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**QUANTIFYING SPATIAL HETEROGENEITY OF WILD BLUEBERRIES AND CROP WATER STRESS
MONITORING USING REMOTE SENSING TECHNOLOGIES**

By

Kallol Barai

B.S. Shahjalal University of Science and Technology, Bangladesh 2018

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Submitted in Partial Fulfillment of the
Requirements for the Degree of
Master of Science
(in Botany and Plant Pathology)

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The University of Maine

August 2022

Advisory Committee:

YongJiang Zhang, Assistant Professor of Plant Physiology, Advisor

Matthew Wallhead, Assistant Professor of Horticulture

Parinaz Rahimzadeh-Bajgiran, Assistant Professor of Remote Sensing of Natural Resources

José Eduardo Meireles, Assistant Professor of Plant Evolution and Systematic

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Thesis Advisor: Dr. Yongjiang Zhang

An Abstract of the Thesis Presented
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The wild blueberry is one of the major crops of Maine, with significant economic value and potential health benefits. Due to global climate change, drought impacts have been increasing significantly in recent years in the northeast region of the USA, causing significant economic losses in the agricultural sectors. It has been predicted to increase further in the future. Changing patterns of the elevated atmospheric temperatures, increased rainfall variabilities, and more frequent drought events have made the wild blueberry industry of Maine vulnerable, suggesting the adoption of novel approaches to mitigate the negative impacts of global climate changes. Also, wild blueberry fields show high spatial heterogeneity, making precise and effective management difficult. Our research focuses on quantifying the spatial heterogeneity in functional traits of wild blueberries, analyzing the impact of historical drought on wild blueberry production, and testing the use of drone-based thermal sensors to quantify spatial heterogeneity in water stress across wild blueberry fields.

In chapter two, we aimed to quantify the inter-genotype variation in several structural, functional, and yield-related traits and to establish the relationship between functional traits and yield-related traits.

We conducted a study during the 2019 harvest season measuring several structural, functional, and yield traits from two wild blueberry farms. We found high variations in structural, functional, and yield-related traits among genotypes but not between the two fields, confirming the spatially heterogeneous nature within wild blueberry fields. We also found negative associations of the leaf mass per unit area and midday leaf temperature with the yield, whereas the leaf chlorophyll concentration was positively associated with the yield. Additionally, we found quadratic relationships between some yield-related traits and stem length, with the optimum stem length for yield at 25 cm. Our results suggest that some leaf and stem functional traits can be used to predict wild blueberry yields.

In chapter three, we analyzed historical drought patterns using a drought index Standardised Precipitation-Evapotranspiration Index (SPEI). We assessed drought impacts on production (yield) and remotely sensed vegetation indices; Enhanced Vegetation Index (EVI) and Normalized Difference Vegetation Index (NDVI) of the wild blueberry fields in Maine, USA. Despite a significant warming pattern, we found no significant changes in SPEI in the past 71 years. We also analyzed the impact of short and long-term water conditions (SPEI) during the growing season on the wild blueberry vegetation condition and production. We found that drought has had a significant impact on vegetation status and production historically. Further, the relationship between the relatively long-term SPEI and vegetation indices EVI and NDVI was significantly more substantial than short-term SPEI, suggesting water conditions in a relatively long-term probably determine crop health. We also compared an irrigated and non-irrigated wild blueberry field at the same location (Deblois, Maine). We found that irrigation decoupled the relationship between SPEI and vegetation indices and yield, suggesting the need for effective irrigation strategies to mitigate drought impacts.

In chapter four, we tested the use of remotely sensed canopy temperature-based crop water stress index (CWSI) to remotely and non-destructively detect the water status of wild blueberries. By detecting crop water status using the CWSI, irrigation can be intelligently controlled in the highly spatially

heterogeneous wild blueberry fields to increase efficiency and profitability. A drone-based thermal sensor was used to acquire the canopy temperature data remotely and then calculate CWSI. CWSI calculated from bio-indicator based T_{wet} and T_{dry} reference was found to be effective ($R^2 = 0.78$; $p < 0.05$) in detecting leaf water potential (LWP), which is superior compared to the statistical T_{wet} and empirical T_{dry} reference-based CWSI. The CWSI-LWP model-based crop water status (LWP) maps showed high variability in crop water stress within irrigated and non-irrigated fields, suggesting the need for precise water stress monitoring and management in wild blueberry fields

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CHAPTER 1: WILD BLUEBERRIES OF MAINE: ECONOMIC IMPORTANCE, NUTRITIONAL VALUE, CURRENT MANAGEMENT PRACTICES & UPCOMING CHALLENGES

The wild lowbush blueberries include a few species in the genus *Vaccinium* (mainly *Vaccinium angustifolium* Aiton) under the Ericaceae family. Wild blueberries are outcrossing, tetraploid ($2n = 4 \times = 48$), rhizomatous, and woody perennial plants (Beers et al., 2019). There are only about four native fruit crops in North America, and the wild blueberry is one of them. As a cultural heritage from the native indigenous people, the wild blueberry has developed into an important crop in the northeast (NE) region of the United States and part of Canada (Percival et al., 2003). Wild blueberries have a rich and unique diversity of flavors due to their genotypic variation along with a very high percentage of antioxidants than other fruits (Prior et al., 1998; Wolfe & Liu, 2007). High antioxidants in blueberries can help neutralize free radicals in human bodies and thus have many potential health benefits, i.e., preventing diseases related to aging, inflammation, cancer and heart diseases by canceling out unstable oxygen molecules associated with these diseases, thus making wild blueberries economically very important (Cho et al., 2004; Duffy et al., 2008; Kalt et al., 2007; Rimando et al., 2004). It is a vital crop in the state of Maine, with an economic value of 27.7 million dollars in 2020 which has been increasing over the years (NASS, 2020). Maine is the top producer of wild blueberries in the world, providing 40% of the world supply and 99% of the wild blueberries consumed in the US (Bell, 2009; Yarborough, 2009a). Within North America, the state of Maine produces about 10% of all blueberries, including both wild and highbush varieties (Yarborough, 2009a).

The wild blueberry production system is a semi-natural system with plants naturally growing in the fields but managed. In Maine, there are approximately 44,000 acres of commercially managed wild blueberries (Yarborough, 2012). Commercial wild blueberry fields have been developed from naturally occurring native wild blueberry plants in the understory of natural NE forests (Yarborough, 2012). Although most commercial fields originated from naturally growing plants, propagation techniques are

still possible, especially by growing from rhizome cuttings and softwood cuttings from the field (Yarborough, 2009A). Each seed produced by a wild blueberry plant forms a distinct plant expanding to form a carpet with multiple aboveground stems, which has been called a clone as it reproduces through cross-pollination. Rhizomes, or underground stems, are present in mature plants and grow just below the soil surface (~10 cm), sprouting new stems above it. The rhizomes grow roots as they mature. The original plant along with its spreading rhizome system is known as a genotype or commonly as a clone by the growers.

Different traits such as leaf size, leaf shape, leaf and stem colors, disease resistance, berry size, flavor, and total yield differ substantially between different genotypes (Bell, 2009; Barai et al., 2022). The size of genotypes varies, but the amount of space they occupy also depends on their age; the younger genotypes use less space. According to Yarborough 2009A, Maine may have more than 4.75 million genotypes. Maine is home to several different wild blueberry species, with *Vaccinium angustifolium* and *Vaccinium myrtilloides* being the most prevalent. Commercial wild blueberry cultivation has a two-year cropping cycle that alternates between a vegetative year and a production or crop year (Bell, 2009). The majority of wild blueberry fields are entirely pruned (or burned) every other year. Due to pruning practice, only half of the fields is available for harvest each year (Bell, 2009). Vegetative and developmental growth occurs in wild blueberries during the growing season immediately after pruning. During the end of this season, flower buds form (Yarborough, 2015). The flowering phase passes between two and four weeks. Cross-pollination is generally required for a successful fruit set in wild blueberries (Bell et al., 2009). Successful pollination forms the fruit set, and the fruit set of the blueberries fully develop and ripens (Bell et al., 2009). Yield is a complex trait determined by multiple factors in crops including wild blueberries. Previously, researchers have found that the major factors affecting yield in wild blueberries are adequate irrigation, effective pollination, pruning practice, weed control, insect-pests control, crop diseases, soil characteristics, genotype-specific genetic composition, and weather factors

such as winter damage, summer drought, and spring frosts. (Bell et al., 2010; Drummond & Collins, 2020; Farooque et al., 2012; Yarborough et al., 2016; Yarborough, 2004a, 2012).

In recent years, severe warming and drought incidents have been reported in the NE region of the USA, which is expected to increase further in the future, making the agricultural systems vulnerable and causing significant economic losses (Fernandez et al., 2020; Sweet et al., 2017; Wolfe et al., 2018). However, the historical trends of drought, their impacts on wild blueberry agricultural systems in this region, and the effect of current irrigation practices to mitigate the impact of drought have not been carefully assessed. Wild blueberry yields range from 4,000 to 5,000 pounds per acre, but with more intensive precise management and high-yielding genotypes, the production could be significantly higher (Yarborough, 2004b). Nevertheless, current conventional management practices (i.e., irrigation, fertilization) treat the field uniformly without considering the spatial variability of genotypes and soil properties within a field. These uniform management practices may result in over or under-application of resources (i.e., water, fertilizer) within a field. Uniform management practices along with recently increased drought conditions can result in reduced yield and profitability and affect the sustainability of water use and agricultural production; overall making the wild blueberry industry vulnerable. So, there is a need for an efficient management system considering the spatial heterogeneity for wild blueberries along with upcoming projected drought events. To do that, the overall objectives of my research were to:

- 1) Quantify the spatial heterogeneity of several functional traits and structural traits and their impact on yield;
- 2) Characterize impacts of historical drought on wild blueberry agricultural systems and the effect of current irrigation practices to mitigate the impact of drought;
- 3) Test the remote detection of crop water stress and detection of the spatial variation of Wild blueberry water status using a UAV-based thermal sensor.

CHAPTER 2: HIGH VARIATION IN YIELD AMONG WILD BLUEBERRY GENOTYPES: CAN YIELD BE PREDICTED BY LEAF AND STEM FUNCTIONAL TRAITS?

2.1 Abstract

Wild lowbush blueberry fields are characterized by high genetic diversity, with a large number of genotypes coexisting in every field. Yield also varies among genotypes, which could be related to the variation in physiological and structural traits, but this has not been rigorously tested. In this study, we aimed to quantify the inter-genotype variation in yield, as well as leaf and stem functional traits, and to establish the relationship between functional traits and yield-related traits in wild blueberries. To do so, we carried out a study during the 2019 harvest sea-son measuring structural and functional traits including stem number, stem length, stem diameter, leaf chlorophyll concentration, leaf mass area, leaf area per stem, leaf number per stem, number of branches per stem, leaf temperature, soil temperature, and soil water content and yield data including yield, berry size (weight of 100 berries), number of berries per stem, and length of berry cluster from two wild blueberry farms. We found high variations in structural, functional, and yield-related traits among genotypes, but not between two fields. We also found negative associations of the leaf mass per unit area and midday leaf temperature with the yield, whereas the leaf chlorophyll concentration was positively associated with the yield. Additionally, we found a quadratic relationship between yield-related traits (weight of 100 berries, number of berries per stem, and length of berry cluster) and stem length, with the optimum stem length for yield at 25 cm. Our results suggest that several leaf and stem functional traits are related with yield-related traits; thereby, those traits can be used to predict wild blueberry yields. Our findings could help growers and breeders select better-yielding genotypes based on structural and functional traits.

2.2 Introduction

Wild blueberry (*Vaccinium angustifolium* Aiton) is an important crop of Maine, USA, Quebec, and maritime Canada. It mostly reproduces by obligate cross-pollination (Drummond, 2019b). Each seed gives rise to a genetically unique plant, which expands horizontally in the field via the vegetative (clonal) growth of rhizomes and forms a patch of connected aboveground stems (Qu & Drummond, 2018). A patch of stems with identical genetic information is called a genotype or clone with a different genetic make-up than other wild blueberry genotypes (Bell et al., 2009). Among genotypes, there are significant differences in morphology, structural and functional traits, physiological performance, and yield (Bell et al., 2009; Drummond, 2020; Tasnim & Zhang, 2021). Crop yield is regulated by several genes named quantitative trait loci and is also influenced by external environmental factors (Wang et al., 2012). Yield is also indirectly determined by several morphological and physiological traits (Wang et al., 2012). An optimal plant architecture is beneficial for higher yields (Xue et al., 2008). In wild blueberries, the high intergenotype variations in structural and physiological traits can indirectly impact the overall yield of wild blueberries, but the associations have not been established.

The yield of crops is strongly associated with and dependent on plant growth and development. Plant architecture, leaf structure, and vascular architecture are major developmental features of plants (Mathan et al., 2016). Plant functional traits can directly reflect the physiological performance of a plant and its response to environmental change and stress (Yan et al., 2021). Many studies have shown that leaf features and plant architectural traits can predict plant growth and performance as the quantity of light interception, photosynthetic capacity, and the source energy of plants are determined by several functional traits (Falster & Westoby, 2003; Mathan et al., 2016). Also, the mobilization of photosynthates from source to sink, which is related to vascular architecture, is crucial for the efficient partitioning of photo-assimilated carbon (Mathan et al., 2016). Thus, these features can be considered the parts of a developmental module that dictates crop performance and yield; the optimization of these

developmental features is essential for the efficient performance of crop plants. Genotypes of wild blueberries with a greater vegetative biomass, which is represented by several structural traits, i.e., stem height and number of leaves, can produce more flowers, which ultimately results in higher fruit biomass (Fournier et al., 2020). Plant stem height and diameter are closely related to biomass production and are important morphological traits affecting yield performance (Yan et al., 2021). Plants with a higher stem height and larger diameter stems normally have larger diameter vessels, leading to a higher hydraulic conductance (Sperry et al., 2006). A higher hydraulic conductance facilitates higher stomatal conductance, leading to more photosynthetic carbon gain (Santiago et al., 2004). Another critical factor in hydraulic conductance is leaf vein density (Brodribb et al., 2007). High leaf vein density leads to higher photosynthetic and growth rates by providing a faster water supply (McDowell et al., 2002). However, with increasing height, the risk of xylem cavitation also increases, leading to more vulnerability to drought compared to shorter plants (McDowell et al., 2002; Phillips et al., 2010). Wild blueberry plants with a higher stem height might also be more prone to winter wind damage and could result in reduced yield. That is why an optimal architecture is required to achieve a high yield (Xue et al., 2008), and an understanding of the optimal architecture for the wild blueberry crop could aid in the selection process of cultivar development.

Physiological traits of crops determine the health of plants and correlate with yield. Crop yield can be increased by (i) increased photosynthesis per unit land area, which can be achieved by effective agronomic practices such as irrigation, fertilization, and pollination and (ii) the increased partitioning of crop biomass to the harvested product, which can be achieved by plant breeding (Richards, 2000). Leaf chlorophyll concentration (LCC) is a good indicator of leaf photosynthetic capacity and is positively correlated with yield (Ghimire et al., 2015). In wheat, researchers have found positive correlations of yield with the rate of CO₂ fixation, LCC, and stomatal conductance and a negative correlation with the loss of chlorophyll (Silva-Pérez et al., 2020). Understanding the effects of LCC on wild blueberry yield can explain

the soil nutrient availability in a field and the uptake of nutrients because LCC production in leaves depends on the nitrogen and moisture availability in the soil (Percival & Sanderson, 2004). Understanding the relationships between LCC and yield can also help facilitate the selection of high-yielding genotypes. Plant canopy temperature is related to stomatal conductance and water stress (Silva-Pérez et al., 2020). A negative correlation between yield and canopy temperature was found in wheat (Silva-Pérez et al., 2020). Canopy temperature can be used to assess the health condition status of a plant and allow an assessment of the current on-farm water use efficiency and the development of mitigation strategies.

