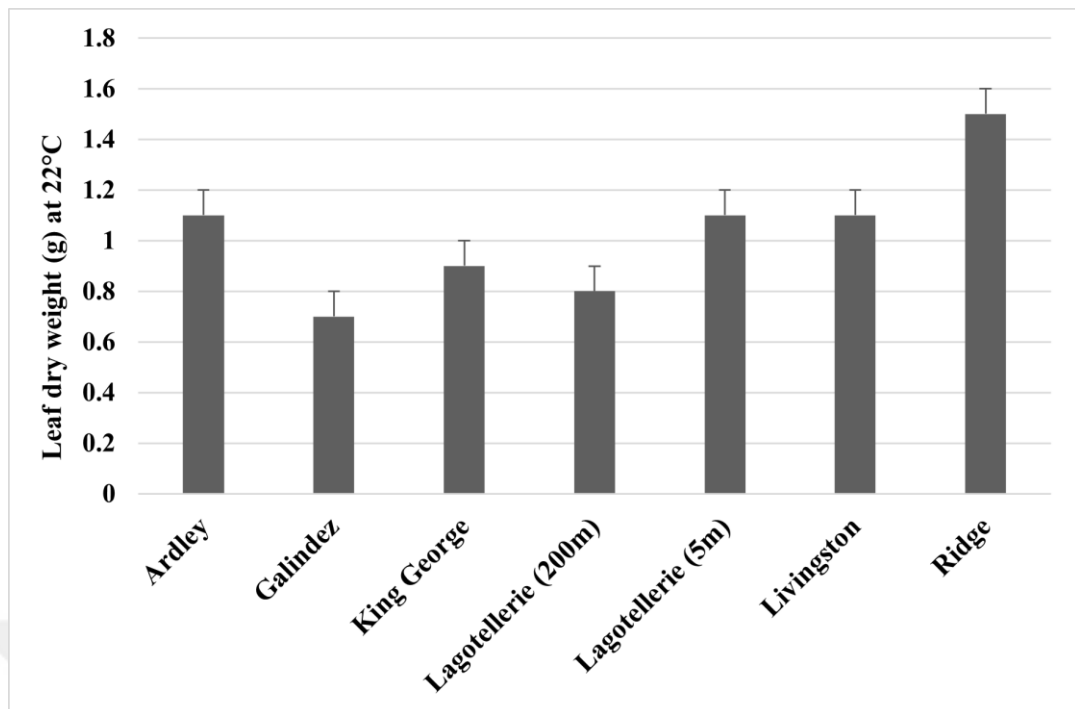


Figure 4. 26. Leaf-dry weight of *D. antarctica* ecotypes grown under 22°C

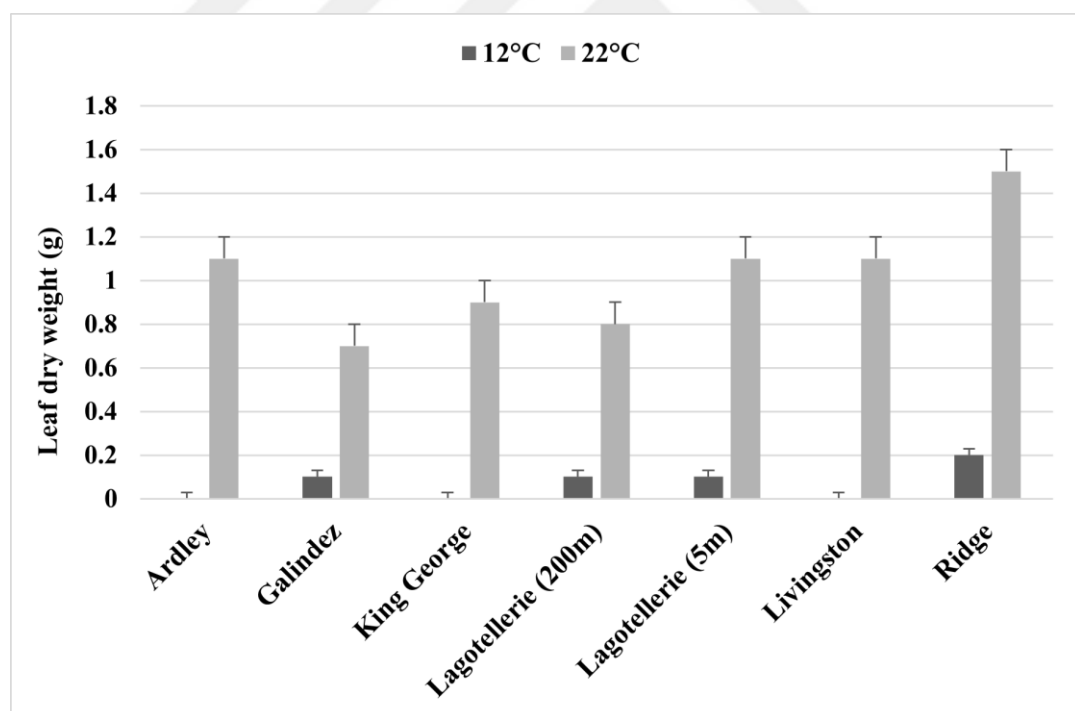


Figure 4. 27. Leaf-dry weight of *D. antarctica* ecotypes grown under two different temperature conditions

These findings related with a leaf fresh and dry weight suggest an ecotype-dependent response to temperature variations in *D. antarctica*. Some ecotypes thrive in warmer

conditions, while other display limited growth across temperature ranges. Understanding such temperature-related variations can offer valuable insights for conservation strategies and agricultural practices concerning these ecotypes, facilitating informed decision-making for their management and preservation.

