EVALUATION OF HEAT TOLERANT CAULIFLOWER VARIETIES ON CALIFORNIA’S CENTRAL COAST

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ABSTRACT

Evaluation of Heat Tolerant Cauliflower Varieties on California’s Central Coast

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Heat tolerant trials of vegetable crops will help to improve food security when it becomes affected by rising temperatures due to climate change. By having heat tolerant vegetable crops, we can ensure the well-being of individuals in our society—nutritionally, economically, and socially. California is responsible for 90% of the cauliflower production in the United States. This research aims to determine the overall productivity of three heat tolerant cauliflower varieties (Bishop, Mardi, Flame Star) during the summer months (July-September) on California’s central coast. Stomatal conductance and chlorophyll fluorescence were measured throughout the growing cycle to evaluate plant stress and photosynthetic rate; the results were similar for each variety but had a large variation among the data collected from each plant. Ecological data, such as soil temperature, herbivory, height and soil moisture, were similar for each cauliflower variety throughout the growing season as well. The Bishop and Flame Star varieties produced similar yields of 27.34 pounds (Bishop) and 33.53 pounds (Flame Star), although the Mardi variety did not produce any marketable curds. The three varieties performed similarly, in terms of photosynthetic productivity and ecological data, which indicates that there are alternate dynamics related to biotic or abiotic stress that need to be evaluated in order to determine why the Bishop and Flame Star varieties yielded quality, marketable cauliflower curds relative to the Mardi variety.

Keywords: heat tolerance, cauliflower, climate change, heat stress, crop productivity
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TABLE OF CONTENTS

List of Tables ................................................................................................................................. 7
List of Figures ................................................................................................................................. 8
1 Chapter 1 ........................................................................................................................................ 10
  1.1 Introduction ............................................................................................................................... 10
  1.2 Climate Change and Agricultural Demand .............................................................................. 11
2 Chapter 2: Literature Review ..................................................................................................... 13
  2.1 Introduction ............................................................................................................................... 13
  2.2 Climate Change and the Agricultural Industry ........................................................................ 13
    2.2.1 Vegetable Industry.............................................................................................................. 14
    2.2.2 Cauliflower ....................................................................................................................... 15
  2.3 Vegetable Production and Environmental Stress .................................................................... 15
    2.3.1 Abiotic Stresses ................................................................................................................ 15
    2.3.2 Photosynthetic Rate ......................................................................................................... 16
3 Chapter 3: Materials and Methods .......................................................................................... 19
  3.1 Introduction ............................................................................................................................... 19
    3.1.1 Study Site .......................................................................................................................... 19
    3.1.2 Cauliflower Varieties ....................................................................................................... 20
    3.1.3 Experimental Design ....................................................................................................... 20
    3.1.4 Measurements and Observations .................................................................................... 22
    3.1.5 Insecticide Treatment ...................................................................................................... 22
    3.1.6 Statistical Analysis .......................................................................................................... 23
4 Chapter 4: Results ...................................................................................................................... 24
  4.1 Weather ..................................................................................................................................... 24
  4.2 Height and Herbivory ............................................................................................................... 24
  4.3 Soil Moisture and Soil Temperature ....................................................................................... 26
  4.4 Chlorophyll Fluorescence and Stomatal Conductance ............................................................. 27
  4.5 Yield ......................................................................................................................................... 30
  4.6 Root Morphology ..................................................................................................................... 31
5 Chapter 5: Discussion ................................................................................................................ 33
  5.1 Future Research Considerations ............................................................................................ 35
References....................................................................................................................................... 37
**LIST OF TABLES**

Table 4-1: The average measurements for stomatal conductance and chlorophyll fluorescence of each variety (Bishop, Mardi, Flame Star) from all dates of data collection................................................................................................................. 29

Table 4-2: The total root weight (grams) of each variety—Bishop, Mardi and Flame Star. .................................................................................................................................................................. 32
LIST OF FIGURES

Figure 3-1: Aerial view of the Student Experimental Farm (SEF); the raised planter boxes are outlined in the black box. Pictured on the left is the satellite view of the SEF and pictured on the right is the satellite view with elevation. (https://www.dronedeploy.com/app2/data/60a31c7391460cb257b4a1c8/layers/elevation) .......................................................... 20

Figure 3-2: Layout of the experimental area and the corresponding garden beds for each variety. Planter beds were randomly selected for each variety—Bishop (green), Mardi (blue) and Flame Star (orange). .......................................................... 21

Figure 4-1: Daily range of temperature and precipitation in July, August and September 2021. The maximum temperature occurred during 1-3PM PST. There was no precipitation recorded. .................................................................................. 24

Figure 4-2: Average height (centimeters) measurements for each cauliflower variety (Bishop, Mardi, Flame Star) from two different collection dates. .................. 25

Figure 4-3: Average herbivory (%) measurements for each cauliflower variety (Bishop, Mardi, Flame Star) from two different collection dates. .................. 26

Figure 4-4: Average soil moisture, represented by volumetric water content (%VWC), for each cauliflower variety—Bishop, Mardi and Flame Star. Soil moisture data was collected over a seven-week period with a TDR probe. .......................................................... 27

Figure 4-5: Average soil temperature (˚C) for each cauliflower variety—Bishop, Mardi and Flame Star. Soil temperature data was collected over a seven-week period with a TDR probe. ........................................................................ 27