Plant breeding can increase crop yield by achieving a high-rate partitioning of crop biomass to the harvested product (Richards, 2000). However, the breeding of wild blueberry is complicated due to the characteristics of heterozygosity, ploidy, complex heritability of QTLs, and long timeframe for establishing mature fruit-bearing progeny (Bell et al., 2009). Hall (1983) noted improvements in wild blueberry cultivars in terms of size and yield resulting from a plant breeding program (Hall, 1983). Researchers also found heritable genetic components linked to wild blueberry yield (Bell, 2009). Griffing (1956) also demonstrated evidence of phenotypic selections that resulted in genetically advanced inbred lines (Griffing, 1956). The overall productivity of blueberry fields can be improved by increasing blueberry cover in between inter-genotype empty spaces in existing fields and replacing the less productive genotypes with high-yielding genotypes (Hepler & Yarborough, 1991). Understanding the direction and strength of the relationships between yield-related traits and functional traits is very important to improve the efficiency of genetic selection in plant breeding programs (Li et al., 2019). Studies have quantified associations between yield-related traits and functional traits in different crops (Ghimire et al., 2015; Li et al., 2019; Reddy, 2019; Reynolds et al., 1994), but these associations have not been quantified in the wild blueberry.

To do so, the objectives of this research were to:

- 1) Quantify the variation of major leaf and stem functional traits and yield-related traits among selected genotypes and between fields of wild blueberries, and
- 2) Establish relationships between plant functional traits and yield-related traits in wild blueberries. This study will help to clarify the magnitudes and directions of correlations among functional traits and yield-related traits in wild blueberries.

This can also be helpful for both growers and breeders in selecting better yielding genotypes based on structural and functional traits.

2.3. Materials and Methods

2.3.1. Study Sites and Plant Materials

The study sites were located in Washington County in the Downeast coastal region of Maine, USA. One site was located at the University of Maine Blueberry Hill Farm (BBHF) in Jonesboro (Latitude: 67°38'53"W, Longitude: 44°38'44"N), Maine, USA, and another site was located at Jasper Wyman & Son (Wyman's) blueberry farm (Latitude: 67°59'58"W, Longitude: 44°44'07"N) in Deblois, Maine, USA. Soils at both sites were well-drained acidic sandy loam with 0–3% slope. The Downeast coastal region of Maine has a temperate climate with an average low of –10.6 °C and a high of 24.2 °C, and a monthly average precipitation low of 85.1 mm and high of 136.4 mm. Wild blueberries are managed on a two-year cycle; in prune years, plants are pruned and grow vegetatively, and in harvest years, the plants flower and produce a fruit crop. The study was conducted in 2019, which was a crop or harvest year for both fields. Wild blueberry fields contain numerous diverse genotypes (Beers et al., 2019); 30 genotypes were selected from BBHF, and 15 genotypes were selected from Wyman's based on their morphological and phenological differences to include a high variety of genotypes. Different genotypes could be easily

identified visually based on morphological and phenological differences including leaf color, stem color, berry density, berry color (Figure 2.1), as well as phenology.



Figure 2.1 High variations in leaf color, stem height, berry density, and berry color among genotypes in a wild blueberry field at the Wyman’s Farm, Deblois, Maine, USA. Different genotypes form patches connecting each other in the field. The photo was taken during the fruit production stage by Dr. Xiaoxue Mo.

2.3.1.1 Leaf and Stem Structural Traits

Six wild blueberry shoots were arbitrarily selected from each randomly selected genotype of the BBHF and Wyman’s fields in August 2019 during the harvest season. Structural traits including stem length and diameter, leaf number, as well as total leaf area and dry leaf biomass in a stem were measured. A caliper was used to measure stem length (from the soil line) and diameter. Leaf area was determined using an LI-3000A area meter (Li-Cor, Lincoln, NE, USA), after which leaves were oven-dried at 70°C to constant mass and weighed. Leaf mass per area (LMA) was determined as leaf dry mass divided by leaf area (g/m^2).

2.3.1.2 Leaf Chlorophyll Concentrations

Six wild blueberry stems were arbitrarily selected from each genotype to measure leaf chlorophyll concentration (LCC). Chlorophyll concentration per unit leaf area was measured in the field with a SPAD Chlorophyll Meter (SPAD 502; Minolta Corp., Osaka, Japan) on samples from the Wyman's Farm and with an atLEAF Digital Chlorophyll Meter (FT Green LLC, Wilmington, DE, USA) on samples from BBHF. Both the SPAD and atLeaf data were converted to values for LCC ($\mu\text{g}/\text{cm}^2$) by the formula relating SPAD/atLEAF values and laboratory-measured chlorophyll values using chemical extraction methods (Zhu et al., 2012).

2.3.1.3 Leaf Temperature, Soil Temperature, and Soil Water Content

Leaf temperature (LeafT) was measured in the field by using a Fluke 62 Max+ handheld infrared thermometer (Fluke Corporation, Everett, WA, USA) on four arbitrarily selected leaves from each genotype. Soil temperature (SoilT) and soil water content (SWC) were measured in the field by a Fieldscout TDR 150 Soil Moisture Meter (Spectrum Technologies Inc., Aurora, IL, USA) from four random places within the boundaries of each genotype. These measurements were taken during midday (12:00 to 14:00) on 4 August 2019, a sunny day, and the leaves were exposed to direct sunlight. The measurements were carried out under similar conditions for all genotypes. These measurements were only taken at Wyman's Farm due to limited resources and the time taken to measure each genotype.

2.3.1.4. Yield and Yield-related Traits

Yield and yield-related traits were quantified by establishing a 0.3 by 0.3-m quadrat within each genotype (Table 2.1). A hand rake was used to collect the fruit sample within each quadrat and then weighed on a portable balance in the field to obtain the total fruit yield (g) per quadrat. The weight (g) of 100 berries (WT) from each quadrat was also quantified as a measure of berry size. The length of berry cluster (LBC) in stem (cm) and the number of berries per stem (NBS) were quantified in the field by selecting six stems in the quadrat. All measurements were conducted at both research sites.

Table 2.1 List of yield-related, morphological, and functional traits used in this study along with their abbreviations and units.

Traits Type	Traits Name	Abbreviation	Unit
Yield Traits	Yield per area	Yield	g/m ²
	Berry size (Weight of 100 berries)	WT	g
	Number of berries per Stem	NBS	#
	Length of berry cluster	LBC	cm
Structural Traits	Number of stems per plot	StemN	#/quadrat
	Stem length	StemL	cm
	Stem diameter	StemD	cm
	Leaf number per stem	LNPS	#
	Number of branches per stem	NBPS	#
Functional Traits	Leaf chlorophyll concentration per area	LCC	µg/cm ²
	Leaf mass per area	LMA	g/m ²
	Leaf area per stem	LAPS	cm ²
	Leaf temperature	LeafT	°C
	Soil temperature	SoilT	°C
	Soil water content	SWC	%

2.3.2. Calculation for Estimating Optimal Sample Size of Genotypes to Estimate Yield

Using estimates of variance from the genotype yield data collected in the two fields and two levels of precision (SE/mean ratios of 0.10 and 0.25), the optimal sample size of genotypes for estimating yield was calculated using the formula developed by Cochran (1977) (*Sampling Techniques, 3rd Edition* | Wiley, n.d.):

$$N = (t_{(0.05)} \times S^2 / (m^2 \times P^2))$$

where N = optimal sample size for a given level of precision; $t_{(0.05)}$ is 1.96 or the t -value for $\alpha = 0.05$ when n approaches ∞ ; S^2 = estimated variance of yield/genotype; m = mean yield/genotype; and P = level of precision defined as the proportion of the standard error/mean.

The use of the t -value in the formula estimates the optimal sample size for a 95% likelihood of obtaining the desired precision in yield for the calculated number of genotypes required.

2.3.3. Data Analyses

Statistical analyses were conducted using SPSS v23 (IBM Corp., Armonk, NY, USA), JMP v15 (SAS Institute Inc., Cary, NC, USA), and RStudio software (RStudio, PBC, Vienna, Austria). The truncated gaussian kernel density estimation was performed using the ggplot package in RStudio. To determine trait differences between the two fields and genotypes within fields, we used a hierarchical nested mixed model with the field as the fixed effect and the genotype nested within the field as the random effect ($\alpha = 0.05$). Harrison et al. suggested this approach for split-plot designs (genotypes within fields) (Harrison et al., 2018). The statistical software JMP version 15 was used to fit the model with the restricted maximum likelihood. The Pearson correlation analysis was conducted using the corrplot package in RStudio at an alpha (α) level of 0.05 and a 95% confidence interval. The structural and functional traits were averaged within genotypes for a regression analysis using SPSS. We analyzed the relationship between functional traits using linear (in the form of $a + bx$) or quadratic (in the form $a + bx + cx^2$) regressions according to which best approximated the structure of the relationship. We determined the statistical significance of the relationship using the coefficient of determination and its significance (α) at $p < 0.05$. With the mean trait values of each genotype, a Principal Component Analysis (PCA) was employed to characterize the variance of structural and functional traits for the two examined fields in RStudio. Multiple linear regression analysis was conducted in RStudio to test the overall contribution of several functional traits (StemL, LMA, and LCC) on the yield for studied fields. We used a generalized linear model with the Gaussian or Normal distribution and the identity linkage function. The model was fit using maximum likelihood.

2.4. Results

2.4.1. Estimation of Optimal Sample Size of Genotypes to Estimate Yield

Based upon the average yields in the two fields, we estimated that 31 genotypes should be sampled if it is desired to estimate the yield of a field with a standard error\mean ratio precision of 0.25. If higher precisions are desired, the necessary sample sizes increase geometrically: 196 genotypes for a precision of 0.10 and 784 genotypes for a precision of 0.05. However, if one is willing to sacrifice 95% confidence in estimating yield with a given precision, then 16, 100, and 400 genotypes would need to be sampled to estimate yield with precisions of 0.25, 0.10, and 0.05, respectively.

2.4.2. Variation in Yield, Structural and Functional Traits between Wyman’s and BBHF Blueberry Fields and among the Genotypes

We found that both Wyman’s and BBHF fields had yield distributions that were heavily skewed toward the yield range 2200–2500 (g/m²) (Figure 2.2). Though the average yield in the BBHF field appeared higher compared to the Wyman’s field yield (Figure 2.2), the difference was not statistically significant (Table 2.2).

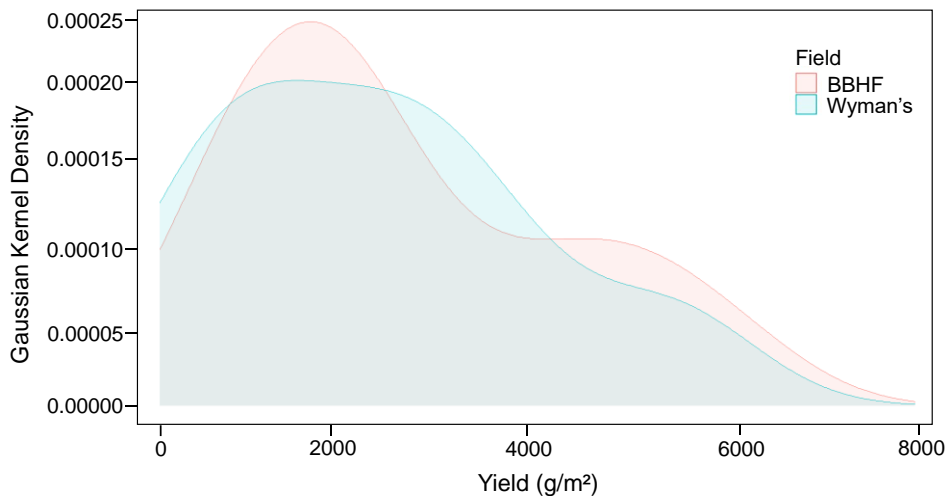


Figure 2.2. Gaussian kernel density estimates of the wild blueberry yield (g/m²) for the two studied wild blueberry fields, Wyman’s and BBHF.

Table 2.2. Comparison between Jasper Wyman & Son Blueberry Farm (Wyman’s) and from Blueberry Hill Farm (BBHF) blueberry fields in yield-related, morphological, and functional traits in the minimum (Min), maximum (Max), mean, and standard deviation (SD) of Yield, WT, NBS, LBC, StemN, StemL, StemD, LCC, LMA, LAPS, LNPS, NBPS, LeafT, SoilT, and SWC. To determine trait differences between the two fields and genotypes within fields, we used a hierarchical nested mixed model with field as the fixed effect and genotype nested with-in field as the random effect. LeafT, SoilT, and SWC data were only available for the Wyman’s field. For definitions of trait abbreviations, please see Table 2.1.

		<i>Yield</i> (g/m ²)	<i>WT</i> (g)	<i>NBS</i> (#)	<i>LBC</i> (cm)	<i>StemN</i> (#/plot)	<i>StemL</i> (cm)	<i>StemD</i> (cm)	<i>LCC</i> (µg/cm ²)	<i>LMA</i> (g/m ²)	<i>LAPS</i> (cm ²)	<i>LNPS</i> (#)	<i>NBPS</i> (#)	<i>LeafT</i> (°C)	<i>SoilT</i> (°C)	<i>SWC</i> (%)
<i>Wyman’s</i>	Min	122.22	40.00	0.83	0.45	126.00	14.73	1.64	0.10	64.50	26.00	29.17	5.33	25.40	31.32	9.75
	Max	5577.70	69.00	20.50	5.75	289.00	28.32	2.47	0.34	82.85	95.85	102.80	17.33	32.26	33.0	22.15
	Mean	2332.30	50.90	7.63	2.87	179.80	20.91	2.05	0.17	72.02	65.03	57.71	7.91	28.42	32.13	14.21
	SD	1685.58	7.81	5.78	1.56	52.91	4.08	0.25	0.06	5.19	21.65	22.81	2.89	2.20	0.61	3.22
	Difference Among Genotypes	NA ¹	NA ¹	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)	NA ¹	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)	No (<i>p</i> = 0.37)	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)
<i>BBHF</i>	Min	177.77	22.00	1.87	0.58	79.00	10.73	1.36	0.09	60.94	29.98	22.33	5.17			
	Max	6355.50	88.00	26.67	3.95	246.00	24.34	2.78	0.31	81.00	105.9	62.33	12.92			
	Mean	2588.80	43.60	10.16	2.27	136.70	17.69	2.17	0.20	70.99	59.02	43.15	8.38			
	SD	1760.60	15.60	5.21	0.89	38.26	3.75	0.34	0.06	5.46	22.91	12.21	1.92			
	Difference Among Genotypes	NA ¹	NA ¹	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)	NA ¹	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)	No (<i>p</i> = 0.98)	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.001)	NA ²	NA ²	NA ²
Difference Between Fields	No (<i>p</i> = 0.59)	No (<i>p</i> = 0.06)	No (<i>p</i> = 0.14)	No (<i>p</i> = 0.11)	Yes (<i>p</i> < 0.001)	Yes (<i>p</i> < 0.05)	No (<i>p</i> = 0.26)	No (<i>p</i> = 0.13)	No (<i>p</i> = 0.55)	No (<i>p</i> = 0.47)	Yes (<i>p</i> < 0.0001)	No (<i>p</i> = 0.53)	NA ²	NA ²	NA ²	

¹ no genotype subsample (stems) contribution. ² no sample for the BBHF. ³# refers to number.