4.1.6 Plant Root Fresh Weight

The ANOVA tables show the effect of temperature on root fresh weight percentage of *D. antarctica* ecotypes grown under two different temperature conditions, 12°C and 22°C. Table 4.18 and 4.19 show that significant differences were noticed in temperature and ecotype interaction under both temperature conditions.

Table 4. 18. ANOVA table showing root fresh weight of *D. antarctica* ecotypes grown in 12°C

Source	DF	Mean Square	F value
Ecotype	6	0.062	2.42
Error	14	0.025	
Total	20		
CV		72.227	

*p≤0.05, **p≤0.01

Table 4. 19. ANOVA table showing root fresh weight of *D. antarctica* ecotypes grown in 22°C

Source	DF	Mean Square	F value
Ecotype	6	0.517	2.80
Error	14	0.184	
Total	20		
CV		29.541	

*p≤0.05, **p≤0.01

Table 4.20 presents the average root fresh weight values of seven different ecotypes of *D. antarctica* grown under two different temperature conditions: 12°C and 22°C. Each value in the table shows the root fresh weight measurement of a specific ecotype at the given temperature condition. From the given data its clear understood

that ecotypes respond differently to temperature changes in terms of root fresh weight. At 12°C, ecotypes have root fresh weights ranging from 0.1 g to 0.4 g, in Lagotellerie (200 m) and Ridge have higher values at 0.4 g. On the other hand, at 22°C, there is a significant increase in root fresh weight, with ecotypes Ardley, Galindez, Livingston and Ridge showing values between 1.6 g and 1.9 g. This indicates a temperature-induced increase in root biomass.

According to the results of the Table 4.20, Ardley ecotype presented interesting variations in root weights when subjected to the 22°C. It demonstrated the highest in 22°C and lowest root fresh weight compared to other ecotypes in its category in 12°C conditions as detailed in the Figures 4.28, 4.29 and 4.30.

Table 4. 20. Average root-fresh weight values result of *D. antarctica* ecotypes developed under two different temperature conditions

Ecotype	12°C	22°C	Mean
Ardley	0.1 ^h	1.9 ^a	1.0
Galindez	0.2 ^g	1.6 ^{b-c}	0.9
King George	0.1 ^h	0.7 ^e	0.4
Lagotellerie (200m)	0.4 ^f	1.4 ^c	0.9
Lagotellerie (5m)	0.3 ^{f-g}	1.1 ^d	0.7
Livingston	0.1 ^h	1.7 ^b	0.9
Ridge	0.4 ^f	1.7 ^b	1.0
Mean	0.2	1.5	0.8