Figure 4-6: Chlorophyll fluorescence measurements collected at six different dates of collection for each cauliflower variety. The black points represent each plant of the designated variety—Bishop, Mardi and Flame Star. The line illustrates the fluctuations of the average chlorophyll fluorescence at that time point. .......... 28

Figure 4-7: Stomatal conductance measurements collected at six different dates of collection for each cauliflower variety. The black points represent each plant of
the designated variety—Bishop, Mardi and Flame Star. The line illustrates the fluctuations of the average stomatal conductance at that time point. .......................... 29

Figure 4-8: The total yield for the Bishop and Flame Star varieties. Curds were of standard size (5-7inches.) and did not have a ‘ricey’ appearance. Mardi did not produce any marketable cauliflower heads.......................................................... 30

Figure 4-9: Mardi cauliflower curds that are less than standard quality. A “ricey” appearance can be clearly seen in the left cauliflower curd; red arrows highlight the ricey appearance on the curds. The middle cauliflower curd, circled in red, is smaller (less than 4 inches) than the standard size (5-7 inches). .......................... 30

Figure 4-10: Flame Star (right) and Bishop (left) harvested cauliflower curds. The cauliflower heads are standard size (5-7 inches) with no ricey/loose curds. ...... 31

Figure 4-11: Average root ball width (cm) and root length (cm) for each variety—Bishop, Mardi and Flame Star.......................................................... 31
1 CHAPTER 1

1.1 Introduction

The California agricultural industry is responsible for supplying over a third of the country’s vegetables and two thirds of the country’s fruits and nuts. In 2018, the value of vegetable crop production in California was $12.9 billion (USDA, 2018). Vegetables provide nutritional balance to one’s diet and can help with overcoming micronutrient deficiencies (Ayyogari et al., 2014; Bhardwaj, 2012; Peña & Hughes, 2007). Vegetables are sensitive to environmental extremes, such as high temperatures and low soil moisture (Ayyogari et al., 2014; Kochler et al., 2007a). These environmental stresses, projected to accelerate due to climate change, threaten lower yields and increased production costs. In order to combat this, vegetable crops will need to overcome heat/drought stress while maintaining yield (Basu et al., 2016). Overall, the goal of this research is to evaluate heat tolerant vegetable cultivars that can be utilized in the agricultural industry to maintain food security.

Cauliflower is a vegetable crop in the Brassica family that has a high socio-economic, commercial, and nutritional value—it is also known to be one of the most important agricultural vegetable crops worldwide (J. E. Olesen & Grevesen, 1993; Rurek, 2010). The consumption of cauliflower has increased over the past decade due to its high nutritional value and beneficial effects on human health (Collado-González et al., 2021). California is the largest producer of cauliflower with approximately 90% of the supply in the United States. Last year (2020), the total crop value was $346 million, total production equaling 9.0 million hundredweight (cwt) (“CA Utilized Vegetable Production Value Shows Slight Decline,” 2021). Globally, the United States is third in the world’s top producers of cauliflower; China is the number one producer and India is second (S & Singh, 2020). Cauliflower is grown in the central and south coast of California; in the counties of Monterey, San Benito, Santa Cruz, San Luis Obispo, Ventura, Santa Barbara, Tulare, Fresno, Stanislaus and San Joaquin (Koike et al., 2009). Cauliflower requires adequate soil moisture throughout the growing cycle, so the crop is typically irrigated with furrow and overhead sprinklers (Koike et al., 2009). Since cauliflower is such a high value vegetable, it is important to evaluate how abiotic factors,
such as temperature, can affect cauliflower productivity (J. E. Olesen & Grevsen, 1993; Rurek, 2010; Torres et al., 2017).

A decrease in a plant’s photosynthetic rate is indicative of plants experiencing resource or environmental related stress—which will ultimately affect overall plant productivity (Gou et al., 2018; Maxwell & Johnson, 2000). Indicators for determining photosynthetic rate are stomatal conductance and chlorophyll fluorescence (Cheng et al., 2001; Maxwell & Johnson, 2000; Medrano et al., 2002; Urban et al., 2017). Stomatal conductance is a measurement of the degree to which the stomates are opening to allow gas exchange between water vapor leaving the plant and carbon entering it (Medrano et al., 2001). Chlorophyll fluorescence is a measurement that indicates the efficiency of a plant’s photochemistry—specifically Photosystem II (Maxwell et al., 2000). Traditional cauliflower varieties have been shown to have a net decrease in photosynthetic rate when exposed to heat stress (Kochler et al., 2007a; J. E. Olesen & Grevsen, 1993; Rurek, 2010). Dry soil has also been shown to increase stress symptoms in traditional cauliflower varieties (Kochler et al., 2007a). By evaluating these indicators, we will be able to determine if heat tolerant cauliflower varieties are undergoing environmental stress and how it correlates to overall yield.