We found a high variation in the observed yields, structural and functional traits among genotypes, and within and between fields (Table 2.2). The yield varied by 46-fold, ranging from 122.22 to 5577.7 g/m² in the Wyman’s field. The yield varied by 36-fold, ranging from 177.77 to 6355.5 g/m² in the BBHF field. The weight of 100 berries varied by 1.7-fold, ranging from 40 to 69 g in the Wyman’s field, and varied by 4-fold, ranging from 22 to 88 g in the BBHF field. The number of berries on a stem varied by 24.7-fold, ranging from 0.8 to 20.5 in the Wyman’s field, and varied by 14.3-fold, ranging from 1.9 to 26.7 in the BBHF field. The length of a berry cluster varied by 12.8-fold, ranging from 0.5 to 5.8 cm in the Wyman’s field, and varied by 6.8-fold, ranging from 0.6 to 4.0 cm in the BBHF field. Stem length varied by 1.9-fold, ranging from 14.7 to 28.3 cm in the Wyman’s field, and varied by 2.3-fold, ranging from 10.7 to

24.3 cm in the BBHF field. Leaf mass area varied by 1.3-fold, ranging from 64.5 to 82.9 g/m² in the Wyman's field, and varied by 1.3-fold, ranging from 60.9 to 81.0 g/m² in the BBHF field. Leaf chlorophyll concentration (LCC) varied by 3.4-fold, ranging from 0.1 to 0.3 µg/cm² in the Wyman's field, and varied by 3.4-fold, ranging from 0.1 to 0.3 µg/cm² in the BBHF field. Leaf canopy temperature varied by 1.3-fold, ranging from 25.4 to 32.3 °C in the Wyman's field.

When comparing between the fields, we did not find any significant differences between Wyman's and BBHF in yield-related traits: yield (g/m²), weight of 100 berries (WT), number of berries on a stem (NBS), and the length of a berry cluster (LBC) (Table 2.2). We found significant differences between the two fields for stem structural traits including the stem number per plot (StemN) and stem length (StemL) but not for the stem diameter (StemD). Both total StemN and mean StemL were higher in the Wyman's field compared to the BBHF field (Table 2.2). In terms of leaf structural and functional traits, a significant difference was only found for leaf number per stem (LNPS), and mean LNPS was higher in the Wyman's field.

When comparing among genotypes of each field, significant differences in yield-related traits; NBS and LBC were found among the genotypes of the Wyman's field as well as the BBHF (Table 2.2). We found significant differences among genotypes for stem structural traits including stem length (StemL), and stem diameter (StemD). In terms of leaf structural and functional traits, we found significant differences among genotypes LCC and leaf area per stem (LAPS) but not between the Wyman's and BBHF fields. The same pattern was found for the number of branches/stem (NBPS). The LNPS was found significantly different between fields as well as among the genotypes in those fields. We did not find any significant difference in leaf mass per area (LMA) between the fields or among the genotypes in those fields. Significant differences among genotypes in Wyman's field were also found with water condition, leaf temperature (LeafT), soil temperature (SoilT), and soil water content (SWC).

2.4.3 Structural and Functional Traits Relationship with Yield and Yield Related Traits of Wild

Blueberry Fields

We used Pearson correlation as well as bivariate linear and quadratic regression relationships to determine if different structural and functional traits can be used to predict yield and yield-related traits WT, NBS, and LBC for the wild blueberries. We found relationships of yield with the LMA, LCC, and LeafT, but the relationships were not always consistent within and across fields (Figures 2.3 and 2.4). We found significant negative linear relationships between the yield and LMA for the Wyman's field (Figure 2.4a: $R^2 = 0.41$, $p < 0.05$) as well as combining the data of the two fields (Figure 2.4c: $R^2 = 0.18$, $p < 0.05$), but not for the BBHF field. We also found significant quadratic relationships for Wyman's field (Figure 2.4a: $R^2 = 0.37$, $p < 0.05$) as well as for the combined field data (Figure 2.4c: $R^2 = 0.10$, $p < 0.05$), whereas the quadratic relationships were not significant for the BBHF field (Figure 2.4b: $R^2 = 0.07$, $p = 0.16$). However, coefficients of determination (R^2) were higher for the linear relationships compared to quadratic relationships between yield and LMA. When we analyzed the relationship between LCC and yield, we did not find any significant linear or quadratic relationships for Wyman's field or the BBHF field. When the data for the two fields were combined, we found a borderline significant quadratic relationship between LCC and yield (Figure 2.4f: $R^2 = 0.11$, $p = 0.08$). We also found significant linear (Figure 2.4g: $R^2 = 0.42$, $p < 0.01$) and quadratic relationships (Figure 2.4g: $R^2 = 0.42$, $p < 0.05$) when we analyzed the relationship between LeafT and Yield(g/m^2).

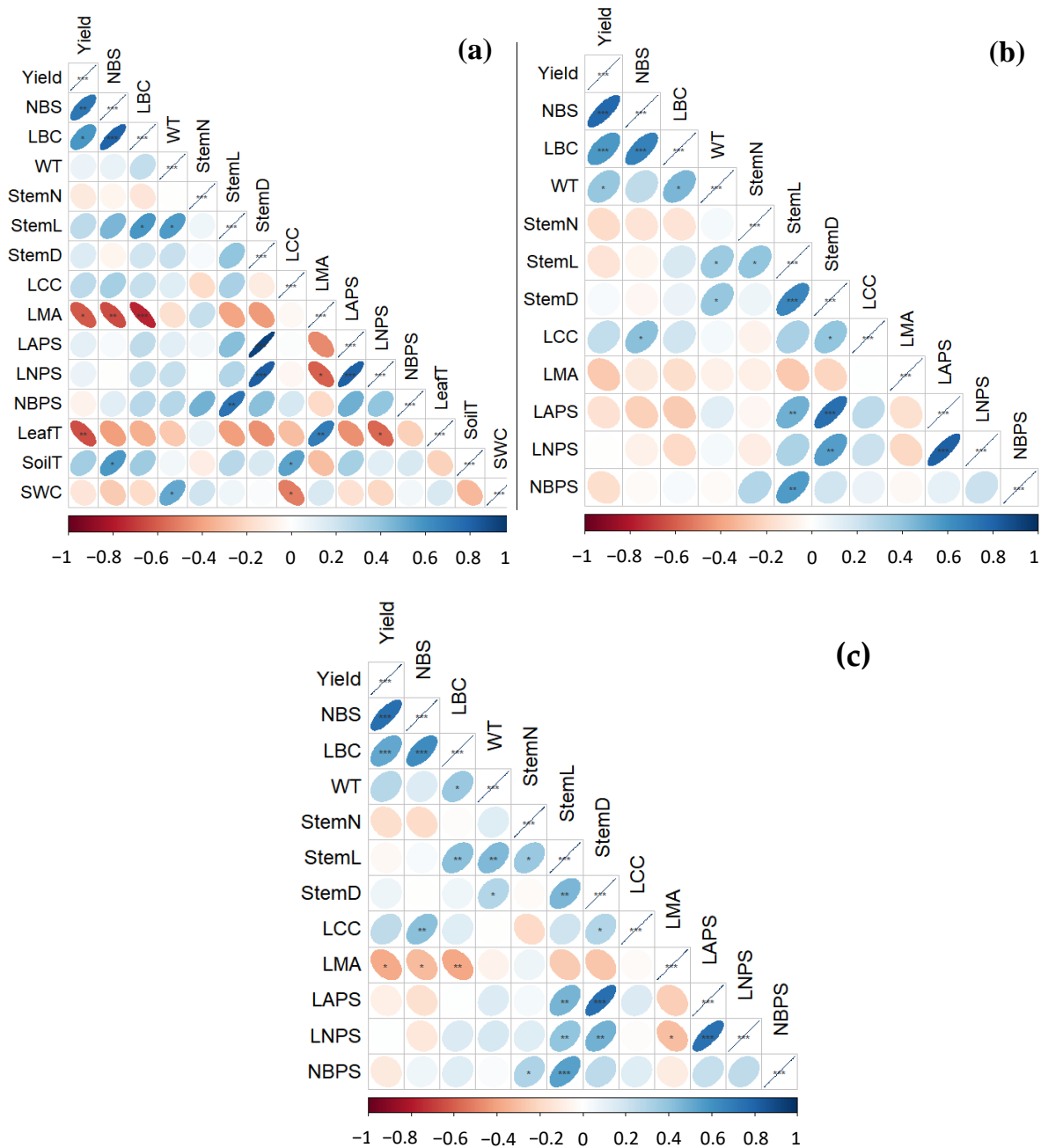


Figure 2.3. Pearson correlation heat map of the structural, functional, and yield-related traits of wild blueberries for (a) Wyman's field, (b) BBHF field, and (c) the combined data from both fields. The horizontal axis represents the degree and direction of Pearson correlation r . The sign of the significance for each correlation is shown as ***, $p < 0.001$; **, $p < 0.01$; *, $p < 0.05$. For definitions of trait abbreviations, please see Table 2.1. LeafT, SoilT, and SWC data were only available for the Wyman's field.

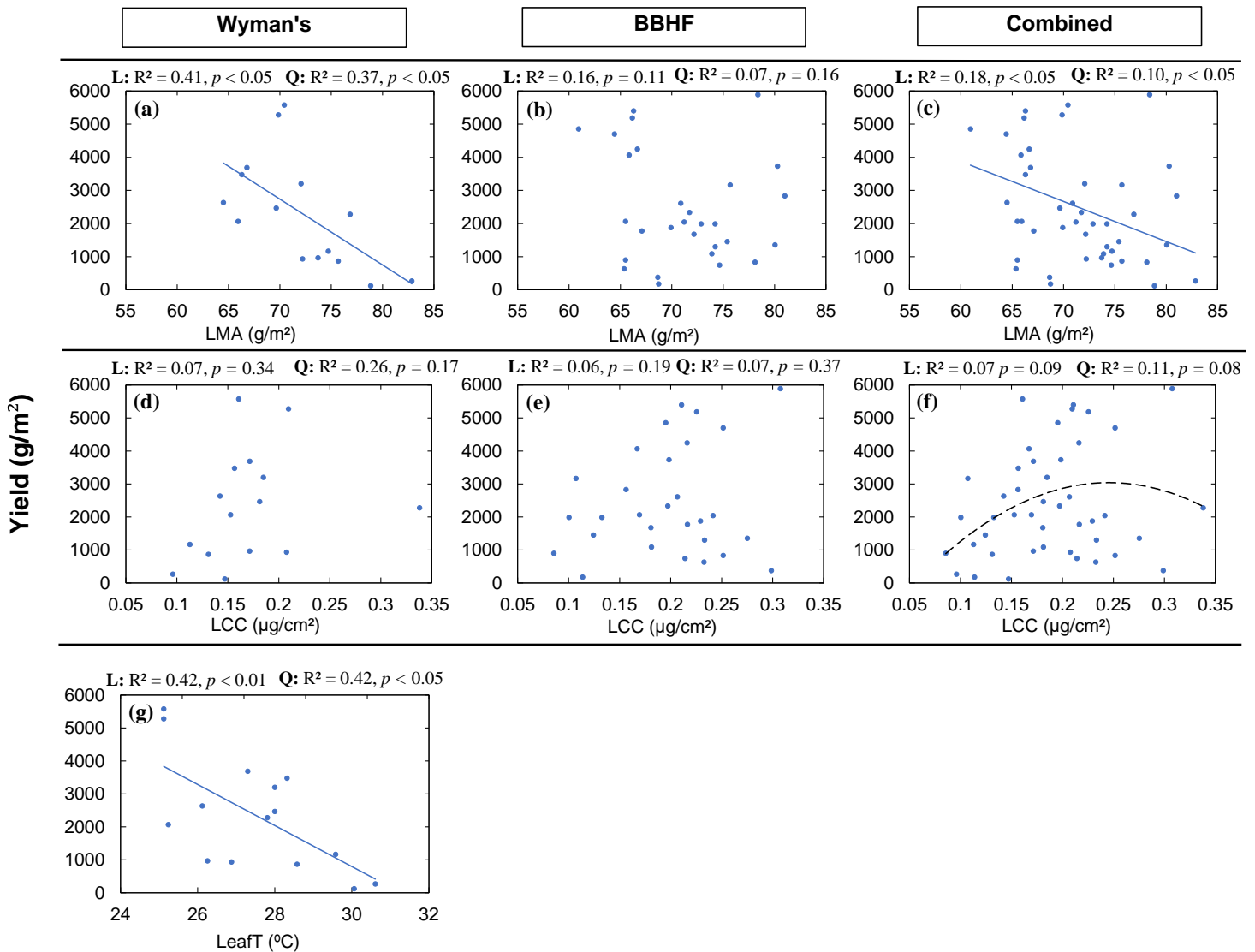


Figure 2.4. Yield (g/m^2) of wild blueberry fields in relation to leaf mass per area (LMA) (a–c); leaf chlorophyll concentration (LCC) (d–f) and leaf temperature (LeafT) (g). The solid lines indicate significant ($p < 0.05$) linear relationships, and black lines indicate significant ($p < 0.05$) quadratic relationships. Dashed lines indicate marginally significant ($p < 0.10$) relationships. Here among all the structural and functional traits, only significant or marginally significant relationships to yield were plotted. The strongest relationship was chosen by the high level of significance (p -value) and higher coefficient of determination (R^2). For definitions of trait abbreviations, please see Table 2.1.

We found relationships of the average number of berries (NBS) with StemL, LCC, LMA, LAPS, and SoilT, but they were not consistent within and across fields (Figures 2.3 and 2.5). We found a marginally significant linear (Figure 2.5a: $R^2 = 0.21$, $p = 0.08$) and quadratic (Figure 2.5a: $R^2 = 0.33$, $p = 0.08$) relationship between NBS and StemL among the genotypes in the Wyman's field. Significant relationships were absent for the BBHF field and when the two fields were combined. We did not find any significant linear or quadratic relationships between LCC and NBS for Wyman's wild blueberry field (Figure 2.5d) but found significant positive linear relationships for the BBHF field (Figure 2.5e: $R^2 = 0.17$, $p < 0.05$) and combined data (Figure 2.5f: $R^2 = 0.17$, $p < 0.05$). For the combined data, we also found a significant quadratic relationship (Figure 2.5f: $R^2 = 0.21$, $p < 0.01$), but for the BBHF field, the quadratic relationship was borderline significant (Figure 2.5e: $R^2 = 0.18$, $p = 0.07$). We found significant negative linear relationships between NBS and LMA for Wyman's field (Figure 2.5g: $R^2 = 0.43$, $p < 0.01$) and the combining data for the two fields (Figure 2.5i: $R^2 = 0.09$, $p < 0.05$), but no significant relationship for the BBHF field. While comparing the leaf structural trait average leaf area per stem (LAPS) with NBS, only a significant quadratic relationship was found for the BBHF field (Figure 2.5k: $R^2 = 0.20$, $p < 0.05$). We also found significant linear (Figure 2.5m: $R^2 = 0.35$, $p < 0.05$) and quadratic relationships (Figure 2.5m: $R^2 = 0.33$, $p < 0.05$) between soil temperature (SoilT) and NBS; however, we found a higher coefficient of determination (R^2) associated with the linear relationship.