Different letters indicate significant differences in by Tukey test at 0.05

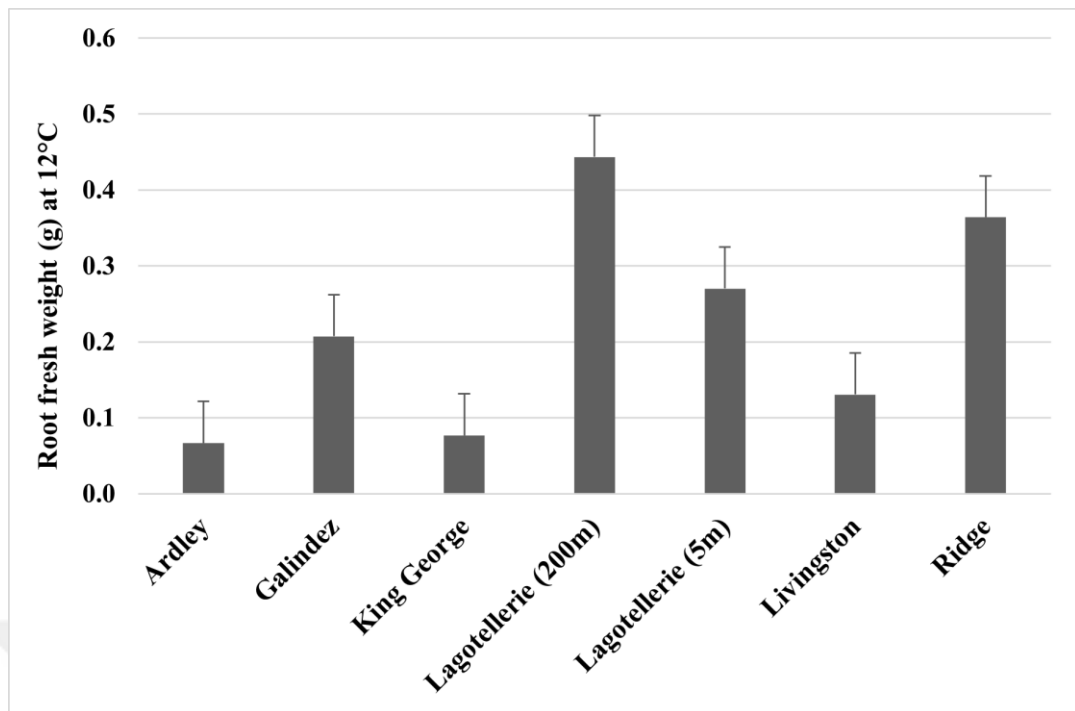


Figure 4. 28. Root-fresh weight of *D. antarctica* ecotypes grown under 12°C

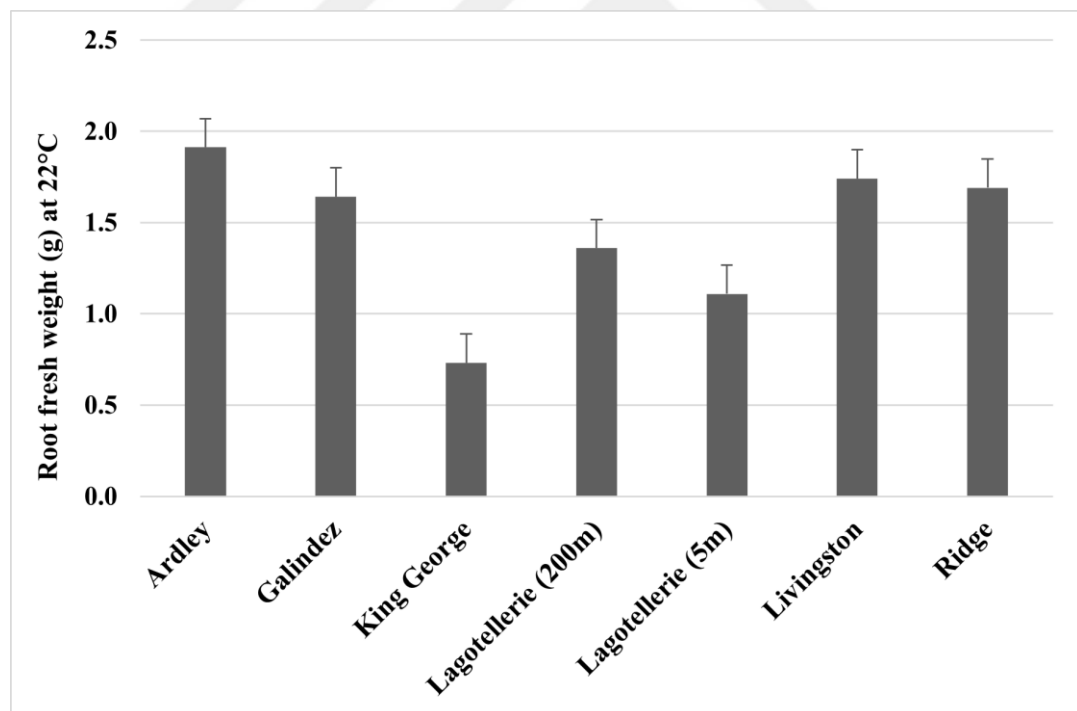


Figure 4. 29. Root-fresh weight of *D. antarctica* ecotypes grown under 22°C

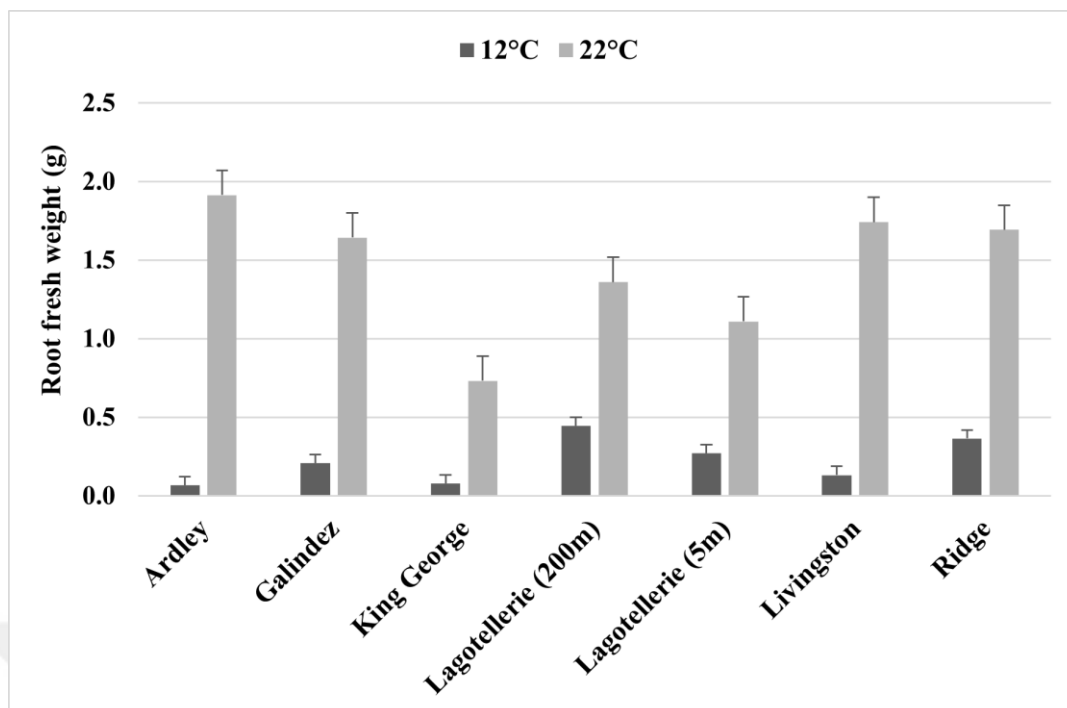


Figure 4. 30. Comparison of the root-fresh weight of *D. antarctica* ecotypes grown under two different temperature conditions

4.1.6 Plant Root Dry Weight

The provided Tables 4.21 and 4.22 present the results of ANOVA for root dry weight after 6-month development of *D. antarctica* ecotypes grown under two different temperature conditions. The data reveals significant differences observed in the interaction between temperature and ecotype under both temperature conditions. .

Table 4. 21. ANOVA table showing root dry weight of *D. antarctica* ecotypes grown in 12°C

Source	DF	Mean Square	F value
Ecotype	6	0.000	1.82
Error	14	0.000	
Total	20		
CV		96.738	

* $p \leq 0.05$, ** $p \leq 0.01$

Table 4. 22. ANOVA table showing root dry weight of *D. antarctica* ecotypes grown in 22°C

Source	DF	Mean Square	F value
Ecotype	6	0.004	3.38*
Error	14	0.001	
Total	20		
CV		27.267	

* $p \leq 0.05$, ** $p \leq 0.01$

In the Table 4.23 showed the average root dry weight values of various ecotypes *D. antarctica* grown under two different temperature conditions. This analysis is essential for understanding the ecotypes reactions to temperature variations and their adaptability within Antarctic environments. The data analysis results once more showed that the impact of two different temperature conditions on root dry weight of plants varied.