1.2 Climate Change and Agricultural Demand

Climate change has led to increased surface temperature, erratic precipitation patterns, increased intensity and duration of drought; these climatic changes will adversely affect food production on a regional and global scale (Bhardwaj, 2012; IPCC — Intergovernmental Panel on Climate Change, n.d.; Leisner, 2020; Moretti et al., 2010). Agriculture production is responsible for feeding the world’s growing population—as the population increases, agricultural demand is expected to increase 50% by 2030 (Wheeler et al., 2000). Climate change will have an impact on crop productivity, ultimately affecting food availability in the face of high demand (Peña & Hughes, 2007; Wheeler et al., 2000; Wojciechowska et al., 2016). The agricultural industry needs to identify alternative, heat tolerant crop varieties so that we can maintain food security without requiring additional nutrient and water-resource input (Ayyogari et al., 2014; Blum, 2009; Urban et al., 2017).
There is a need for research into the physiological and ecological mechanisms involved with heat stress and how plant breeding selection can mitigate the effect of climate change on vegetable production (El-Soda et al., 2014; Kage et al., 2001; J. E. Olesen & Grevsen, 1993; Rurek, 2010). Heat stress has the capability to alter a plant’s gene expression, metabolism, growth rate and crop yield (Aleem et al., 2021; Bray et al., 2000). Evaluation of heat stress in heat tolerant vegetable cultivars will help us to understand the mechanisms involved in maintaining plant productivity during prolonged periods of environmental stress (Bray et al., 2000). Alternate genotypes of the same plant species will have the ability to survive differing amounts of stress; heat tolerant trials of a single vegetable will demonstrate this fact (Bray et al., 2000; Rurek, 2010).

Characterizing heat resistant vegetable cultivars will have positive effects on all three pillars of sustainability—environmentally (having improved resource utilization), economically (stable vegetable production, maintaining food prices) and socially (increasing nutritional well-being, decreasing inequality by lowering food production costs).

The objectives of this research are to investigate (1) differences in plant stress (stomatal conductance) and photosynthetic rates (chlorophyll fluorescence) between three cauliflower varieties (2) the relationship between plant stress, photosynthetic rate, and yield. We hypothesize that: (1) the cauliflower variety that has an increased photosynthetic rate will be more heat tolerant compared to alternate varieties and (2) the most heat tolerant cauliflower variety will produce the greatest yield despite higher temperatures during the growing/harvest season.
2 CHAPTER 2: LITERATURE REVIEW

2.1 Introduction
Agriculture production is responsible for feeding the world’s growing population, which will ultimately lead to an increased demand for food production (Wheeler et al., 2000). Climate change negatively affects crop productivity, which will then adversely affect food availability as global demand grows (Peña & Hughes, 2007; Wheeler et al., 2000; Wojciechowska et al., 2016). Vegetables, in particular, improve dietary health and nutrition (Ebert, 2017). The agricultural industry needs to look into alternative, heat tolerant crop varieties so that we can maintain food security without requiring additional nutrient and water-resource input (Ayyogari et al., 2014; Blum, 2009; Urban et al., 2017). This review will discuss the effects of climate change on California’s vegetable industry; how vegetables are affected by environmental stresses; and the methods in which we can evaluate those factors.

2.2 Climate Change and the Agricultural Industry
Climate change poses a great threat to global food production—environmental extremes associated with climate change will have negative effects on the production of our agricultural commodities. (Bhardwaj, 2012; IPCC — Intergovernmental Panel on Climate Change, n.d.; Leisner, 2020; Moretti et al., 2010). The global population in 2050 is projected to consist of 9 billion people (the population in 2021 is currently 7.9 billion people), which is why we need to find ways to maintain food security and lower our environmental impact in the face of increased demand (Yadav et al., 2015). Research is increasingly focused on mitigation and adaptation of alternate crops and farming systems in order to develop what is known as “climate smart food systems” (Fujimori et al., 2018; Yadav et al., 2015). These “climate smart” food systems believe that sustainably transforming the agricultural sector is key to maintaining food security. Key features of creating these sustainable agricultural food systems are (1) to increase food security by increasing crop productivity and (2) to build resilience within our crops so that it allows them adapt to climate change (Branca et al., 2013)
Global surface temperature is increasing; it is expected to increase, on average, between 0.8°C and 1.0°C by mid-century (2040-2050) and by end of century it is expected to increase between 2°C and 4°C \( (IPCC — Intergovernmental Panel on Climate Change, \text{n.d.; Leisner, 2020; Wheeler et al., 2000; Yadav et al., 2015}) \). The California counties that are responsible for the majority of cauliflower production are expected to have a 7-8°F increase of the average annual maximum temperature by the end of the 21st century \( (“Central Coast Region Report,” \text{n.d.}) \). An increase of the average annual temperature in these regions pose a great risk to some of our largest agricultural production sectors in California. California agricultural plays a large role in maintaining food security on a national level; which is why it is imperative for research to consider how to mitigate the effect of climate change on agricultural production \( (USDA - National Agricultural Statistics Service - About NASS - Agency Overview, \text{n.d.}) \).