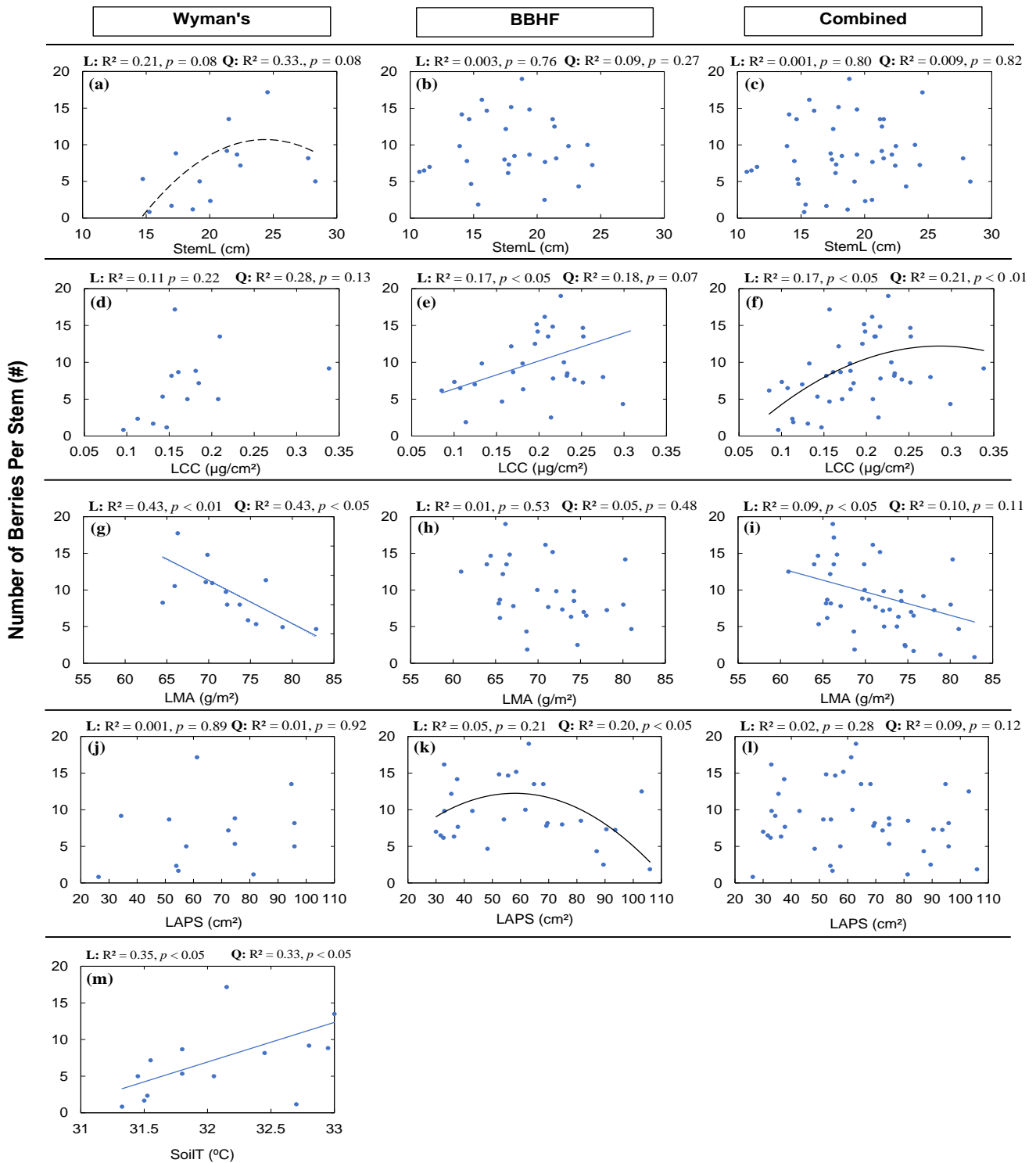


Figure 2.5. Average number of berries per stem (NBS) in wild blueberry fields in relation to average stem length (StemL) (a–c); average leaf chlorophyll concentration (LCC) (d–f); average leaf mass per area (LMA) (g–i); average leaf area per stem (LAPS) (j–l); and average soil temperature (SoilT) (m). The solid blue lines indicate significant ($p < 0.05$) linear relationships, and black lines indicate significant ($p < 0.05$) quadratic relationships. Dashed lines indicate marginally significant ($p < 0.10$) relationships. Here, among all the structural and functional traits, only significant or marginally significant relationships to NBS were plotted. The strongest relationship was selected from the high level of significance (p -value) and higher coefficient of determination (R^2).

The length of a berry cluster in stem (LBC) was significantly related to StemL, LMA, and LeafT. However, relationships within and across fields were not consistent (Figures 2.3 and 2.6). This was also the case for yield (Figure 2.3) and NBS (Figure 2.5). We found a significant positive linear (Figure 2.6a: $R^2 = 0.34$, $p < 0.05$) and marginally significant quadratic (Figure 2.6a: $R^2 = 0.35$, $p = 0.07$) relationship of LBC with the StemL among the genotypes in the Wyman's field, whereas significant relationships were absent for the BBHF field. However, we found significant linear and quadratic relationships (Figure 2.6c) for the combined data from the two fields. The relationships of LMA with the LBC were similar to what we found for NBS (Figure 2.6d–f). We found significant negative linear and quadratic relationships between LBC and LMA for the Wyman's field (Figure 2.6d: $R^2 = 0.58$, $p < 0.001$) and for the combined data of the two fields (Figure 2.6g: $R^2 = 0.15$, $p < 0.05$). No significant relationship was found for the BBHF field. A quadratic relationship was found between LeafT and LBC (Figure 2.6g: $R^2 = 0.38$, $p < 0.05$).

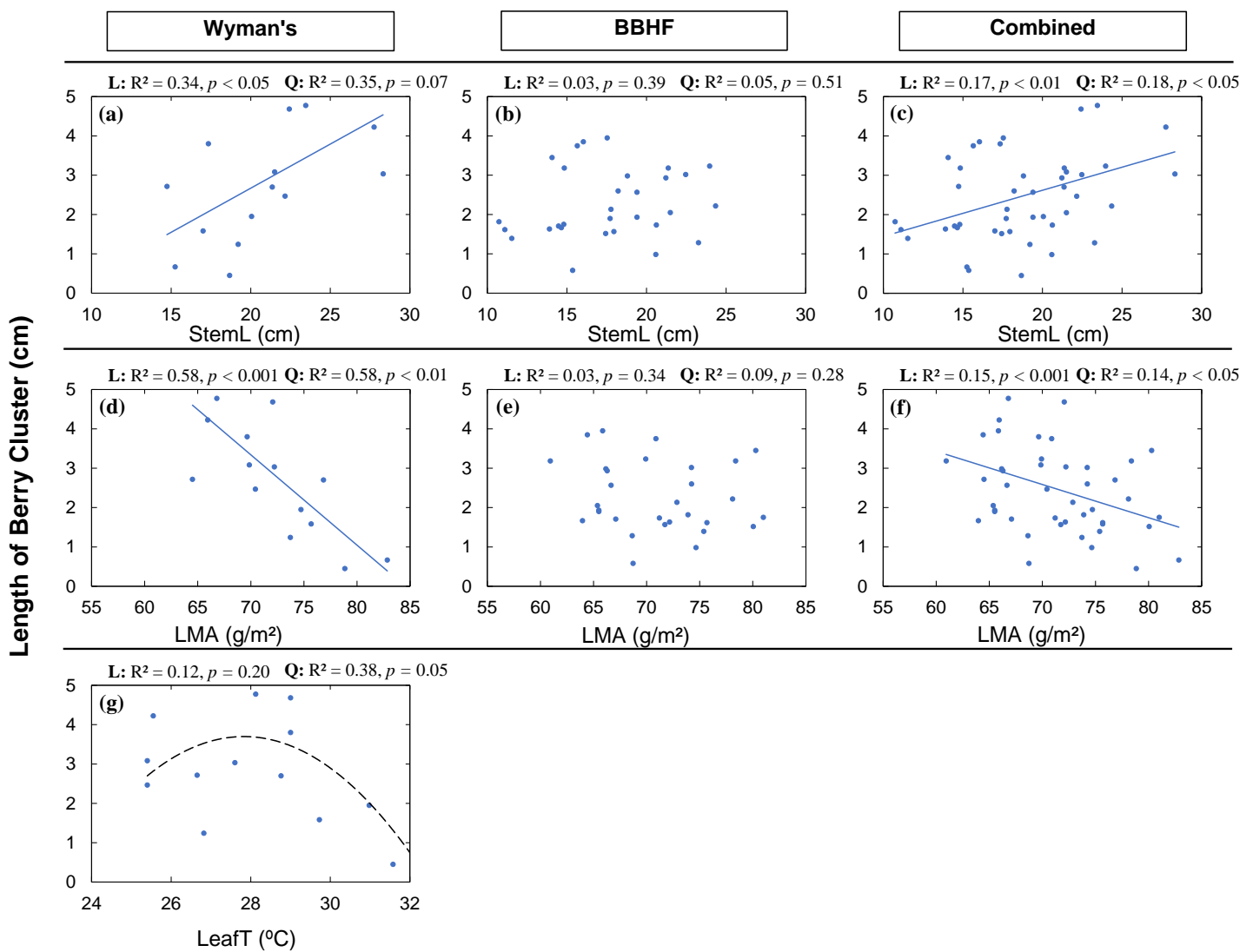


Figure 2.6. Average length of berry cluster (LBC) in wild blueberry fields in relation to average stem length (StemL) (a–c); average leaf mass per area (LMA) (d–f); and average leaf temperature (LeafT) (g). The solid blue lines indicate significant ($p < 0.05$) linear relationships, and black lines indicate significant ($p < 0.05$) quadratic relationships. Dashed lines indicate marginally significant ($p < 0.10$) relationships. Here, among all the structural and functional traits, only significant or marginally significant relationships to LBC were plotted. The strongest relationship was selected from the high level of significance (p -value) and higher coefficient of determination (R^2).

Berry size, indicated by the weight of 100 berries (WT), was found to be related to StemL, StemD, and SWC. These relationships were consistent among fields for StemL and WT (Figure 2.7a–c), but not for StemD (Figure 2.7d–f). We found a significant positive linear (Figure 2.7a: $R^2 = 0.32$, $p < 0.05$) and marginally significant quadratic (Figure 2.7a: $R^2 = 0.32$, $p = 0.10$) relationship for WT with StemL among genotypes in the Wyman's field, whereas for the BBHF field, we found no significant relationships for the BBHF field (Figure 2.7b) or for the combined data (Figure 2.7c). We found significant positive linear and

quadratic relationships (Figure 2.7e) for the BBHF field as well as for the combined data of two fields between StemD and WT, but no correlation was found for the Wyman's field. We also found a significant increase in WT with an increase in SWC (Figure 2.7g: $R^2 = 0.27$, $p < 0.05$).

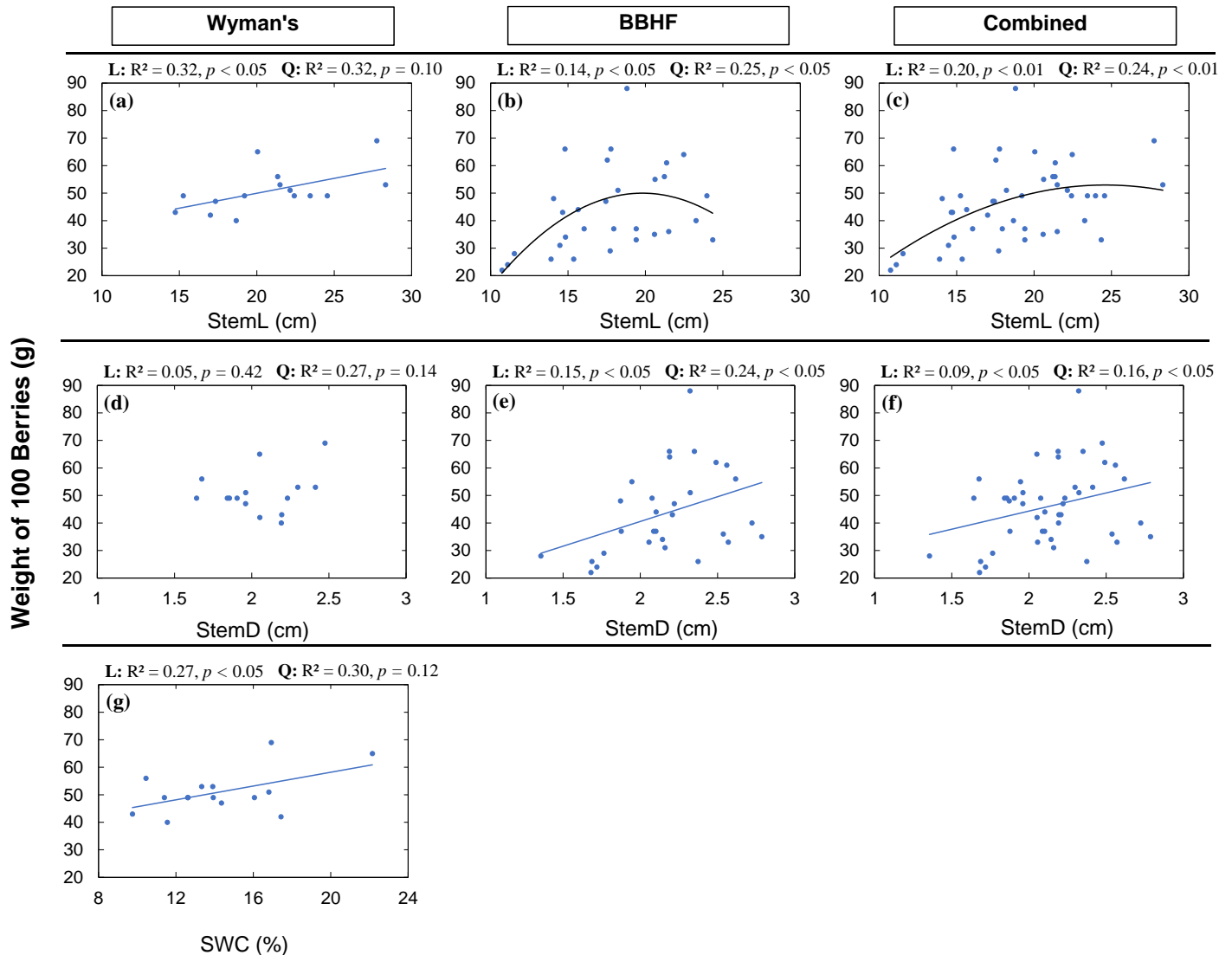


Figure 2.7. Berry size (weight of 100 berries) (WT) in relation to average stem length (StemL) (a–c); average stem diameter (StemD) (d–f); and average soil water content (SWC) (g). The solid blue lines indicate significant ($p < 0.05$) linear relationships, and black lines indicate significant ($p < 0.05$) quadratic relationships. Dashed lines indicate marginally significant ($p < 0.10$) relationships. Here, among all the structural and functional traits, only significant or marginally significant relationships to WT were plotted. The strongest relationship was selected from the high level of significance (p -value) and higher coefficient of determination (R^2).

Combining the data from both fields, we found that the multiple linear regression model tested was statistically significant ($R^2 = 0.345$, $p = 0.01$), and the fitted regression model was $\text{Sqrt yield} = -0.93 \times \text{StemL} + 73.50 \times \text{LCC} - 1.17 \times \text{LMA} - ((\text{LCC} - 0.19) \times (\text{StemL} - 18.76) \times (-30.07))$. LMA ($\beta = -1.17$, $p < 0.05$) and LCC ($\beta = 73.50$, $p < 0.05$) significantly predicted yield (Table 2.3). The effect of LMA was negative on the yield, whereas the effect of LCC was found to be positive. It was also found that StemL did not significantly predict yield ($\beta = -0.93$, $p = 0.057$). When the parameters are centered and scaled, the relative importance of the predictors determining a unit of yield are: -25.3 , 28.2 , -41.4 , and -42.1 for the parameters StemL, LCC, LMA, and StemL \times LCC, respectively. The R^2 of 0.345 of this multiple linear regression was higher than those of single traits (Figure 2.4).

Table 2.3. Multiple linear regression analysis to predict yield (g/m^2) using several functional traits stem length (StemL), leaf mass area (LMA), and leaf chlorophyll concentration (LCC) for the two studied fields combined. The best model, chosen after Lasso variable selection (least absolute shrinkage and selection operator) had an AICc of 382.1 and a coefficient of determination of $R^2 = 0.345$. None of the predictors were correlated ($p > 0.05$), and the predictor distributions were not significantly different than a Normal distribution.

Model Parameters	Parameter Values ¹	Standard Error	Chi-Square	<i>p</i> Value ²
intercept	135.059	31.429	18.466	<0.0001
StemL	-0.926	0.482	3.627	0.057
LCC	73.496	35.391	4.313	0.038
LMA	-1.169	0.411	8.114	0.004
StemL \times LCC	-30.072	12.397	5.884	0.015

¹ values are the constant for the intercept and linear slopes for the predictors. ² bold values are where $p < 0.05$, otherwise $p < 0.1$.