Notably, at 12°C, all ecotypes displayed a root dry weight of 0.0 g, indicating limited root development under this cooler condition. In contrast, at 22°C, several ecotypes such as Ardley, Galindez and Ridge maintained a root dry weight of 0.2 g. This suggests that these ecotypes have ability to adaptations to higher temperatures, allowing for more substantial root growth compared to the cooler conditions (Figure 4.31).

Table 4. 23. Average root-dry weight values result of *D. antarctica* ecotypes developed under two different temperature conditions

Ecotype	12°C	22°C	Mean
Ardley	0.0 ^c	0.2 ^a	0.1
Galindez	0.0 ^c	0.2 ^a	0.1
King George	0.0 ^c	0.1 ^b	0.1
Lagotellerie (200m)	0.0 ^c	0.1 ^b	0.1
Lagotellerie (5m)	0.0 ^c	0.1 ^b	0.1
Livingston	0.0 ^c	0.1 ^b	0.1
Ridge	0.0 ^c	0.2 ^a	0.1
Mean	0.0	1.1	0.1

Different letters indicate significant differences in by Tukey test at 0.05

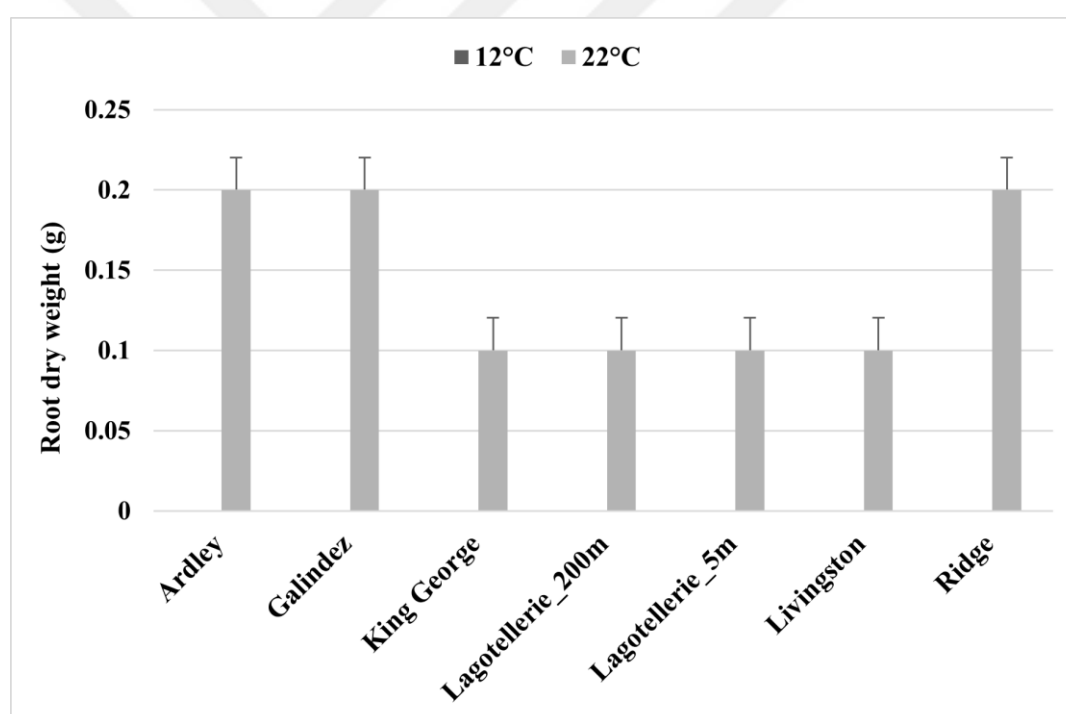


Figure 4. 31. Comparison of the root-dry weight of *D. antarctica* ecotypes grown under two different temperature conditions

To summarize, the examination of various ecotypes in different temperature conditions highlights the intricate reactions of Antarctica plant. Although certain ecotypes displayed unique characteristics in terms of leaf and root weights, the study aligns with Xiong et al.'s findings, indicating a consistent growth pattern across diverse temperature regimes (Xiong et al., 2000). These parallel observations

suggest a shared physiological response among Antarctica plants: *D. antarctica* and *C. quitensis* towards temperature fluctuations, emphasizing the significance of understanding their adaptive strategies in the context of changing environmental conditions. Further investigation into the mechanisms driving these consistent growth patterns could provide valuable insights into the adaptive resilience of Antarctic vascular plants in a warming climate (Xiong et al., 2000). This deeper understanding is crucial for predicting and managing the impacts of climate change on Antarctica ecosystems, ensuring the preservation and sustainability of these unique species.

4.1.7 Plant Chlorophyll Content

The variance analysis for chl *a*, chl *b* and total chl content after 6-month development of *D. antarctica* ecotypes grown under two different temperature conditions are given in Tables 4.24 and 4.25.

Table 4. 24. ANOVA table showing Chlorophyll content of *D. antarctica* ecotypes grown in 12°C

Source	DF	Chl <i>a</i> **	Chl <i>b</i> **	Total chl**
Ecotype	4	0.066	0.019	0.146
Error	10	0.007	0.003	0.018
Total	24			
CV		7.919	11.218	8.511

* $p \leq 0.05$, ** $p \leq 0.01$

Table 4. 25. ANOVA table showing Chlorophyll content of *D. antarctica* ecotypes grown in 22°C

Source	DF	Chl <i>a</i> **	Chl <i>b</i> **	Total chl**
Ecotype	4	0.216	0.096	0.586
Error	10	0.020	0.005	0.037
Total	24			
CV		14.921	15.455	13.659

* $p \leq 0.05$, ** $p \leq 0.01$

The mean of Chlorophyll content data presented in Tables 4.26 and 4.27 provide a detailed understanding of the chl *a*, chl *b* and total chl production comparison between different ecotypes of *D. antarctica* under two different temperature conditions. It's important to note that the leaves of King George and Ardley ecotypes, which were grown at 12°C temperature condition were not utilized for chlorophyll analysis due to a limited number of sibling samples (refet to the Figure 4.15).