### 2.2.1 Vegetable Industry

Increased surface temperature has the ability to adversely affect the future production value of California’s vegetable industry. Vegetables are sensitive to environmental extremes, such as high temperatures and low soil moisture \( (Ayyogari et al., 2014; Kochler et al., 2007a) \). Environmental stresses attributed to climate change, such as increasing temperatures, reduced irrigation, flooding, and salinity, have been shown to limit vegetable production productivity and sustainability—these factors can decrease crop productivity by 65 to 87% \( (Bray et al., 2000; Rao et al., 2016) \). High temperatures and increased \( CO_2 \) levels have also been shown to negatively affect annual yield and overall product quality. Yield losses attributed to temperature stress will negatively affect agricultural productivity and will be detrimental to the quality of the vegetable supply \( (Peña & Hughes, 2007) \). Irrigation is a commonly utilized practice in agriculture to alleviate heat stress since it maximizes the amount of water that reaches the plant \( (Nouri et al., 2019) \). In addition to adapting our agricultural practices, modern breeding tools can be utilized to explore and develop new cultivars adapted to elevated temperatures \( (Peña & Hughes, 2007) \). There needs to be increased research to mitigate the effects of climate change on some of California’s most prominent vegetable crops, such as, cauliflower.
2.2.2 Cauliflower

Cauliflower is a high value vegetable crop that is at risk of decreased production outputs. Cauliflower (*brassica oleracea var. botrytis*) is typically bred to be a cool season crop and is sensitive to extreme temperature changes. Cauliflower production and overall consumption has increased over the past decade due to its high commercial and nutritional value (Collado-González et al., 2021; J. E. Olesen & Greven, 1993; Rurek, 2010). Temperature plays an important role on its vegetative and generative phases during production (Ara et al., 2009). Research on cauliflower production in California has been associated with the crop’s susceptibility to verticillium wilt disease rather than its response to environmental extremes (Ebert, 2017; Jensen et al., 1999; Koike et al., 1996). Countries, such as China, Pakistan, Europe and India, have studied aspects of the biochemical processes associated with heat tolerance in cauliflower varieties, although research is limited (Aleem et al., 2021; Kenny et al., 1993; Kochler et al., 2007a; J. Olesen & Greven, 1997). Climate projections vary on the global scale, which is why crop productivity research should be conducted at the local environment. California is responsible for the majority (90%) of the cauliflower production in the United States, which is why it is important to research how abiotic factors, such as temperature, can affect cauliflower productivity in the future (J. E. Olesen & Greven, 1993; Rurek, 2010; Torres et al., 2017).

2.3 Vegetable Production and Environmental Stress

2.3.1 Abiotic Stresses

Abiotic stresses, or environmental related stress, can affect morphological and physiological responses within a plant. These abiotic stresses have the ability to affect the root, shoot, and leaf structure along with the rate of photosynthesis, respiration and transpiration (Kochler et al., 2007b; Rao et al., 2016; Rurek, 2010; Tardieu, 2013). These abiotic stresses have the power to significantly affect plant growth and overall productivity, which would lead to an overall decrease in yield. Abiotic stress causes plants to go into an “alarm phase,” which leads to a decline in function in order for the plant to survive (Wojciechowska et al., 2016). High temperatures are one of the most important abiotic factors that can affect the cultivation of many plant species (Collado-González et al., 2021). It is imperative that we evaluate the many mechanisms involved
with the heat stress response to understand how we can make our plants more resilient in the face of increased environmental stressors.

2.3.1.1 Response to Heat Stress

Plants have developed heat tolerant mechanisms—biochemical, molecular and morphophysiological—to modify their response to heat stress (Bray et al., 2000; Formisano et al., 2020). Common avoidance and acclimation mechanisms in response to heat stress can include altering membrane phospholipids, changing leaf orientation, reducing leaf size and reducing water loss by controlling stomates (Formisano et al., 2020). Heat tolerant grasses have been shown to have an extensive root system to facilitate water intake and increase transpirational cooling in response to higher temperatures (Xu & Huang, 2001). Alternate studies of heat tolerant crops demonstrate that there is a decrease in normal protein synthesis and an acceleration in the transcription and translation of Heat Shock Proteins (HSPs) in response to heat stress (Bray et al., 2000). The HSPs are responsible for stabilizing integral proteins and cell membranes during heat stress to prevent plant death (Aleem et al., 2020). The responses to heat stress vary among plant genotypes; it is important to evaluate these adaptations across different genotypes if we are going to produce a superior heat tolerant plant cultivar.

2.3.2 Photosynthetic Rate

A suite of photosynthetic traits can be used to assess plant stress (Kochler et al., 2007b; Medrano et al., 2002; Rao et al., 2016; Urban et al., 2017). Reduced photosynthetic rate is indicative of plants experiencing resource or environmental related stress—which will ultimately affect overall plant productivity (Gou et al., 2018; Maxwell & Johnson, 2000). Indicators for determining photosynthetic rate are stomatal conductance and chlorophyll fluorescence; net photosynthesis can also be measured by evaluating CO₂ uptake (Cheng et al., 2001; Maxwell & Johnson, 2000; Medrano et al., 2002; Urban et al., 2017).

2.3.2.1 Stomatal Conductance

Stomatal conductance is a measurement of the degree to which the stomates are opening to allow gas exchange between water vapor leaving the plant and carbon entering it (Medrano et al., 2001). Stomatal conductance to water (GSW) is a measure of the degree of stomatal openness and the number of stomata. If GSW is (0), then the stomata are
closed; if the GSW is (.99), then the stomata are open. Stomatal openings regulate the exchange of water vapor and CO₂ between a leaf and the surrounding air (Medrano et al., 2001). Early closure of stomata reduces photosynthetic efficiency, thus hindering dry matter production (Liu & Stützel, 2002). Stomatal openness can indicate a physiological response to environmental conditions; as plants become more stressed, it is expected for more stomates to close. Evaluation of stomatal conductance will indicate the plant’s physiological response while the evaluation of chlorophyll fluorescence will provide information about a plant’s quantum efficiency.