In the PCA analysis of the observed common structural and functional traits of the Wyman's and BBHF field, principal component axis 1 (PC1) explained 28.4% while principal component axis 2 (PC2) explained 21.2% of the total variance (Figure 2.8). The PC1 was positively associated with stem structural traits StemL, StemD, and leaf size-related traits (LeafN and LAPS) and negatively with LMA. The PC2 was positively associated with leaf size-related traits (LeafN and LAPS), and negatively associated with yield and the yield-related traits NBS, LBC, and WT. The first principal component axis (PC1) represented a trade-off between investment in leaf toughness and endurance vs. investment in structural features, while

the second principal component (PC2) represented a trade-off between productivity and investment in leaf structure.

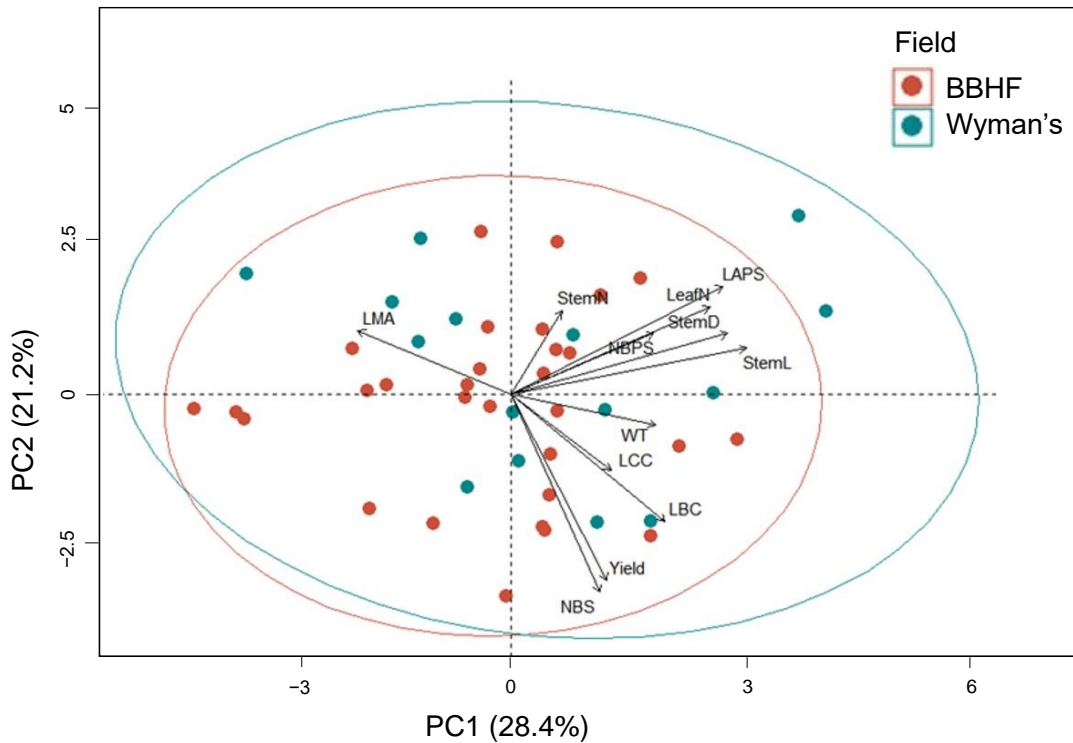


Figure 2.8. Principal Component Analysis (PCA) of mean values of studied common structural and functional traits. The red circle represents BBHF’s field, and the blue circle represents Wyman’s field mean trait values. For definitions of trait abbreviations, please see Table 2.1.

2.5 Discussion

We found high variations in structural, functional, and yield-related traits among genotypes in both wild lowbush blueberry fields. Particularly, the yield varied by 46-fold among genotypes in the Wyman’s field and 36-fold in the BBHF field, which are surprisingly high and suggest the potential for breeding programs and precise management to increase yield. The high variations within fields could be explained by the fact that wild blueberry farms are semi-natural ecosystems with plants naturally growing in the field. Interestingly, no significant differences were found between the two studied fields in traits except StemN, StemL, and LNPS, despite different management practices and environmental conditions. To guide further studies, we found that the optimal sample size to estimate the yield of an entire field is 31

genotypes with a standard error\mean ratio precision of 0.25. Additionally, our results suggest the possible use of leaf mass per area, leaf chlorophyll concentration, stem length, and midday leaf temperature in predicting yield for wild blueberries. An R^2 of 0.345 of the multiple linear regression model suggests important contributions to yield by other factors such as pollination and diseases. Nevertheless, the significant relationships revealed here suggest important biological causal factors in determining yield, which could be used to direct the design of controlled experiments in wild blueberries and other berry crops.

Wild blueberries are known to exhibit a high degree of intra-species diversity in terms of structural and functional traits and yield (Kloet, 1978; Smagula et al., 1997). This is confirmed in our study. Additionally, we did not find any significant differences in any traits except StemN, StemL, and LNPS between the two studied fields (Table 2.2). This pattern agrees with the genetic variation across genotypes and fields, with more than 75 to 92% of the total genetic variation explained by inter-genotype variance within fields and only 8 to 25% explained by between-field variance (Beers et al., 2019; Bell, 2009). Our results suggest overlaps in yield and many functional traits at the two farms despite different management practices. This could be explained by the fact that wild blueberry fields are semi-natural systems with blueberry plants naturally growing in the field and competing with each other. Thus, different genotypes with diverse functional performance are filling different niches of the ecosystem, resulting in a high range of functional traits in different farms. The absence of significant differences between these two farms might also be partially explained by seed dispersal by mammals and birds. Genetic differentiation among blueberry fields does not occur until between-field distances reach 12.5 km (Beers et al., 2019), suggesting that the effective genetic neighborhood is quite large, and gene flow through both pollen and seed dispersal operates at km distances. Therefore, fields close to one another will have genotypes that are genetically similar. Structural and functional features of plants also can be influenced by micro-climatic conditions (Albert et al., 2010). We found a high variation in soil water content in the Wyman's field (Table

2.2), which suggests the heterogeneity of soil moisture retention capability and other influencing environmental factors within the field. High inter-genotype diversity along with the spatial heterogeneity of the micro-climatic conditions might together shape the significant variation of structural and functional traits in wild blueberries (Albert et al., 2010).

We found that with an increase of LMA, the yield-related traits indicated by Yield, NBS, and LBC decreased. A leaf with a high leaf mass per area (LMA) has more fiber content and mass density resulted from the high N accumulation in the cell wall (Onoda et al., 2011; Witkowski & Lamont, 1991). This type of investment in leaf structure increases plant endurance and resistance to environmental stress (Zhang et al., 2017). However, high N concentrations in the cell wall reduce N concentrations in the photosynthetic machinery, reducing the overall photosynthetic performance (Onoda et al., 2011; Witkowski & Lamont, 1991). Moreover, thicker cell walls increase leaf endurance but increase CO₂ diffusion resistance to chloroplasts, resulting in reduced photosynthetic capacity (Niinemets, 1999; Reich et al., 1997). A higher LMA is linked to better survival but slower growth and yield performance; thus, it reduces overall yield (Rüger et al., 2012), whereas a lower LMA results in better resource acquisition and usage efficiency, producing faster growth and higher yield (Lambers & Poorter, 2004). Our finding in the regression analysis is also supported by our PCA analysis, where PC1 was shown to be positively correlated with stem structural features and leaf size-related features and negatively correlated with LMA. The first axis (PC1) indicated a trade-off in investment between leaf toughness and durability vs. structural features related to resource acquisition for photosynthesis, whereas the second axis (PC2) represented a trade-off between productivity and leaf structure. This investment in leaves vs. reproduction tradeoff is also supported by the finding that the removal of flowers of wild blueberry plants enhanced earlier leaf production and higher leaf production rates (Bajcz & Drummond, 2017).

A positive link of yield with CO₂ fixation rate, LCC, and stomatal conductance and a negative correlation with LCC has been found in wheat (Silva-Pérez et al., 2020). In general, LCC correlates positively

with yield performance of crops as it reflects photosynthetic capacity (Ghimire et al., 2015). We also found a positive impact of LCC on yield (Figure 2.4f: borderline significance) and yield-related traits like NBS (Figure 2.5e,f). Chlorophyll production in leaves is dependent on soil nitrogen availability as well as the effectiveness of plants to uptake nitrogen from soil (Percival & Sanderson, 2004). The significant variation of LCC among wild blueberry genotypes and between fields might be related to the variability of soil nitrogen availability as well as variability in the effectiveness to uptake nitrogen from soils due to distinct genotypic features or a combination of both.

We found quadratic relationships between yield-related traits and StemL, suggesting that there is an optimum height for maximum wild blueberry yields. Plants with a higher stem length and larger diameter normally have bigger vessels, which can lead to higher hydraulic conductivity (Sperry et al., 2006), facilitating higher stomatal conductance and photosynthetic carbon gain (Santiago et al., 2004). Higher hydraulic conductivity and photosynthesis are also related to higher plant growth rates (Brodrribb et al., 2007), ultimately impacting yield performance. This explains positive associations between yield-related traits and stem structural traits, which need further physiological studies to confirm. It was previously observed in wild blueberries that greater stem length is related to higher fruit biomass (Fournier et al., 2020). Additionally, genotypes with a higher StemL might be benefitted in terms of successful pollination by bees, which is one of the major yield determining factors of wild blueberries (Yarborough, 2004b). However, as length increases, the danger of xylem cavitation increases, making plants more susceptible to drought (McDowell et al., 2002; Phillips et al., 2010). Additionally, taller wild blueberry plants might be more prone to winter wind damage, which could result in reduced yield.

We found a negative association between the LeafT with Yield (Figure 2.4g) and a quadratic association with the yield-related trait LBC (Figure 2.6g). LeafT is a good indicator of leaf transpiration and water status (Gonzalez-Dugo et al., 2014). There is a direct correlation between leaf temperature and plant water status. When water is limited, the plant reduces its transpiration rate, resulting in higher leaf

temperatures than non-stressed well-watered plants (Gonzalez-Dugo et al., 2014). We did not find any significant relationship between yield and soil water content, but we found a significant positive association between WT and soil water content (Figure 2.7g). A reason for the positive association might be that the weight of berries mostly consists of water; thus, a higher soil water content might be a driver of berry weight.

Between the two fields, we found that plants in Wyman's field showed significantly higher investment in vegetative features compared to the genotypes in BBHF, as we can see significantly higher numbers of stems per quadrat, stem length, and leaf number per stem. However, the difference in yield-related traits between the two studied fields was not statistically significant. Notably, winter damages were found in the fields in 2019, and Wyman's field with taller stems (higher stem length) may experience higher winter damages and more reduction in yield. Overall, although the difference in management practices and environmental conditions between these two farms resulted in differences in vegetative features, it seems they had limited effects on yield, at least in 2019.

2.6 Conclusions

Our study confirms the presence of high levels of variation among wild blueberry genotypes in structural, functional, and yield-related traits. The negative association of leaf mass per area (LMA) with yield-related traits suggests that there is a tradeoff between maintenance traits and yield performance traits, which is to be expected based on cost/benefit balance theory. This interesting tradeoff could exist in other berry crops as well, which deserves further investigation. Meanwhile, although genotypes with a high LMA showed lower yield, they may be more drought resistant and produce a higher yield in drought years. As drought is frequent in wild blueberry fields (Barai et al., 2021), and future warming will decrease the relative humidity and enhance drought effects (Chen et al., 2022; Tasnim et al., 2021), the functional diversity in this semi-natural system could be important in maintaining the stability of wild blueberry

production under increasing climate variability. Overall, our findings imply that a number of leaf and stem functional traits are linked to yield-related traits, and thus these traits can be used to predict wild blueberry productivity to facilitate precise management. We also identified that 25 cm was the optimal stem length for maximizing yield. Our findings are useful for growers and breeders in selecting superior yielding genotypes based on structural and functional features. Increased high-yielding blueberry genotypes planted in between inter-genotype unoccupied spaces in existing fields, as well as replacing less productive genotypes with high-yielding genotypes, would enhance crop production.

CHAPTER 3: IS DROUGHT INCREASING IN MAINE AND HURTING WILD BLUEBERRY PRODUCTION?

3.1 Abstract

A few severe drought events occurred in the Northeast (NE) USA in recent decades, and caused significant economic losses, but the temporal pattern of drought incidents and their impacts on agricultural systems have not been well-assessed. Here, we analyzed historical changes and patterns of drought using a drought index [Standardized Precipitation-Evapotranspiration Index, (SPEI)], and assessed drought impacts on remotely sensed vegetation indices [Enhanced Vegetation Index (EVI) and Normalized Difference Vegetation Index (NDVI)] and production (yield) of the wild blueberry fields in Maine, USA. We also analyzed the impact of short and long-term water conditions of the growing season on the wild blueberry vegetation condition and production. No significant changes in SPEI were found in the past 71 years, despite a significant warming pattern. Also, there was a significant relationship between the relatively long-term SPEI and vegetation indices (EVI and NDVI), but not the short-term SPEI. This suggests that the crop health is probably determined by water conditions in a relatively long term. Neither SPEI nor vegetation indices (EVI & NDVI) are good predictors of wild blueberry yield, possibly because wild blueberry yield does not only depend on climate variables and crop vigor, but also other more important variables such as pollination and fruit-set. We also compared an irrigated and a non-irrigated wild blueberry field at the same location (Deblois, Maine) where we found that irrigation decoupled the relationship between SPEI and NDVI or EVI.

3.2. Introduction

Elevated atmospheric temperatures, increased rainfall variabilities, and more frequent extreme drought events associated with anthropogenic climate change have been significantly damaging agricultural systems and crop production globally (Lesk et al., 2016; Wilhelmi & Wilhite, 2002).

Additionally, local and microclimate changes could be more intense and significantly different compared to the reported average global or regional climate changes in terms of temperatures and precipitation (Fernandez et al., 2020). For instance, a recent study on wild lowbush blueberry crops in Downeast, Maine, USA has revealed that summer temperatures of wild blueberry fields have been increasing significantly faster in the past forty years compared to that of the region (the state of Maine, Northeast (NE), USA) (Tasnim et al., 2021). Such higher increasing temperature patterns in agricultural lands will exacerbate the impacts of drought events due to increased water loss (Carrão et al., 2018; Dai, 2013). Severe drought incidents have been reported in recent decades in NE USA and caused significant economic losses (Sweet et al., 2017; Wolfe et al., 2018). However, the historical trends of drought and their impacts on agricultural systems in this region have not been carefully assessed.

The wild blueberry (*Vaccinium angustifolium* Aiton) is one of the culturally and economically valuable crops in NE USA, which has been growing naturally for hundreds of years at the coastal barrens of the state of Maine in the USA, Atlantic Canada, and the state of Quebec in Canada. It is quite a unique agricultural system as ~1,500 genetically distinct wild blueberry plants can be found in a large field (~ 5 ha.) (Yarborough, 2012). This crop grows in a two-year production cycle where the stems, leaves, and buds develop during the first year, referred to as a prune year, and those plants bloom and produce fruits during the second year, referred to as a crop year (Yarborough, 2012). After harvesting the berries from the end of July to early August in a crop year, plants are pruned to the ground by mowing or burning and the crop cycle starts again the following prune year. It is still unknown how this unique wild agricultural system will respond to the unprecedented changes in rainfall patterns and decreasing soil moisture in this region (Dai, 2013; Tasnim et al., 2021). In fact, summer temperatures and hence potential evapotranspiration of the wild blueberry fields at Downeast, Maine have been increasing significantly in the past decades (Tasnim et al., 2021). Yet, we do not know whether wild blueberry fields experienced drought stress over the years not to mention we do not have any scientific evidence of how this crop has

been responding to drought incidents over the years. Although some controlled drought experiments revealed that wild blueberry plants are drought tolerant based on one growing cycle (2 years) (Glass, 2001; Murray et al., 2002), we still do not know whether drought has short-term and long-term effects on the vigor and production of this crop. Hence, the historical drought patterns that the wild blueberry barrens have been experiencing and their impacts on crop vigor and production need to be analyzed to guide management practices in the future.