Table 4.26, reveals variations in chl *a*, chl *b* and total chl of the five ecotypes: Galindez, Lagotellerie (200 m), Lagotellerie (5 m), Livingston and Ridge grown at 12°C. On the other hand, Table 4.27 includes the variations in chl content of all seven ecotypes grown in 22°C. These differences highlight the unique responses of each ecotype to environmental stimuli, providing insights into their adaptivity in the context of changing temperatures. It's clearly understood from results that some ecotypes displayed higher chl content at one temperature compared to another, indicating varying thermal acclimatization capabilities.

For instance, ecotypes such as Galindez demonstrate varying chl *a*, chl *b* and total chl suggesting adaptability to temperature changes, because Galindez shows reduced chl *a*, chl *b* and total chl content at 22°C compared to 12°C. Conversely, ecotypes like Livingston and Ridge exhibit increased chl *a*, chl *b* and total chl content at 22°C, possibly indicating optimal thermal conditions or improved photosynthetic efficiency.

Additionally, the variations in chl content across ecotypes under different temperature conditions reflect their responses to thermal changes. Analyzing the average chl content values across *D. antarctica* ecotypes at 12°C and 22°C, provides a comprehensive view of chl production patterns, contributing to a holistic understanding of their physiological responses to varying thermal environments (Figure 4.32 and 4.33).

Table 4. 26. Average Chlorophyll content values result of *D. antarctica* ecotypes developed under 12°C

Ecotype	Chl <i>a</i> **	Chl <i>b</i> **	Total chl**	Mean
Galindez	1.3 ^a	0.6 ^a	1.9 ^a	1.2
Lagotellerie (200m)	1.1 ^c	0.5 ^b	1.6 ^c	1.1
Lagotellerie (5m)	0.9 ^d	0.4 ^c	1.3 ^d	0.8
Livingston	1.2 ^b	0.5 ^b	1.7 ^b	1.1
Ridge	1.1 ^c	0.5 ^b	1.6 ^c	1.1
Mean	1.1	0.5	1.6	1.1

Different letters indicate significant differences in by Tukey test at 0.05

Table 4. 27. Average Chlorophyll content values result of *D. antarctica* ecotypes developed under 22°C

Ecotype	Chl <i>a</i> **	Chl <i>b</i> **	Total chl**	Mean
Ardley	0.8 ^d	0.3 ^d	1.1 ^e	0.7
Galindez	1.0 ^c	0.5 ^b	1.5 ^d	1.0
King George	0.7 ^e	0.4 ^c	1.1 ^e	0.7
Lagotellerie (200m)	0.6 ^f	0.3 ^d	0.9 ^f	0.6
Lagotellerie (5m)	1.0 ^c	0.5 ^b	1.6 ^c	1.1
Livingston	1.4 ^a	0.8 ^a	2.2 ^a	1.4
Ridge	1.2 ^b	0.5 ^b	1.7 ^b	1.1
Mean	1.0	0.5	1.4	1.1

Different letters indicate significant differences in by Tukey test at 0.05

In conclusion, our study revealed significant influences of both temperature and ecotype on chlorophyll content, with a notable interaction effect between the two factors. These findings indicate that both temperature and ecotype play crucial roles in affecting chlorophyll content, and their combined effect has a notable impact on specific chlorophyll types as chl *a*, chl *b* and total chl in *D. antarctica* ecotypes as illustrated in Figures 4.34, 4.35 and 4.36.

Specifically, ecotypes such as Galindez and Lagotellerie (200 m), at higher temperature (22°C) tended to result in lower chlorophyll concentration compared to

measurements at 12°C as detailed in the Figure 4.34, 4.35 and 4.36. This corresponds with the study conducted by Xiong et al. (2000) which found that the rates of photosynthesis per leaf area were found to be highest at 12°C; however, plants grown at 20°C compensated for this by significantly increasing their leaf-area production. Despite limited evidence of photosynthetic acclimation with increasing growth temperatures, *D. antarctica* fully acclimated its dark respiration rates across varying temperature conditions. *D. antarctica* maintained higher leaf chlorophyll concentrations at 12°C than 7°C or 20°C, without significant changes in chlorophyll *a:b* ratios (Xiong et al., 2000).

The results in the study conducted by Sáez et al. (2019), aligns with our findings regarding the optimal temperature range for chlorophyll production in *D. antarctica* plants. They observed that *D. antarctica* exhibited the highest chlorophyll (*a+b*) content at moderate temperatures of 10°C and 16°C. This observation is consistent with our study, where ecotypes as Galindez and Lagotellerie (200m) grown at 12°C (similar to the moderate temperatures observed by Sáez et al. (2019) showed favorable chlorophyll levels.

On the other hand, ecotypes such as Lagotellerie (5 m), Livingston and Ridge exhibited the highest chlorophyll content at 22°C as detailed in Figure 4.32, indicative of their specific temperature preferences or adaptations. These variations highlight the intricate nature of plant responses to temperature and emphasize the importance of genetic diversity and local adaptations in ecological research.