2.3.2.2 Chlorophyll Fluorescence
Chlorophyll fluorescence is a measurement that indicates the efficiency of a plant’s photochemistry—specifically Photosystem II (PSII) (Maxwell et al., 2000). Light energy absorbed by a chlorophyll molecule can be used in three different processes: photosynthesis, heat emittance or re-emittance of light (fluorescence) (Maxwell & Johnson, 2000; Murchie & Lawson, 2013). The competition between these processes allows us to evaluate the overall efficiency of PSII.

PSII is a measure of the efficiency of photon emission as defined by the ratio of the number of photons emitted to the number of photos absorbed (Murchie & Lawson, 2013). As the percentage of PSII increases, it indicates that the light absorbed is being utilized for photochemistry (Maxwell et al., 2000). By evaluating the photochemistry mechanisms of a plant, we can compare the overall efficiency of alternate plant species and their response to environmental stresses.

Further research is needed in order to mitigate the effect of climate change on California’s vegetable industry—more specifically, the production of cauliflower. Vegetables are highly sensitive to extreme abiotic factors, which makes their overall productivity and production value very susceptible to decline in the future. It is imperative to evaluate the physiological mechanisms taking place in alternate plant varieties when exposed to increased temperatures. In doing so, researchers can determine
which vegetable cultivars will maintain their productivity in the face of extreme climatic changes/events.
3 CHAPTER 3: MATERIALS AND METHODS

3.1 Introduction

This research will be used to determine the overall efficiency of heat tolerant cauliflower varieties at the cellular and ecological level. To determine the photosynthetic rate, stomatal conductance and chlorophyll fluorescence will be evaluated. The evaluation of stomatal conductance and chlorophyll fluorescence will allow us to determine if the plants are experiencing stress due to abiotic factors, such as an increase in temperature (Maxwell & Johnson, 2000; Medrano et al., 2002). Furthermore, this research will also evaluate soil components, height, herbivory, root morphology and yield of each cauliflower variety.

3.1.1 Study Site

The site is located at California Polytechnic State University’s Student Experimental Farm (SEF) in San Luis Obispo, CA (35.31052392244775 N, -120.6730471601539 W) (Fig. 1). The cauliflower varietals were evaluated/grown in above ground (raised) planter boxes that are 8’ by 4’. The planter boxes contained sandy loam soil; this soil is comprised of 40% compost, 40% sand and 20% coconut core. Bone and alfalfa meal mix was incorporated into the soil of the garden beds—consisting of 4% Water Soluble Nitrogen, 6% Available Phosphate, 3% Soluble Potash and 7.5% Calcium. The climate of San Luis Obispo, CA is classified as a warm-summer Mediterranean climate (Köppen-Geiger Climate Classification).
3.1.2 Cauliflower Varieties

Three cauliflower (*brassica oleracea var. botrytis*) varieties were chosen for this experiment: Bishop, Flame Star and Mardi. The Mardi and Bishop are white cauliflower varieties; the Flame Star is an orange cauliflower variety. Mardi and Flame star have a 62 day growing period and Bishop has a 65 day growing period (*Johnny’s Selected Seeds*, 2021). These specific varietals were bred to be “very good” for heat tolerance and can be transplanted in the spring/summer and harvested in summer or fall (*Johnny’s Selected Seeds*, 2021). The cauliflower transplants were planted 18 inches apart and each row was planted 24 inches apart. The raised planter boxes will have 10 plants per bed (approximately two rows with five plants each).

3.1.3 Experimental Design

Prior to planting, seeds were germinated under LED lights for four weeks until they were ready for transplant at the site location (SEF). This experiment was conducted in a randomized complete block design that was replicated four times for each variety (Kage et al., 2001; Torres et al., 2017).
Figure 3-2: Layout of the experimental area and the corresponding garden beds for each variety. Planter beds were randomly selected for each variety—Bishop (green), Mardi (blue) and Flame Star (orange).

The planter beds were watered with a drip irrigation system three times a week for forty minutes each session. After the initial transplant, spot irrigation was utilized for the first two weeks in conjunction to the regular schedule.

Daily maximum and minimum temperatures (F’) were recorded each day along with total precipitation (inches) (Jensen et al., 1999; Kage et al., 2001; Torres et al., 2017). Soil moisture (% VWC) and soil temperature (˚C) were measured at 40cm, 80cm, 120cm, 160cm and 200cm (Koike et al., 2009). Stomatal conductance and chlorophyll fluorescence were measured using a LI-600 porometer/fluorometer (LICOR).
3.1.4 Measurements and Observations

Data measurements and observations took place over a ten-week period during the summer (July through September, 2021). LICOR and TDR data were collected once a week during the 1 PM - 3 PM PST time frame, when temperature is typically at its highest in San Luis Obispo, CA during the summer.

Stomatal conductance and chlorophyll fluorescence were measured using the LI-600 Porometer and Fluorometer on leaves of each cauliflower varietal. The LI-600 instrument was clamped onto the newest fully expanded leaf. A TDR (time-domain reflectometry) sensor was used to determine the percentage of soil volumetric water content (VWC) and soil temperature at different soil transect points in the garden bed (Gaiser et al., 2004).

Height and herbivory were measured twice during the growing season for every cauliflower plant; the measurements were taken during week 5 and week 9. The height was measured in centimeters using a standard ruler. The observed herbivory was recorded from randomly selected leaves from each cauliflower plant.