Drought has been a great threat to the agricultural systems worldwide, which has been extensively studied in different regions on varieties of crops (Illston & Basara, 2003; Potop et al., 2012). Droughts in agricultural lands are related to a lack of precipitation and inadequate water supply to crops. In order to analyze drought severity, several meteorological drought indices have been developed based on different combinations of precipitation, temperature, soil moisture availability, and vegetation conditions. Widely used drought indices include Palmer Drought Severity Index (PDSI) (Palmer, 1965), Standardized Precipitation Index (SPI) (McKee et al., 1993), Standardized Precipitation-Evapotranspiration Index (SPEI) (Vicente-Serrano et al., 2010) etc. Here, in order to analyze historical drought patterns for wild blueberry fields in Maine, we adopted SPEI over other drought indices as it is determined based on precipitation, temperature, and potential evapotranspiration (Vicente-Serrano et al., 2010). Moreover, SPEI would be the most useful index to determine the water conditions (dry/wet) and drought severity of an agricultural system during both short period and long period (Vicente-Serrano et al., 2010; Zarei & Moghimi, 2019). This is because SPEI's multi-scalar character enables it to detect, monitor, and analyze droughts more effectively as it can quantify the water conditions (dry/wet) and drought severity according to its duration and intensity (Vicente-Serrano et al., 2010). The SPEI also allows the comparison of drought severity through time and space since it can be calculated over a wide range of climates.

Besides determining the historical drought patterns for the wild blueberry barrens, we also assessed the impacts of drought on the crop vigor and production of wild blueberries over the years. In

order to determine the crop vigor, which could be indicated by their greenness and biomass, widely used remotely sensed vegetation indices such as - Normalized Difference Vegetation Index (NDVI) and Enhanced Vegetation Index (EVI) were used to analyze the vegetation condition of wild blueberry fields and their response to historical drought incidents. NDVI and EVI are widely used to assess vegetation health and biomass production as they represent a composite property of canopy cover, canopy structure, canopy greenness, leaf area, and chlorophyll content (Keersmaecker et al., 2017; Glenn et al., 2008; Pettorelli et al., 2005; Son et al., 2014; Tasnim et al., 2021). Moreover, we analyzed the effects of short and long-term water conditions (or water deficits; indicated by SPEI) on the wild blueberry crop vegetation status and production in Maine, and the impacts of irrigation in alleviating drought effects and securing production. Our study will provide a complete understanding of how this crop has been affected by frequent erratic changes in rainfall and drought.

Therefore, the objectives of this study were to:

- 1)** Test whether drought incidents and severity were increasing in past 71 years by analyzing the historical changes in temperature, precipitation, drought index (SPEI), as well as changing pattern of EVI and fruit production of wild blueberry fields in the past two decades in the study sites of the major wild blueberry production region of Maine, USA.
- 2)** Determine the impacts of drought on the vegetation condition and production of the wild blueberry crops in Maine by establishing relationships among the drought index (SPEI), vegetation indices (NDVI and EVI), and yield.
- 3)** Test whether irrigation alleviated the impacts of drought on the vegetation health and production of the wild blueberry crops in Maine by comparing nearby irrigated and non-irrigated fields.

3.3. Materials and Methods

3.3.1. Study Area

We analyzed the wild blueberry fields in Maine, USA as a whole, and two specific fields with a good record of yield data (Figure 3.1). The wild blueberry fields in the major production region are located in the Washington and Hancock counties of Maine (referred to as “Maine WB Fields”, Figure 3.1 a & b). The two specific fields selected were the Airport wild blueberry field at Deblois, Maine (referred to as "Airport", Figure 3.1c), and Baxter wild blueberry field at Deblois, Maine (referred to as "Baxter", Figure 3.1c). The soil of wild blueberry fields in Maine is well-drained sandy loam acidic soil (Drummond et al., 2009). The studied region has a four-season climate with an average annual minimum temperature of -10.6°C and a maximum temperature of 24.2°C , and monthly average precipitation as low as 85.1 mm and as high as 136.4 mm (Arguez et al., 2010).

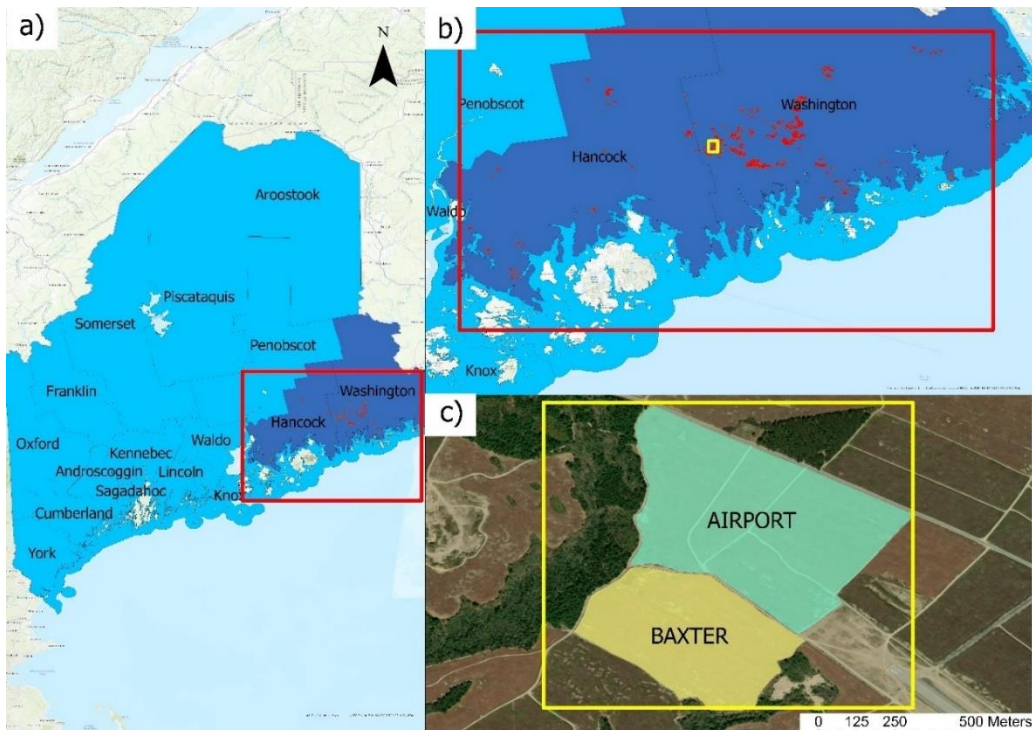


Figure 3.1. Location of the study sites: (a) A map of the state of Maine (light blue color), USA showing the location of major wild blueberry production region (Washington and Hancock County) in dark blue color; (b) A map of the major wild blueberry (WB) production region in Maine showing 89 wild-blueberry fields in red polygons for this study with an area of 0.06 km^2 ($250\text{m} \times 250\text{m}$) or larger; (c) Airport and Baxter wild blueberry field of Wyman’s in Deblois, Washington county, Maine, USA.

3.3.1.1 Wild-Blueberry Fields in Major Wild-Blueberry Production Region of Maine, USA

Washington and Hancock counties together are the largest producer (~90%) of wild blueberries in Maine (“Maine Wild Blueberry Production Statistics - Cooperative Extension,” n.d.). In this study, a total of 89 wild blueberry fields were considered, among which 69 fields were in Washington County, and 20 fields were in Hancock County (Figure 3.1 a & b). Among all the wild blueberry fields of that region, 89 fields with 0.06 km² (250 m*250 m) or larger areas were selected for this study. The land area threshold of 0.06 km² was used because the remote sensing data products that we used had a spatial resolution of 250 m. The selected wild blueberry fields are represented in red color in Figure 3.1 a & b.

3.3.1.2 Airport and Baxter Wild-Blueberry Fields in Deblois, Maine

The Airport and Baxter fields are adjacent to each other (Figure 3.1c). These two fields are part of commercial blueberry fields owned by Jasper Wyman and Son in Deblois (Longitude: -68.0001° N, Latitude: 44.7350° W), Washington county, Maine, USA. In terms of agricultural management, these two fields are historically treated equally except for irrigation. The Airport field is irrigated during the growing season whereas the Baxter field is non-irrigated. During the growing season from May to September, the airport field was irrigated when needed with Nelson Full-Circle Impact sprinklers (Walla Walla, WA, USA) uniformly across the field. The irrigation system was set to ensure a 1 inch of water supply per acre every 3 to 4 days by compensating for natural precipitation. The area of the Airport field is 77 acres (0.31 km²), and the Baxter field is 39 acres (0.16 km²). These two fields were selected to understand and differentiate the effectiveness of current uniform irrigation practices in a single production region with same climatic conditions.

3.3.2 Data Acquisition and Methodology

A Keyhole Markup language Zipped (KMZ) file locating the wild blueberry fields of Maine was produced based on a field survey carried out by the University of Maine Cooperative Extension. The polygons of the 89 wild blueberry fields (area > 0.06 km² in the major wild blueberry production region,

Maine (Figure 3.1 a & b) were acquired from the KMZ file. The Airport and Baxter field polygons were acquired from a KMZ file based on a field survey by Jasper Wyman & Son, Deblois, Maine.

In this study, SPEI was used as a drought index, which is calculated based on precipitation, temperature, and potential evapotranspiration data. The SPEI was chosen over another popular drought index the PDSI, because PDSI has a fixed temporal scale (between 9-12 months) which prevents understanding drought severity on different temporal scales (Guttman, 1998). In addition, SPEI was chosen over widely used the SPI because SPI only considers precipitation data and does not include air temperature and evapotranspiration data, which could also significantly influence understanding drought impacts on agriculture (Vicente-Serrano et al., 2010). The SPEI's multi-scalar character enables it to detect, monitor, and analyze droughts more effectively as it can quantify the drought severity according to its duration and intensity (Vicente-Serrano et al., 2010). The SPEI allows the comparison of drought severity through time and space since it can be calculated over a wide range of climates. In this study, the SPEI data was collected from the readily available open access database "SPEI Global Drought Monitor" (<https://spei.csic.es/map/maps.html>; accessed on 10 July 2020) in netcdf format. The netcdf files containing the SPEI data were transferred to ArcGIS Pro Version 2.7 (ESRI, Redlands, California, USA) to acquire SPEI data for the study sites (Airport, Baxter and Major wild blueberry region of Maine) over 71 years from 1950 to 2020 using zonal statistics tool in ArcGIS. The SPEI data were acquired on different temporal scales ranging from 1 month (SPEI_1) to 48 months (SPEI_48). These data were provided on a per-pixel basis at 4 km spatial resolution. SPEI (SPEI_6 of September) of only the growing season (April-September) was considered in this study. SPEI_6 of September represents the water conditions of a growing season (April-September). To understand the long-term (multi-year) impact of water conditions on vegetation indices (EVI and NDVI) and yield of wild blueberry crops, average SPEI (SPEI_6 of September) of two, three and four consecutive years were also calculated. A positive SPEI value represents a wet condition whereas a negative SPEI value indicates a dry condition.

The dataset of climate variables such as, total precipitation and mean temperature during the growing season (May to September) over 71 years from 1950 to 2020 for the study sites (Airport/Baxter and Major wild blueberry region of Maine) were acquired from the online tool “Climate Engine” (<https://clim-engine.appspot.com/climateEngine> accessed on 17 July 2021) of Desert Research Institute, University of California, USA. Here, total precipitation refers to an average of monthly total precipitation (mm) and mean temperature refers to an average of monthly mean air temperature at 2 m from the ground surface for the growing season. The original data sources for the climate variables were obtained from the AN81m dataset of PRISM Climate Group (<https://prism.oregon-state.edu/explorer/> accessed on 17 July 2021). These data were provided on a per-pixel basis at 4 km spatial resolution for the conterminous United States with a temporal resolution of one month (daily mean temperature and total precipitation were averaged monthly). This AN81m dataset is available from January 1895. Extracted data were transferred into Microsoft Excel (Microsoft, Redmond, WA, USA) to calculate the average total precipitation, mean temperature of the summer months (May to September) of each year (Figure 3.3).

In order to quantify vegetation responses to drought, satellite-based remotely sensed EVI and NDVI data for 21 years (2000 to 2020) of the studied wild blueberry fields were acquired from Google Earth Engine. These data were originally obtained from the Moderate Resolution Imaging Spectroradiometer (MODIS) product MOD13Q1 (<https://lpdaac.usgs.gov/products/mod13q1v006>; accessed on 25 July 2021). MOD13Q1 dataset is preprocessed, readily available, and have open access. The MOD13Q1 Version 6 data have a spatial resolution of 250 meters, generated every 16 days. For the development of the MOD13Q1 product, an algorithm was used to select pixels with low clouds, low view angle and maximum index value to obtain the best available pixels over the 16-day-period image acquisitions (Didan, 2015). The MOD13Q1 product has two vegetation layers: NDVI and EVI. The NDVI is the most common one used for characterizing canopy leaf chlorophyll content based on the reflectance contrast between the red and the NIR (near-infrared) wavebands (Lu et al., 2018). However, NDVI has

some limitations such as, (1) it saturates in dense vegetation, (2) it does not consider the canopy background noise, and (3) its rationing properties to eliminate noise (Zhengxing et al., 2003). These limitations were improved in EVI to some extent, and thus EVI has several advantages over NDVI as it has improved sensitivity over high biomass regions (Zhengxing et al., 2003). This dataset is readily available in the Google Earth Engine. Vegetation indices values over the summer months were extracted for the study sites using a java-script based API in the Google Earth Engine (<https://code.earthengine.google.com/> accessed on 25 July 2021) using extraction command “ui.Chart.image.seriesByRegion”. Extracted data were transferred into Microsoft Excel (Microsoft, Redmond, WA, USA) to calculate the average EVI and NDVI of the summer months (May-September) of each year. The historical yield data of Maine was collected from the United States Department of Agriculture (USDA), National Agricultural Statistics Service using a Quick Stats Ad-hoc Query Tool, available at (<https://quickstats.nass.usda.gov/>; accessed on 26 July 2021). Historical yield data of the entire state of Maine (million lb) were available from 1924 to 2020, but the Yield per production area data (lbs./acre) were only available from 2012-2020 (except 2013). It should be noted that, the yield data were considered from all over the state of Maine, where ~90% of yield was typically from the major wild blueberry production region (Washington and Hancock counties). Historical yield data of the Airport and Baxter fields at Deblois, Maine were provided by Jasper Wyman & Son, Maine. Yield (lbs./acre) data for the Airport and Baxter fields were available from 1993-2019 for every alternate year (except 2001 for the Baxter field).