The study by Gielwanowska et al. (2005) focused on investigating the photosynthetic behaviors of two Antarctic vascular plants, *C. quitensis* and *D. antarctica*, near Palmer Station on the Antarctic Peninsula. An interesting finding emerged during the study: these plant species displayed minimal net photosynthetic rates on warm, sunny days when the temperature in the canopy air exceeded 20°C. Conversely plants exhibited higher rates of photosynthesis on cooler days when the temperature in the canopy air dropped below 10°C. This temperature-dependent response was a significant characteristic observed in both *C. quitensis* and *D. antarctica* within environment of Antarctic Peninsula (Gielwanowska et al., 2005). Significantly, our finding confirms these results, particularly in the ecotype –

Lagotellerie (200 m) – where the observed trend indicates a decrease in chlorophyll concentration with higher temperatures, similar to the findings in the previous studies by Xiong et al. (2000) and Gielwanowska et al. (2005).

These findings highlight the presence of an ideal temperature range, approximately between 10°C and 16°C, conducive to optimal chlorophyll production in *D. antarctica* plants. However, it's important to note that certain ecotypes may exhibit specific temperature preferences or adaptations, leading to variations in chlorophyll content among them. These variations in chlorophyll content among different ecotypes emphasize the intricate nature of plant responses to temperature and the significance of considering genetic diversity and local adaptations in ecological research.

For instance, ecotypes such as Galindez, Livingston and Ridge exhibit distinct chlorophyll concentration at both temperatures as shown in the Figure 4.32 and 4.33, the results imply that these ecotypes exhibit distinct responses to changes in their surroundings, specifically when it comes to temperature fluctuations. The differences in chlorophyll levels observed among these ecotypes at varying temperatures suggest that their chlorophyll production is significantly affected by environmental factors. The fluctuations in the environment, especially temperature changes, are likely to trigger physiological responses within these ecotypes, which in turn affect the synthesis or breakdown of chlorophyll. Further exploration and examination of the precise physiological and genetic mechanisms underlying these temperature responses in various *D. antarctica* ecotypes can offer valuable insights into their niche preferences and adaptive strategies in the context of changing environmental conditions, including climate change.