The mature cauliflower curds were harvested and weighed to determine the overall yield of each cauliflower variety. All plant material above the soil was harvested (Parsons et al., 2007). The curds harvested were measured in diameter and assessed for market quality—assessments will be based on the U.S. No. 1 and No. 2 grades of cauliflower (Cauliflower Grades and Standards | Agricultural Marketing Service, n.d.).

The roots were carefully removed from the soil by digging a trowel in a circular motion about 5 inches from plant base. The roots were lightly shaken to detach loose dirt; the roots width and length were measured using a standard ruler. Roots were then rinsed with water and left to dry for five minutes. The roots were then weighed in grams on a scale.

3.1.5 Insecticide Treatment

The plants developed aphid colonies on their leaves during the growing season, increasing the rate of herbivory. The cauliflower plants were treated with an organic insecticide to minimize damage and overall stress to the plants. The organic insecticide
was comprised of 1 tablespoon of Dr. Bronner’s Pure Castille Peppermint Liquid Soap and 60 ounces of warm water. The organic insecticide was used once during week 5 to control the aphid population; it was applied to all cauliflower plants that had aphids present.

3.1.6 Statistical Analysis

Data was analyzed using a mixed model in JMP Pro to determine statistical significance between the varieties for height, herbivory, chlorophyll fluorescence, and stomatal conductance over different time points. The mixed model has a random effect for plot, which controls for the correlation of plants within the same plot. Data collected for root morphology and yield was analyzed with an ANOVA in JMP Pro (Collado-González et al., 2021). Weather data, TDR data and LI-600 data will be stored, recorded in an Excel document, and archived in OneDrive.
4 CHAPTER 4: RESULTS

4.1 Weather

Daily temperature and precipitation values were obtained from Cal Poly’s ITRC Weather Station. Weather data was obtained for the months of July, August, and September (Figure 4-1). This data represents the weather for the duration of the experiment (from planting the transplants on 07/01/2021 to pulling out the roots on 09/06/2021). Maximum daily temperature was reached during the 1PM to 3PM PST time period. Data collection occurred during this time period. There was no precipitation recorded during these months (Figure 4-1).

![Weather Data (July, August, September 2021)](image)

*Figure 4-1: Daily range of temperature and precipitation in July, August and September 2021. The maximum temperature occurred during 1-3PM PST. There was no precipitation recorded.*

4.2 Height and Herbivory

The average heights of the cauliflower plants were similar between the three varieties. Over the course of the experiment, the Flame Star variety was consistently taller than the Bishop and Mardi varieties (Figure 4-2). The initial mean height measurement of the Bishop variety at 35.77 centimeters (the shortest) did not affect its final average growth
height of 56.3cm; the second tallest of the three varieties. All of cauliflower varieties increased in height (by at least 10 centimeters) over time; the Bishop variety had the greatest increase in height (~20 cm). There was not a significant difference in height between the Bishop (M= 56.3, SD= 12.198), Mardi (M= 55.15, SD= 7.468) and Flame Star (M= 57.35, SD= 6.830) varieties; 0.01 (2); p = 0.93.

![Average Height](image)

Figure 4.2: Average height (centimeters) measurements for each cauliflower variety (Bishop, Mardi, Flame Star) from two different collection dates.

As the cauliflower plants developed, there was an increased amount of herbivory (Figure 4-3). The Mardi had the highest percent of herbivory (38.93%) on the second date of data collection (08/31); the Flame Star had the lowest rate of herbivory (29.88%) on the second data collection date. There was not a significant difference in herbivory between the Bishop (M= 36.55, SD= 28.442), Mardi (M= 38.925, SD= 26.589) and Flame Star (M= 29.875, SD= 22.186) varieties; 0.55 (2); p=0.1736.
4.3 Soil Moisture and Soil Temperature

The soil temperature and moisture decreased gradually over the growing season (Figure 4-4; Figure 4-5). The soil temperature and moisture were similar across each variety’s plot. The Bishop variety maintained an increased soil moisture value over time, except for August 26th when it was equal to the Flame Star’s average soil moisture (16.045%). The Mardi’s average soil moisture was lowest (10.16%) on the fifth day of collection (08/18).

Figure 4-3: Average herbivory (%) measurements for each cauliflower variety (Bishop, Mardi, Flame Star) from two different collection dates.
4.4 Chlorophyll Fluorescence and Stomatal Conductance

Each cauliflower variety had similar chlorophyll fluorescence and stomatal conductance measurements throughout the course of the experiment. Every plant from each variety...
was evaluated at six different data collection dates. The variation of the chlorophyll fluorescence measurements over time were similar across each variety (Figure 4-6).

![Figure 4-6: Chlorophyll fluorescence measurements collected at six different dates of collection for each cauliflower variety. The black points represent each plant of the designated variety—Bishop, Mardi and Flame Star. The line illustrates the fluctuations of the average chlorophyll fluorescence at that time point.](image)

There are 120 data points representing the 40 plants for the Bishop, Mardi and Flame Star varieties. Bishop had the highest total average chlorophyll fluorescence of 0.692; the Mardi and Flame Star were very similar to the Bishop with 0.68 being the average chlorophyll fluorescence measurement (Table 4-1). The distribution of the data points (representative of every plant) of each variety are also similar. After the third date of collection, the average chlorophyll fluorescence increased till the last date of collection. There was not a significant difference in chlorophyll fluorescence between the Bishop (M= 0.692, SD= 0.074), Mardi (M= 0.680, SD= 0.078) and Flame Star (M= 0.680, SD= 0.085) varieties; 0.03 (2); p = 0.2690.
Table 4-1: The average measurements for stomatal conductance and chlorophyll fluorescence of each variety (Bishop, Mardi, Flame Star) from all dates of data collection.