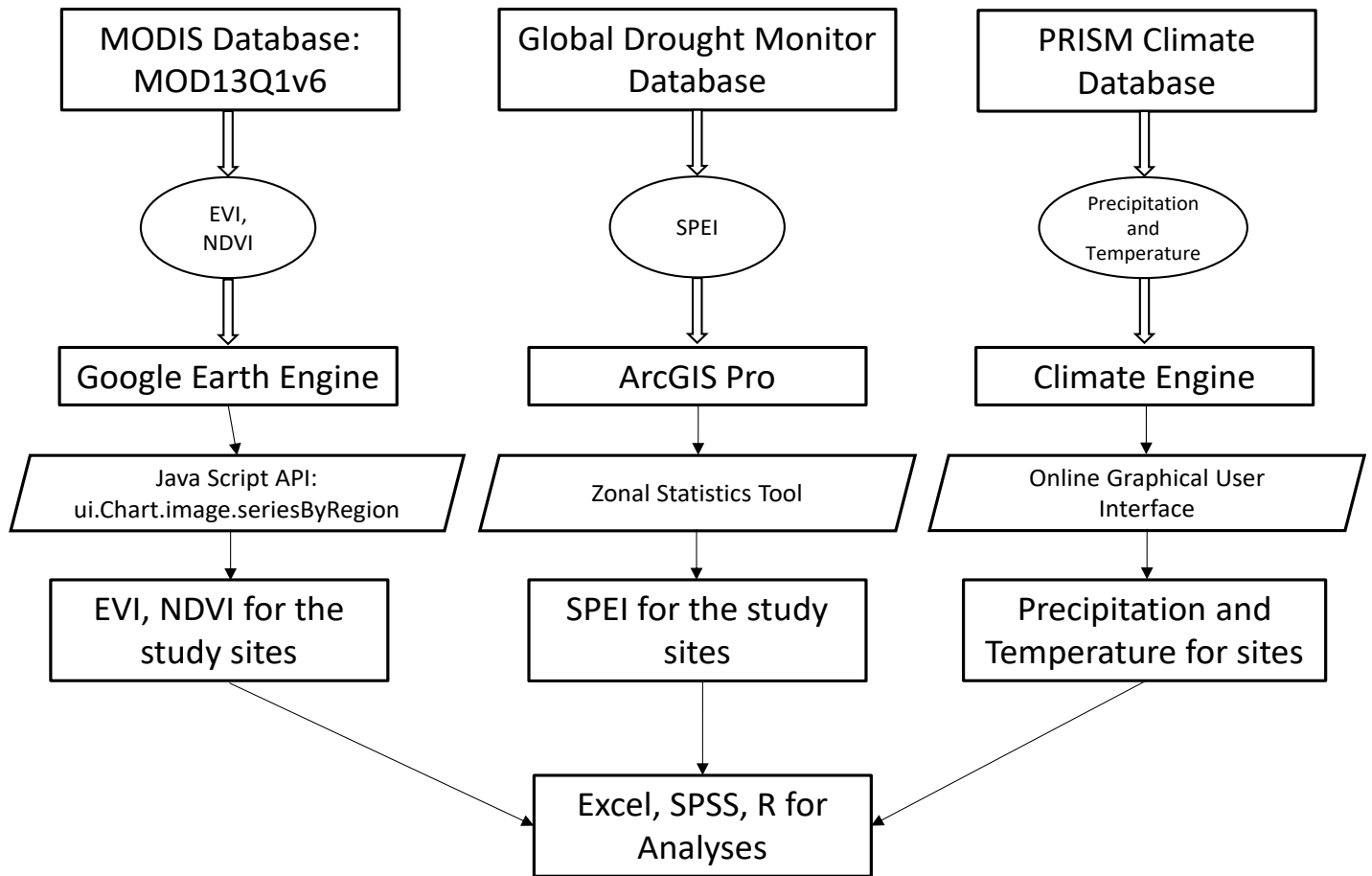


Figure 3.2. A flowchart showing the steps of data acquisition and analysis for this study.

3.3.3 Statistical Analysis

In this study, SPSS V23 (IBM Corp., Armonk, NY, USA), and RStudio software (RStudio, PBC, Vienna, Austria) were used for statistical analysis. Trend analyses of the climate variables (SPEI, Precipitation, Temperature,) over the last 71 years (1950-2020) at the studied wild blueberry fields (Airport/Baxter and Maine) were conducted by Mann–Kendall trend test, Sen’s slope estimator using the XRealStats (Addinsoft, New York, NY, USA) add-on in Microsoft excel. The “pheno” package in RStudio software was used to analyze the forward (UF) and backward (UB) curves of the sequential Mann–Kendall test statistics. Trend analyses of EVI and Yield over the last 21 years (2000-2020) at the studied wild blueberry fields (Airport/Baxter and Maine) were conducted by Mann–Kendall trend test, Sen's slope estimator using the XRealStats tool. To assess the statistical significance of the Mann-Kendall trend

analysis, the significance level (α) was set to 0.05. A Pearson correlation analysis was conducted between different temporal scales of SPEI and EVI and Yield to understand the drought impact on vegetation (EVI) and yield at different temporal scales using SPSS V23. To assess the statistical significance of the Pearson correlation analysis, the significance level (α) was set to 0.05. To understand the short- to long-term (SPEI_1_Year to SPEI_4_Year) effects of drought on EVI, NDVI (average of the growing season: May-September) for the studied wild blueberry fields over 21 years (2000–2020), linear (in the form of $a + bx$) and non-linear (in the form $a + bx + cx^2$) regression analyses were also conducted using SPSS V23. A similar analysis was conducted to understand the short- to long-term impact of water conditions (indicated by SPEI_1_Year to SPEI_4_Year) on yield of the studied wild blueberry fields. We determined the statistical significance of the relationship using the coefficient of determination and its significance (α) at $p < 0.05$.

3.4 Results

3.4.1. Historical Changes in SPEI, Climate Variables, EVI, and Productivity of Wild Blueberry Systems in Maine, USA

During the last 71 years (1950 – 2020), drought index (SPEI, Figure 3.3 a & b) and precipitation (Figure 3.3 c & d) tended to increase marginally (Figure 3.4a-4d; Table 3.1) in the studied wild blueberry fields in Maine (Figure 3.3 a,c & 3.4 a,c) as well as considering two specific fields (Airport/Baxter) at Deblois, Maine (Figure 3.3 b,d & 3.4 b,d). However, the mean atmospheric temperature has been increasing significantly on the wild blueberry fields in Maine overall (Figure 3.3 e & f; Table 3.1), and the two fields in Deblois, ME. These patterns were also supported graphically by the upward UF curve (forward trend) mostly being > 0.0 and UB (backward trend) curve mostly being < 0.0 (Figure 3.4 e & f).



Figure 3.3. Historical (1950 to 2020) pattern in the (a), (b) SPEI_6 of September; (c), (d) Mean precipitation (Average of May–September); (e), (f) average temperature (average of May–September) throughout the major wild blueberry (WB) production region in Maine as well as at the Airport/Baxter wild blueberry fields in Deblois, ME. A positive SPEI value represents a wet condition where a negative SPEI value indicates a dry condition. Here, mean precipitation refers to an average of monthly total precipitation (mm), and mean temperature refers to an average of monthly air temperature at 2 m from the surface for the growing season.

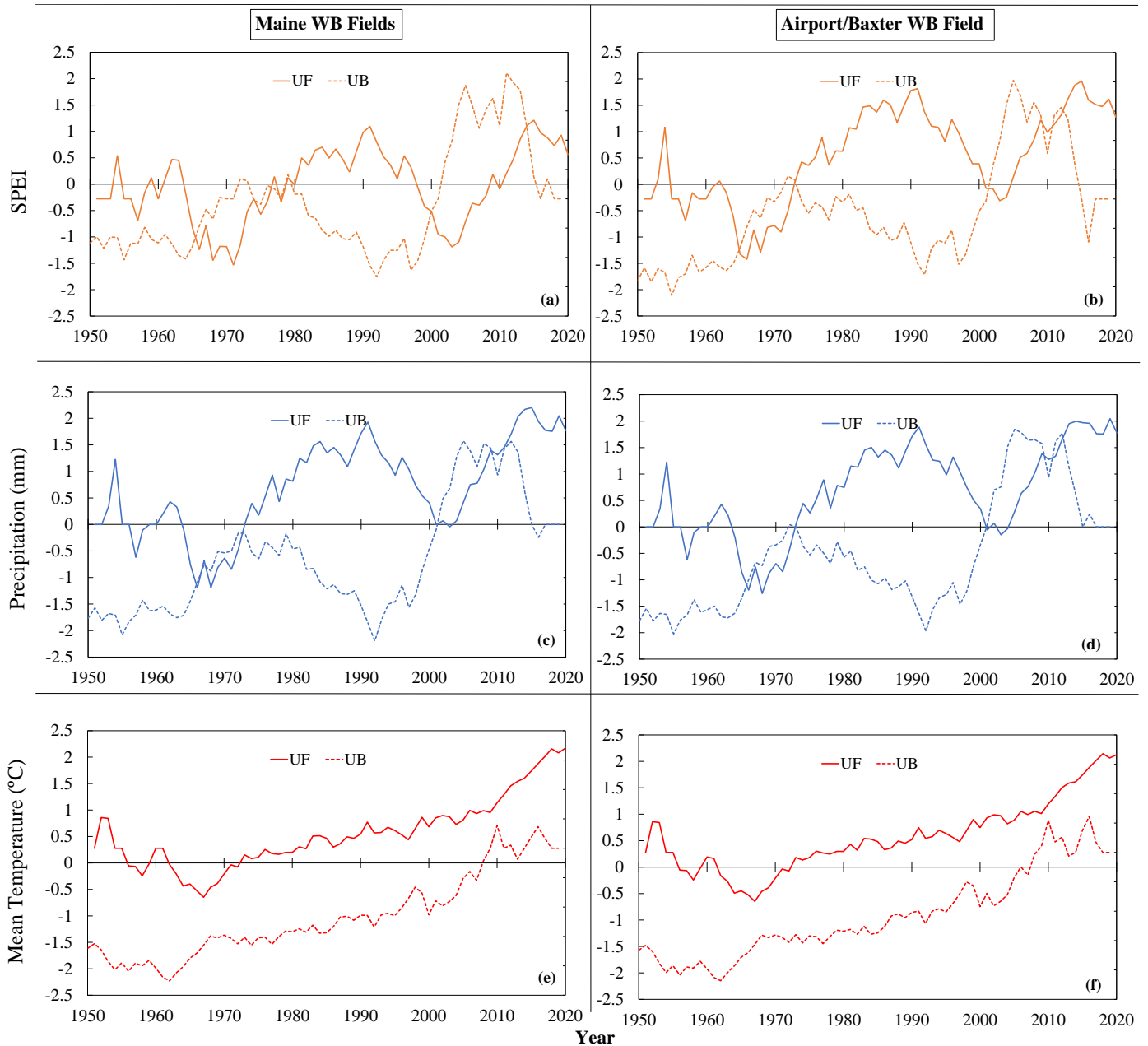


Figure 3.4. Sequential Mann–Kendall test statistics (UF and UB values) calculated from the (a), (b) SPEI₆ of September; (c), (d) Mean precipitation (Average of May–September); (e), (f) average air temperature (average of May–September) throughout the major wild blueberry (WB) production region in Maine as well as at the Airport/Baxter wild blueberry fields in Deblois, ME. Here, the upward UF curve (forward trend) mostly being > 0.0 , UB (backward trend) curve mostly being < 0.0 , and UF and UB being not intersected with each other indicate the significant increasing trends of mean temperature. Whereas the intersections of those curves with 0.0 line as well as with each other represent non-significant changing (increasing/decreasing) trends of SPEI and precipitation.

Table 3.1. Sequential Mann–Kendall trend analysis of Standardized Precipitation-Evapotranspiration Index (SPEI), Precipitation and T_{mean} at different wild-blueberry study zones: Airport/Baxter wild blueberry fields (Deblois, ME), and Maine wild blueberry fields (Washington & Hancock counties, ME). Here, SPEI refers to SPEI_6 of September. It represents the SPEI of the growing season (April-September) and indicates drought severity. T_{mean} represents the average air temperatures during the growing period (May to September).

Mann–Kendall Test	Maine WB Fields			Irrigated/Non-irrigated Airport/Baxter, Deblois, ME		
	SPEI	Precipitation	T_{mean}	SPEI	Precipitation	T_{mean}
Kendall’s Tau	0.062	0.144	0.276	0.114	0.144	0.270
Mann–Kendall Stat (S)	153.000	357.000	687.000	283.000	359.000	671.000
Var (S)	40588.33	40588.33	40588.33	40588.33	40588.33	40588.33
<i>p</i>-value (two-tailed)	0.45	0.07	0.001	0.16	0.07	0.001
Alpha	0.05	0.05	0.05	0.05	0.05	0.05
Trend	Increasing (Non-significant)	Increasing (Non-significant)	Increasing (Significant)	Increasing (Non-significant)	Increasing (Non-significant)	Increasing (Significant)
Sen’s Slope Q	0.005	1.344	0.013	0.008	1.304	0.012

Also, based on the yield data from crop years (every alternative year from 1993-2019), the wild blueberry yield had a marginal increment (non-significant; Table 3.2) in the Airport field (Irrigated) (Figure 3.5a). Similar marginal increments (non-significant, Table 3.2) were also observed in EVI during the growing season (April - September) over the last 21 years (2000-2020) (Figure 3.5d). In contrast, both the yield (Figure 3.5b) and EVI (Figure 3.5e) had significant increments in the Baxter field (non-irrigated; Table 3.2). No significant changes in yield were observed from the studied wild blueberry fields of Maine over the last 21 years (2000-2020) (Figure 3.5c). A significant increase in EVI during the growing season (April - September) was observed over the last 21 years (Figure 3.5f).

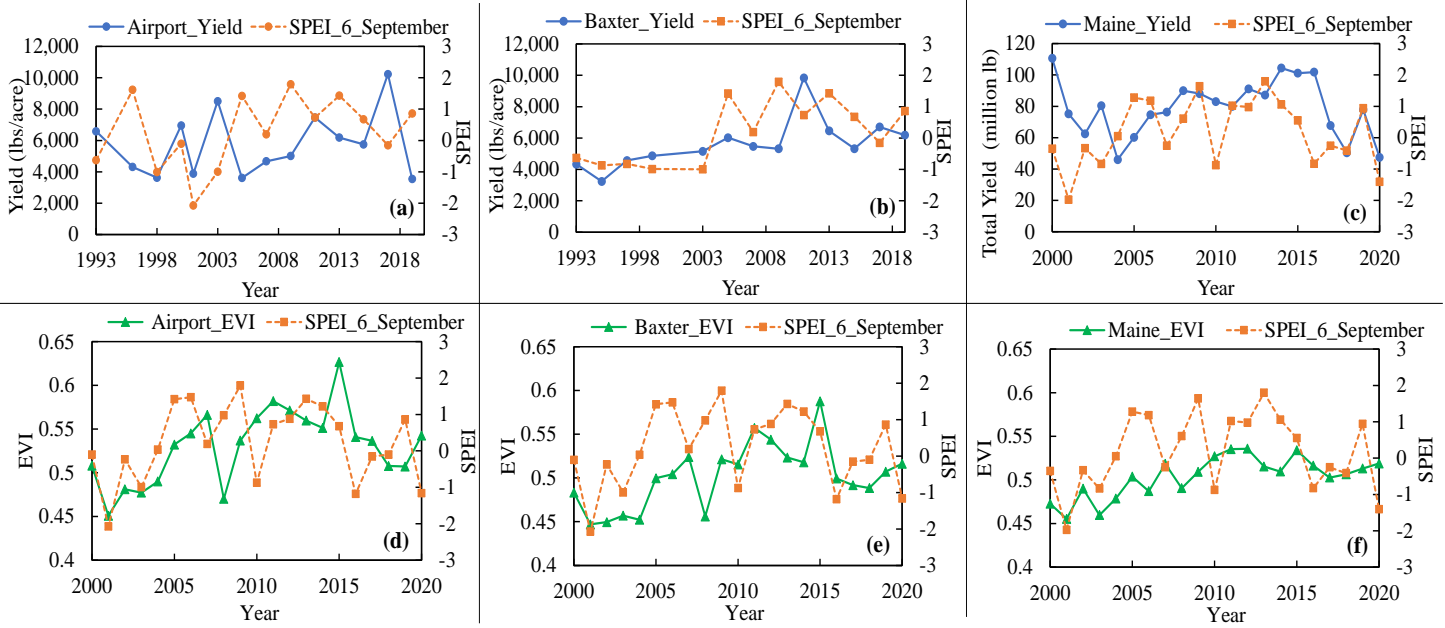


Figure 3.5 (a and d) Historical values of Yield and SPEI (a; 1993-2019), and of EVI and SPEI (d; 2000-2020) for Airport (Irrigated) field, Deblois, ME. (b and e) Historical values of Yield and SPEI (b; 1993-2019), and in EVI and SPEI (e; 2000-2020) for Baxter (Non-Irrigated), Deblois, ME. (c and f) Historical values of Yield and SPEI (c; 2000-2020), and in EVI and SPEI (f; 2000-2020) for major wild blueberry (WB) production region in Maine. Here, orange dashed lines indicate SPEI, blue solid lines indicate yield, green solid lines indicate EVI. Here SPEI refers to SPEI_6 of September. It represents the SPEI of the growing season (April-September) and indicates water conditions. A positive SPEI value represents a wet condition where a negative SPEI value indicates a dry condition.