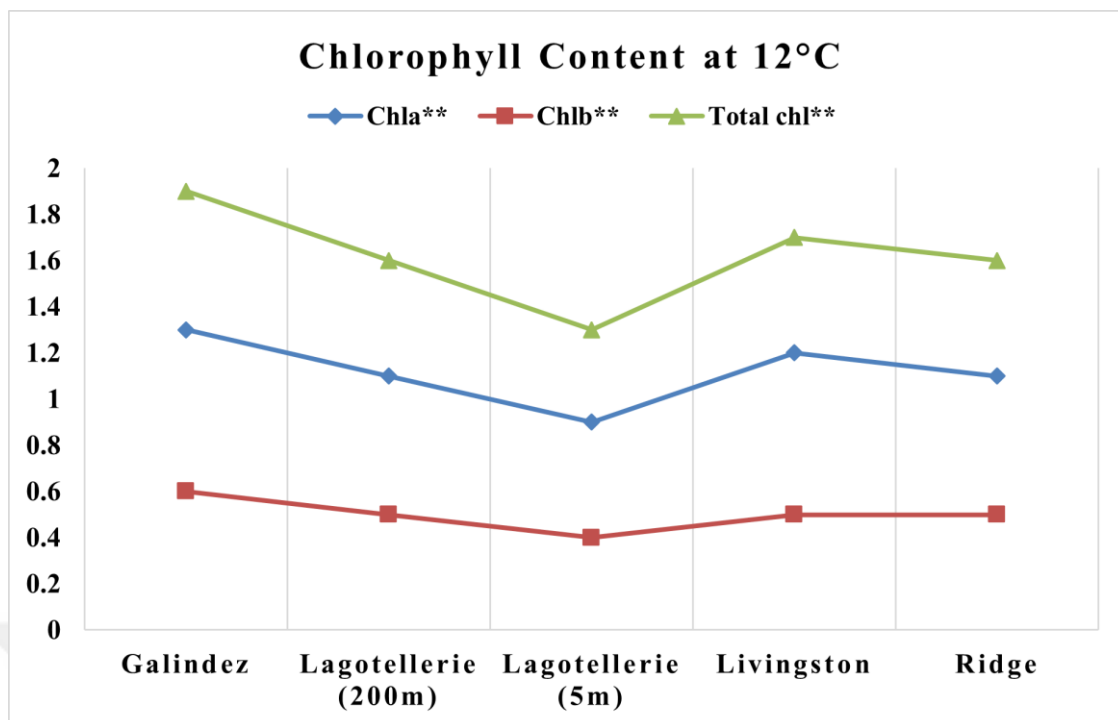


Figure 4. 32. Chlorophyll content of *D. antarctica* ecotypes grown under 12°C

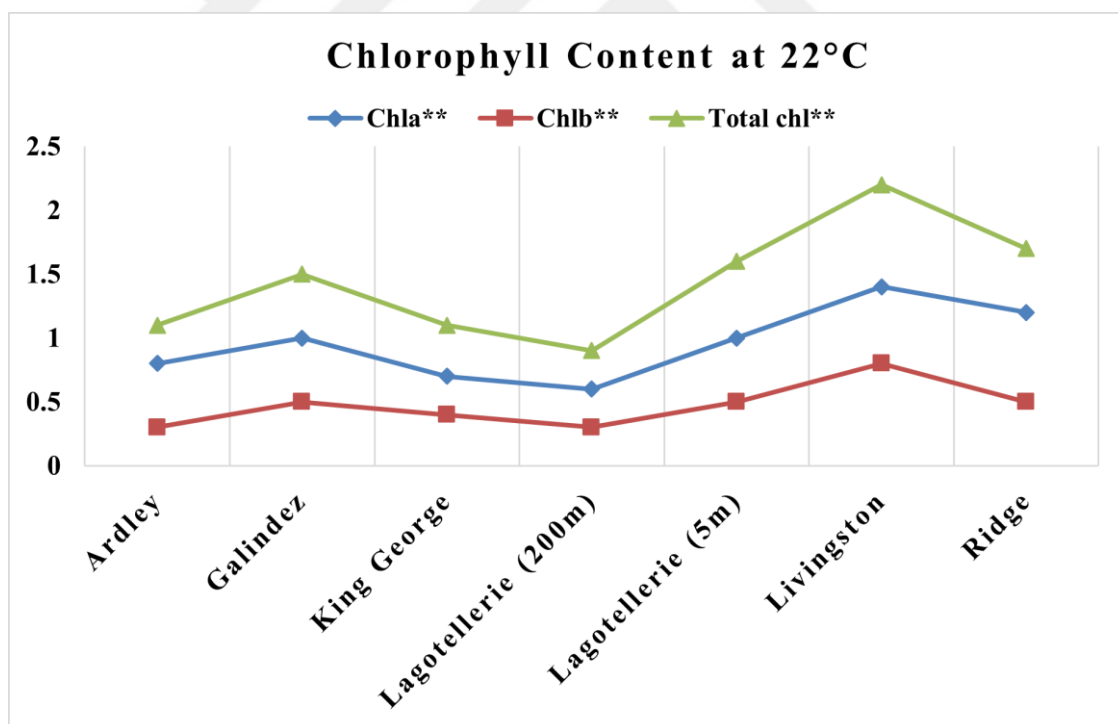


Figure 4. 33. Chlorophyll content of *D. antarctica* ecotypes grown under 22°C

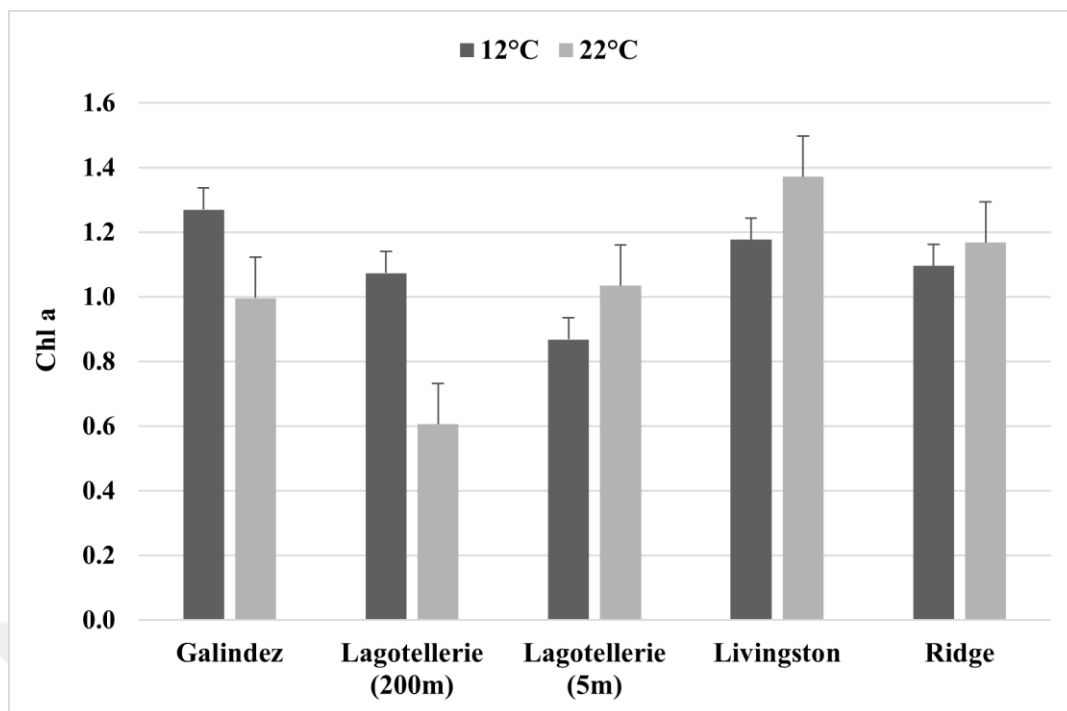


Figure 4. 34. Comparison of the chlorophyll a content of *D. antarctica* ecotypes grown under two different temperature condition