<table>
<thead>
<tr>
<th>Variety</th>
<th>Average Stomatal Conductance</th>
<th>Average Chlorophyll Fluorescence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bishop</td>
<td>0.379</td>
<td>0.692</td>
</tr>
<tr>
<td>Mardi</td>
<td>0.402</td>
<td>0.680</td>
</tr>
<tr>
<td>Flame Star</td>
<td>0.453</td>
<td>0.680</td>
</tr>
</tbody>
</table>

The Flame Star variety had the highest average stomatal conductance of 0.453. The Mardi had the next highest average stomatal conductance of 0.402, while the Bishop’s average was 0.379. The Mardi’s stomatal conductance has the largest range of fluctuation over time (Figure 4-7). The distribution of the data points is consistent across the three varieties. There was not a significant difference in stomatal conductance between the Bishop (M= 0.379, SD= 0.154), Mardi (M= 0.402, SD= 0.141) and Flame Star (M= 0.453, SD= 0.15) varieties; -1.78 (2);p = 0.2189.

![Stomatal Conductance vs. Time](image)

Figure 4-7: Stomatal conductance measurements collected at six different dates of collection for each cauliflower variety. The black points represent each plant of the designated variety—Bishop, Mardi and Flame Star. The line illustrates the fluctuations of the average stomatal conductance at that time point.
4.5 Yield

The Flame Star variety had the greatest overall year, totaling 33.53 pounds (Figure 4-8). The Bishop variety produced approximately six pounds less than the Flame Star, totaling 27.35 pounds.

![Figure 4-8: The total yield for the Bishop and Flame Star varieties. Curds were of standard size (5-7 inches) and did not have a ‘ricey’ appearance. Mardi did not produce any marketable cauliflower heads.]

The Mardi yield data was excluded due to the cauliflower heads not developing into the standard curd size and presentation. The Mardi variety produced smaller cauliflower heads with ‘ricey’ and loose curd appearance (Figure 4-9). The Bishop and Flame Star varieties produced large, standard (5-7 inches) sized curds (Figure 4-10). The visual appearance of the Mardi production prompted it to be excluded from the total yield data. There was not a significant difference in yield between Bishop and Flame Star varieties; -0.53(1); p = 0.613.

![Figure 4-9: Mardi cauliflower curds that are less than standard quality. A “ricey” appearance can be clearly seen in the left cauliflower curd; red arrows highlight the ricey appearance on the curds. The middle cauliflower curd, circled in red, is smaller (less than 4 inches) than the standard size (5-7 inches).]
4.6 Root Morphology

The Flame Star cauliflower variety had the greatest average root ball width and the heaviest total root weight compared to the Bishop and Mardi varieties. The Bishop variety had the greatest root length, 16.9 centimeters; the Mardi and Flame Star varieties had average root lengths of 15.5 cm and 15.425 cm, respectively (Figure 4-11). There was not a significant difference in root length between the Bishop (M= 16.9, SD= 3.58), Mardi (M= 15.5, SD= 2.16) and Flame Star (M= 15.43, SD= 4.25) varieties; 1.53(2); p = 0.3539. There was not a significant difference in root ball width between Bishop (M= 12.55, SD= 3.27), Mardi (M= 11, SD= 1.96) and Flame Star (M= 14, SD= 3.35) varieties; 0.04 (2); p =0.1372.

![Figure 4-11: Average root ball width (cm) and root length (cm) for each variety—Bishop, Mardi and Flame Star.](image)
The average total root weight was highest with the Flame Star (6114.7 grams) variety compared to the Bishop (5360.8 grams) and Mardi (5865.2 grams) (Table 4-2). Cauliflower varieties with increased root weight had shorter average root lengths.

Table 4-2: The total root weight (grams) of each variety—Bishop, Mardi and Flame Star.

<table>
<thead>
<tr>
<th>Variety</th>
<th>Total Root Weight (grams)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bishop</td>
<td>5360.8</td>
</tr>
<tr>
<td>Mardi</td>
<td>5865.2</td>
</tr>
<tr>
<td>Flame Star</td>
<td>6114.7</td>
</tr>
</tbody>
</table>
This experiment was conducted to investigate (1) differences in plant stress (stomatal conductance) and photosynthetic rates (chlorophyll fluorescence) between three cauliflower varieties (2) the relationship between plant stress, photosynthetic rate, and yield. Developing heat tolerant vegetable cultivars for our most profitable agricultural commodities will aid us in the face of rising temperatures attributed to climate change. The methods used for this experiment seek to understand the ecological and biological responses in heat tolerant plants and how they can be used to predict/assess heat tolerance in the future. The results among the three cauliflower varieties—Bishop, Mardi, Flame Star—indicate that there is no significant difference between them. Height, herbivory, chlorophyll fluorescence, stomatal conductance and root morphology are all very similar among the three varieties. Despite these similarities, there was a significant difference between the Mardi yield compared to the Bishop and Flame Star yield. The Mardi variety produced zero marketable yield; while the Bishop and Flame Star produced 27.34 and 33.53 pounds, respectively.

The mixed model used for the statistical analysis of the results accounted for any random effect that would occur within the same plot. By accounting for this random effect, the results produced no statistical significance between the alternate cauliflower varieties. The consistent pattern seen in the results is that the null hypothesis is true and that there is not a significant difference between the heat tolerant varieties.

Soil temperature and soil moisture were similar for each garden bed, indicating uniform environmental conditions among each variety. Any large fluctuations in the soil data would indicate a potential source of abiotic stress to the plants, although none was observed in this experiment. The soil moisture and soil temperature both decreased gradually over the growing period. This is most likely attributed to the plants maturing—as the cauliflower plants developed and grew larger, the plants required increased water uptake and their larger canopy structure lowered the soil temperature.
Root morphology was also very similar among the varieties; typically root morphology is an indicator of heat tolerance as it allows plants to access more water in times of heat stress. The results of the root characteristics offered no statistical difference between the varieties; nor did it show an above average performance from these varieties. Cauliflower normally has a shallower root system, averaging 12 to 18 inches in length—the Bishop, Mardi and Flame Star variety root length was within this range, averaging 15-16 inches in length.

There was an exponential increase in chlorophyll fluorescence and stomatal conductance during the six data collection dates—occurring between the second and fifth collection dates. Increased stomatal conductance and chlorophyll fluorescence indicate decreased plant stress and increased plant productivity (Liu & Stützel, 2002; Maxwell & Johnson, 2000; Medrano et al., 2002). The crops are possibly increasing their photosynthetic activity to store energy for curd formation. The cauliflower plants started to develop curds on the sixth data collection day, when the stomatal conductance and chlorophyll fluorescence values started to decline.

The Mardi variety is not recommended for heat tolerance. The curds produced a “ricey” appearance and were smaller than the standard size (Figure 4-9). The curd heads did not exceed 4 inches in width, while the Bishop and Flame Star curds were at least 5-7 inches in width. The Mardi yield was excluded since the curds were not considered marketable relative to the standard production requirements. The Mardi was exposed to the same environmental conditions and bred for heat tolerance, although the yield data indicates that this variety is not suitable for higher temperatures.

A ‘ricey appearance’ is indicative of environmental stress (Koike et al., 2009). The environmental conditions were similar for each variety (illustrated by the weather and soil data) and the photosynthetic activity was also very similar, which is why it is difficult to determine what exactly caused stress to the Mardi plants. The Mardi’s stomatal conductance curve is similar to the Bishop and the Flame Star, but it does have greater fluctuation from time 3 to time 5 (Figure 4-7). This fluctuation was not statistically
significant although it could indicate that large fluctuations in stomatal conductance could influence curd development. Additional research involving the measurement of alternate parameters of photosynthesis could potentially indicate another biochemical process that influences the curd formation/quality.

The failure of the Mardi variety to produce marketable curds illustrates the importance of the heat tolerant trials for our vegetable commodities in California. Cauliflower is very susceptible to heat stress, which will affect its physical appearance thus affecting its overall market value (Aleem et al., 2021). Agricultural operations cannot afford to risk lower production rates, which is why research is needed to test the varieties deemed ‘heat tolerant’ at the local level. Cauliflower varieties that are marketed as heat tolerant in other countries or states within the U.S. does not mean they will be successful in California’s climate conditions.

5.1 Future Research Considerations

To truly evaluate the relationship between plant stress, photosynthetic rate and yield, this type of experiment needs to assess each individual plant rather than the population for each variety. The large distribution of data points for the stomatal conductance and chlorophyll fluorescence illustrates the large amount of variance among the genotypes within each cauliflower variety. It is recommended to have the individual plant yield to compare with the individual plant stress (stomatal conductance) and photosynthetic rates (chlorophyll fluorescence). By having the data for each individual plant, it could also be used for breeding superior genotypes of that particular cauliflower variety in the future.

Another aspect that could have influenced our similar results were that the climate conditions were too mild—meaning the plants were not stressed enough to show any noticeable signs of stress or major differences in yield (between the Bishop and the Flame Star). To determine if this could have influenced our results, it is recommended to introduce a cool season control variety into this study. Evaluating a cool season cauliflower crop on California’s central coast during the summer can act as a proxy for
what winter will become due to climate change. The data obtained from a cool season variety would be expected to illustrate a large contrast to the data obtained from the heat tolerant varieties.

There is another parameter, not accounted for in this study, that is causing either biotic or abiotic stress; specifically affecting the Mardi variety. It would be helpful to reassess the cauliflower varieties for heat tolerance by evaluating the characteristics of their cellular and metabolic responses to heat stress (Aleem et al., 2021; Asthir, 2015; Bray et al., 2000). A specific response to heat stress that could be evaluated is the synthesis of proteins, specifically heat shock proteins (HSPs). An increased synthesis of HSPs and a decrease in normal protein synthesis is a signature response to heat stress, which is an indicator that can be measured in future research experiments. Incorporating the assessment of metabolic responses to increased temperatures in heat tolerant vegetable cultivars will help to account for an additional parameter that could influence the difference among the yield results.
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