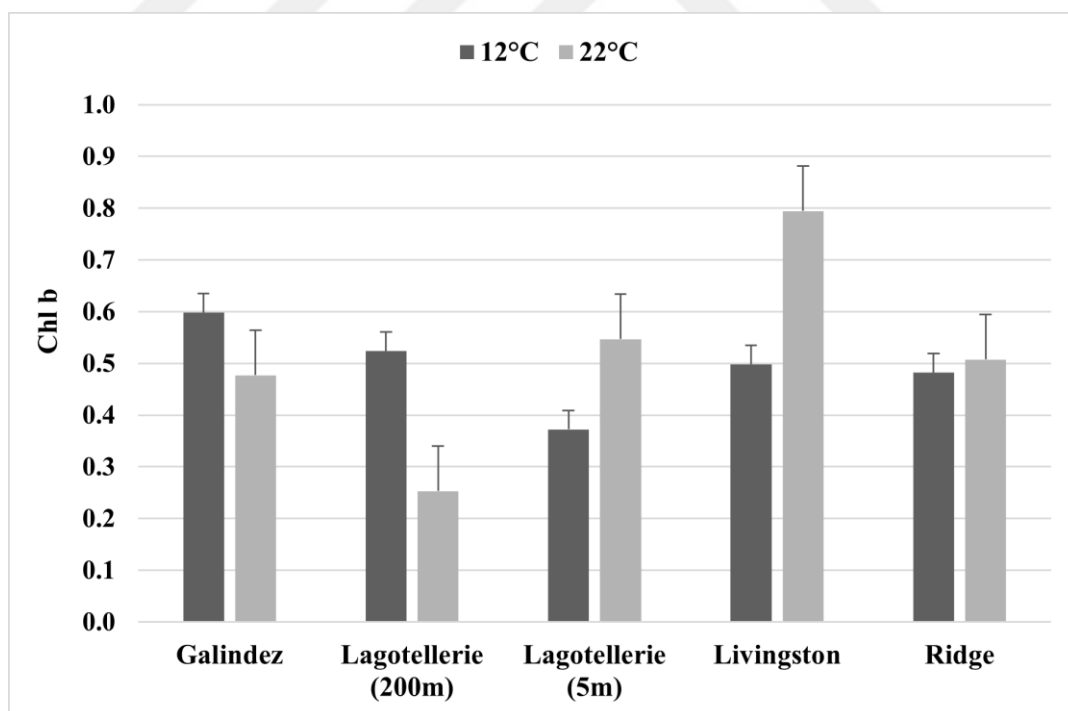


Figure 4. 35. Comparison of the chlorophyll b content of *D. antarctica* ecotypes grown under two different temperature condition

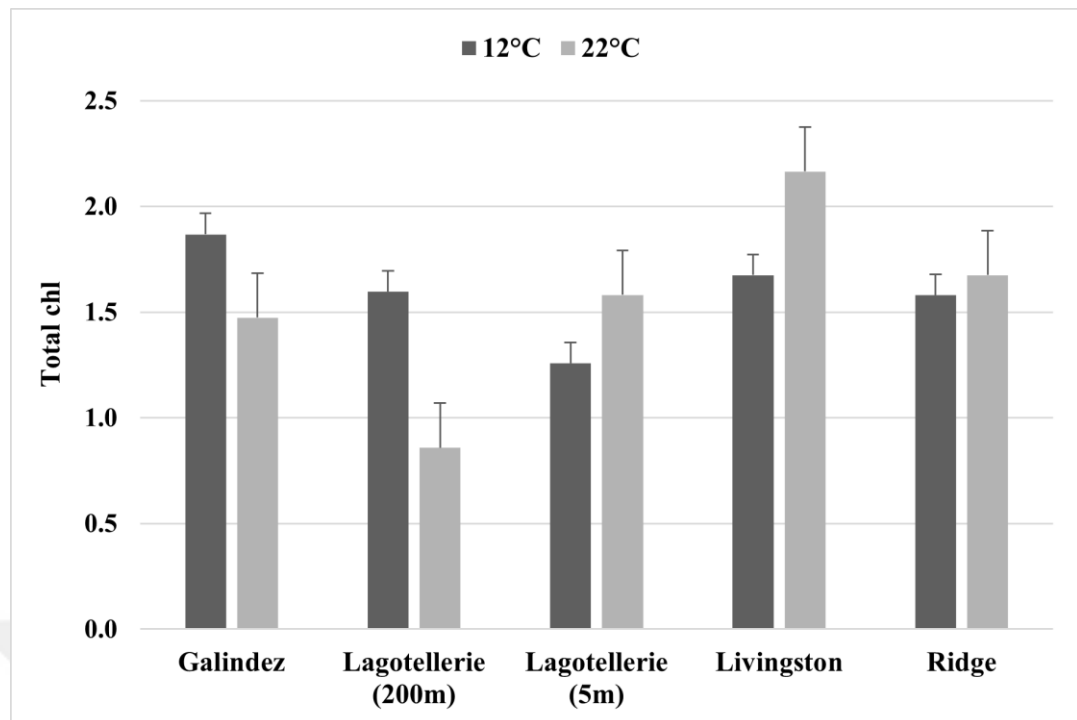


Figure 4. 36. Comparison of the total chlorophyll content of *D. antarctica* ecotypes grown under two different temperature condition

CHAPTER V

CONCLUSIONS

The study on the comparing the development and physiology of *Deschampsia antarctica* ecotype collections grown under two different temperature conditions, 12°C and 22°C, revealed differential responses to these temperature variations during a 6-month growth period. The Livingston ecotype consistently stood out as one of the tallest ecotypes, showcasing its robust growth potential under both 12°C and 22°C temperature conditions. Notably, ecotypes such as Galindez, King George and Ridge showed a significant height increase at 22°C compared to 12°C, indicating a temperature-dependent growth pattern. On the other hand, Lagotellerie (200 m), demonstrates a unique response by displaying a decrease in plant height at 22°C temperature condition. This suggests potential temperature sensitivity in its growth, which differs from the pattern seen in other ecotypes.

Among these ecotypes, Ridge stood out for its remarkable adaptability, as shown by its impressive performance in terms of stem and root characteristics at both temperature levels. Its ability to flower even under stressful conditions at 22°C, further highlights its resilience.

Moreover, the study highlights the impact of temperature on reproductive yield, with ecotypes grown at 22°C producing more siblings than those at 12°C, this notices the importance of considering temperature variations when studying the reproductivity of *D. antarctica*.

The findings from this study may offer valuable insights into how these ecotypes respond to different conditions, as observing the health and behavior of *D. antarctica* can provide crucial information about climate change in Antarctica. Any significant changes in growth patterns, reproductive success, or distribution of this species could potentially indicate environmental shifts caused by climate change. Our study stands out due to its unique focus on the investigation of seven ecotypes collected from two distinct regions in Antarctica. Unlike many other studies that concentrate on specific species, *D. antarctica* from one side, our research takes a broader approach by

examining seven different ecotypes of *D. antarctica*. This comprehensive analysis allows for a deeper understanding of how different ecotypes from various regions respond to temperature variations. By adopting this diverse approach, we are able to gain valuable insights into the potential responses of Antarctic vascular plants to changing environmental conditions across different geographical locations within Antarctica.



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