Table 3.2 Sequential Mann–Kendall trend analysis of Yield and Enhanced Vegetation Index (EVI) at three different wild-blueberry study zones: Airport (Irrigated field, Deblois, ME), Baxter (Non-Irrigated field, Deblois, ME), and Maine wild blueberry fields (Washington county and Hancock County, ME).

Mann–Kendall Test	Airport, Deblois, ME (Irrigated field)		Baxter, Deblois, ME (Non-Irrigated field)		Maine WB fields	
	Yield	EVI	Yield	EVI	Yield	EVI
Kendall’s Tau	0.099	0.257	0.667	0.333	0.057	0.476
Mann–Kendall Stat (S)	9.000	54.000	52.000	70.000	12.000	100.000
Var (S)	333.667	1096.667	268.667	1096.667	1096.667	1096.667
p-value (two-tailed)	0.667	0.110	0.002	0.037	0.740	0.003
Alpha	0.050	0.050	0.050	0.050	0.050	0.050
Trend	Increasing (non-significant)	Increasing (non-significant)	Increasing (Significant)	Increasing (Significant)	Increasing (non-significant)	Increasing (Significant)
Sen’s Slope Q	54.10	0.003	89.91	0.003	0.301	0.003

3.4.2. Relationships between SPEI and vegetation indices in wild blueberry fields of Maine

Based on the relationships of both short-term and long-term average SPEI with EVI and NDVI (Figure 3.6 & 3.7), long-term SPEI showed stronger influence on vegetation indices (EVI in Figure 3.6 & NDVI in Figure 3.7) of wild blueberries compared to the short term SPEI. While analyzing the impact of short-term SPEI (SPEI_1_Year in Figure 3.6-3.7, and SPEI_1 to SPEI_11 in Table S1) on EVI (Figure 3.6 a-c) and NDVI (Figure 3.7 a-c) during the growing season (May-September), no significant relationship was observed for the studied wild blueberry fields in Maine. On the contrary, while observing the impact of long-term SPEI (2 to 4 consecutive years) on both EVI and NDVI of the wild blueberry fields during the growing season, we found both significant linear and quadratic relationships between SPEI and EVI (Figure 3.6 d-l) as well as between SPEI and NDVI (Figure 3.7 d-l). Among the significant linear and quadratic relationships between an average SPEI of 2 consecutive years (SPEI_2 year) and vegetation indices for Airport (Figure 3.6d & 3.7d), Baxter (Figure 3.6e & 3.7e) and studied wild blueberry fields in Maine (Figure 3.6f & 3.7f), the coefficient of determination (R^2) was higher for the quadratic relationships. Moreover,

the strength (R^2 values) of both linear and quadratic relationships was higher while considering more consecutive years such as, SPEI_3 Year (Figure 3.6 g-i & 3.7 g-i) and SPEI_4 Year (Figure 3.6 j-l & 3.7 j-l) compared to the SPEI_2_Year (Figure 3.6 d-f & 3.7 d-f). Although both relationships between SPEI and EVI (Figure 3.6) as well as SPEI and NDVI (Figure 3.7) were significant while considering long-term drought index, the coefficient of determination (R^2) was higher for the relationships between SPEI and EVI compared to the relationships between SPEI and NDVI. Because of the stronger relationship between SPEI and EVI, we further analyzed the impact of short- and long-term SPEI on wild blueberry yield as well as monthly SPEI impacts on EVI and yield during the growing season. Interestingly, while considering the impact of monthly SPEI (different temporal SPEI in Table S1) during the growing season, SPEI of the early season (April-June) showed more impacts on the EVI of wild blueberry fields compared to the SPEI later in the season (July-August).

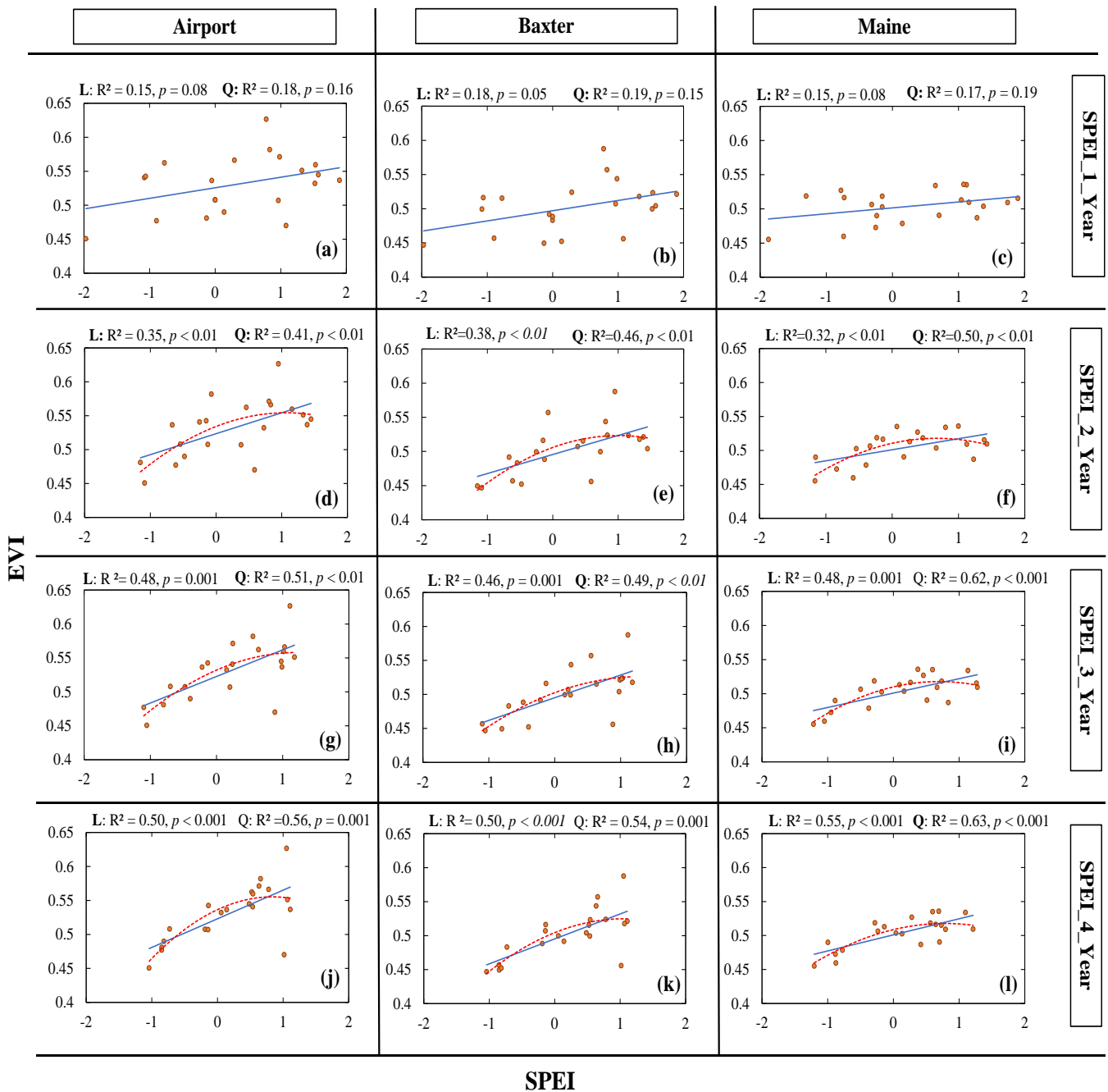


Figure 3.6. Average Enhanced Vegetation Index (EVI) of wild-blueberry fields during growing season (May to September) for three different study zones: Airport (Irrigated), Baxter (Non-irrigated), and major wild blueberry (WB) production region in Maine in relation to (a), (b), (c) SPEI_1_Year ; (d), (e), (f) SPEI_2_Years (average SPEI of two consecutive years); (g), (h), (i) SPEI_3_Years (average SPEI of three consecutive years); (j), (k), (l) SPEI_4_Years (average of SPEI of four consecutive years). Here SPEI refers to SPEI_6 of September and it represents the SPEI (drought severity) of a growing season (April-September). A positive SPEI value represents a wet condition where a negative SPEI value indicates a dry condition. The blue solid lines indicate significant ($p < 0.05$) or marginally significant ($p < 0.10$) linear relationships. The dashed red lines indicate significant ($p < 0.05$) or marginally significant ($p < 0.10$) significant quadratic relationships. The time-period of EVI and SPEI data was from 2000 to 2020.

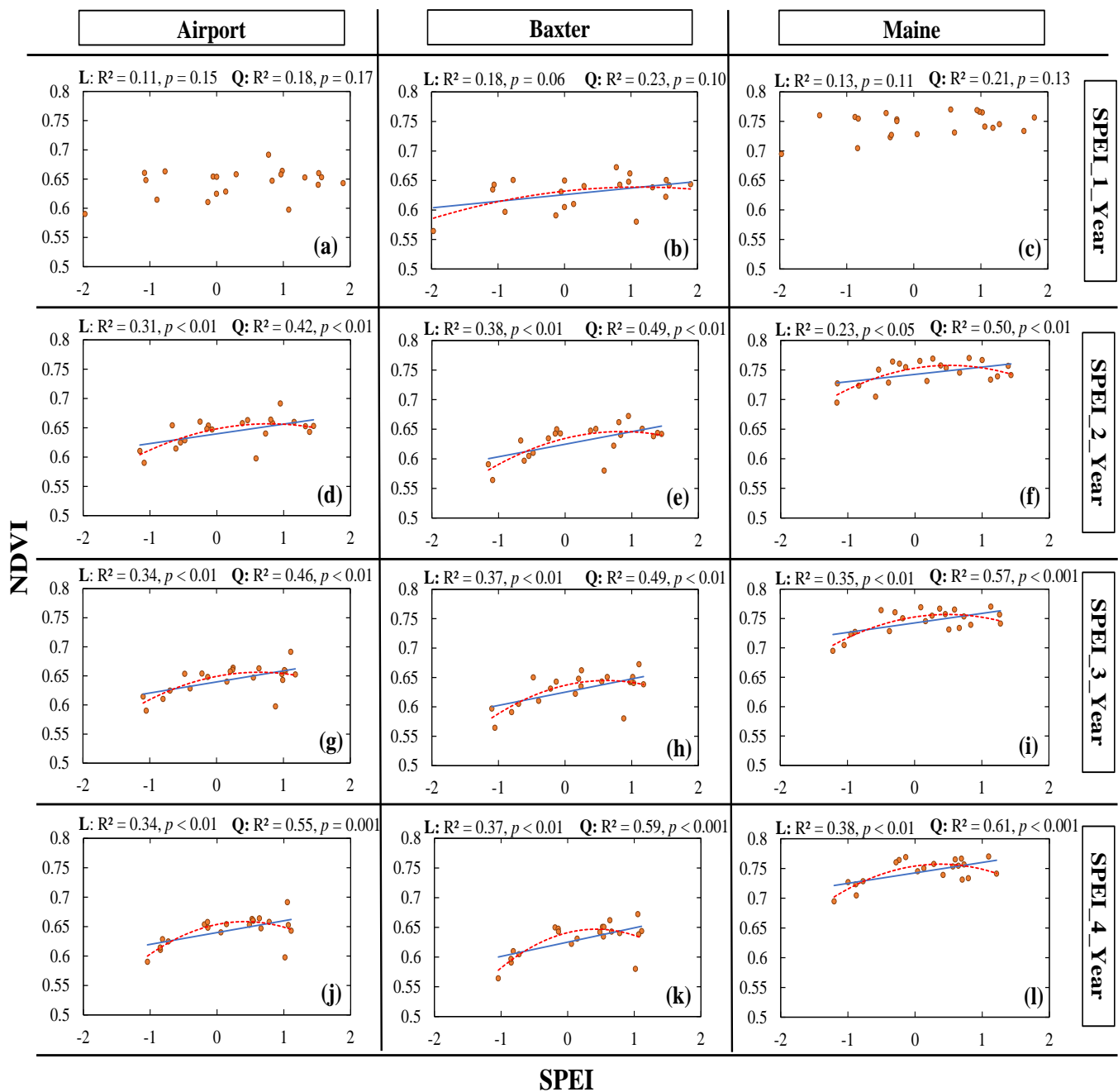


Figure 3.7. Average Normalized difference vegetation index (NDVI) of wild-blueberry fields during the growing season (May to September) for three different study zones: Airport (Irrigated), Baxter (Non-irrigated), and major wild blueberry (WB) production region in Maine in relation to (a), (b), (c) SPEI_1_Year ; (d), (e), (f) SPEI_2_Years (average SPEI of two consecutive years); (g), (h), (i) SPEI_3_Years (average SPEI of three consecutive years); (j), (k), (l) SPEI_4_Years (average SPEI of four consecutive years). Here SPEI refers to SPEI_6 of September and it represents the SPEI (drought severity) of a growing season (April-September). A positive SPEI value represents a wet condition where a negative SPEI value indicates a dry condition. The blue solid lines indicate significant ($p < 0.05$) or marginally significant ($p < 0.10$) linear relationships. The dashed red lines indicate significant ($p < 0.05$) or marginally significant ($p < 0.10$) quadratic relationships. The time-period of EVI and SPEI data was from 2000 to 2020.

3.4.3. Relationships between SPEI and yield of wild blueberry fields in Maine

The impacts of short-term and long-term SPEI on the wild blueberry yield (Figure 3.8 and Table S2) were different from the relationships between SPEI and EVI during the growing season (April-September) (Figure 3.6 and Table S1). A significant and positive linear relationship was found between the short-term SPEI (SPEI_1 Year) and yield for the non-irrigated Baxter field (Figure 3.8b) whereas short-term SPEI (SPEI_1 Year) and yield were negatively correlated (marginally significant, $p = 0.058$) for the irrigated Airport field (Figure 3.8a). For the irrigated field, we observed a decrease in yield with an increase in SPEI. For the wild blueberry fields in Maine as a whole, we found a marginally significant ($p < 0.1$) and positive linear relationship between the short-term drought index (SPEI_1 Year) and wild blueberry yield (Figure 3.8c). We found a significant quadratic relationship between short-term SPEI (SPEI_1 Year) and yield for only the non-irrigated Baxter field (Figure 3.8b) but not for the irrigated Airport field and the studied wild blueberry fields of Maine as a whole. While considering the impact of monthly droughts (different temporal SPEI in Table S2) during the growing season, the correlation between SPEI and yield was significant for the non-irrigated Baxter field whereas it was not significant for the irrigated Airport field.

While analyzing the impact of long-term SPEI (2 to 4 consecutive years) on the yield of irrigated Airport field and non-irrigated Baxter field, no significant linear and quadratic relationships were found (Figure 3.8 d & e; SPEI_3 Year in Figure 3.8 g & h; SPEI_4 Year in Figure 3.8 j & k). However, stronger relationships were observed between the long-term SPEI during the growing season and yield while considering the wild blueberry fields of Maine as a whole (Figure 3.8 i & l). We found significant positive linear and quadratic relationships between the average yield of the wild blueberry fields in Maine and the long-term SPEI (Figure 3.8 i & l) except for the SPEI_2 Year (Figure 3.8f), where the linear relationship was marginally significant ($R^2 = 0.36$, $p = 0.08$) and quadratic relationship was not significant. In fact, the quadratic relationships were stronger between yield and the average SPEI of consecutive 3 and 4 Years (Figure 3.8i & l). Also, while considering cumulative impacts for more consecutive years, both the linear

and quadratic relationships were observed to be stronger for the wild blueberry fields in Maine as a whole (Figure 3.8 f, i & l).

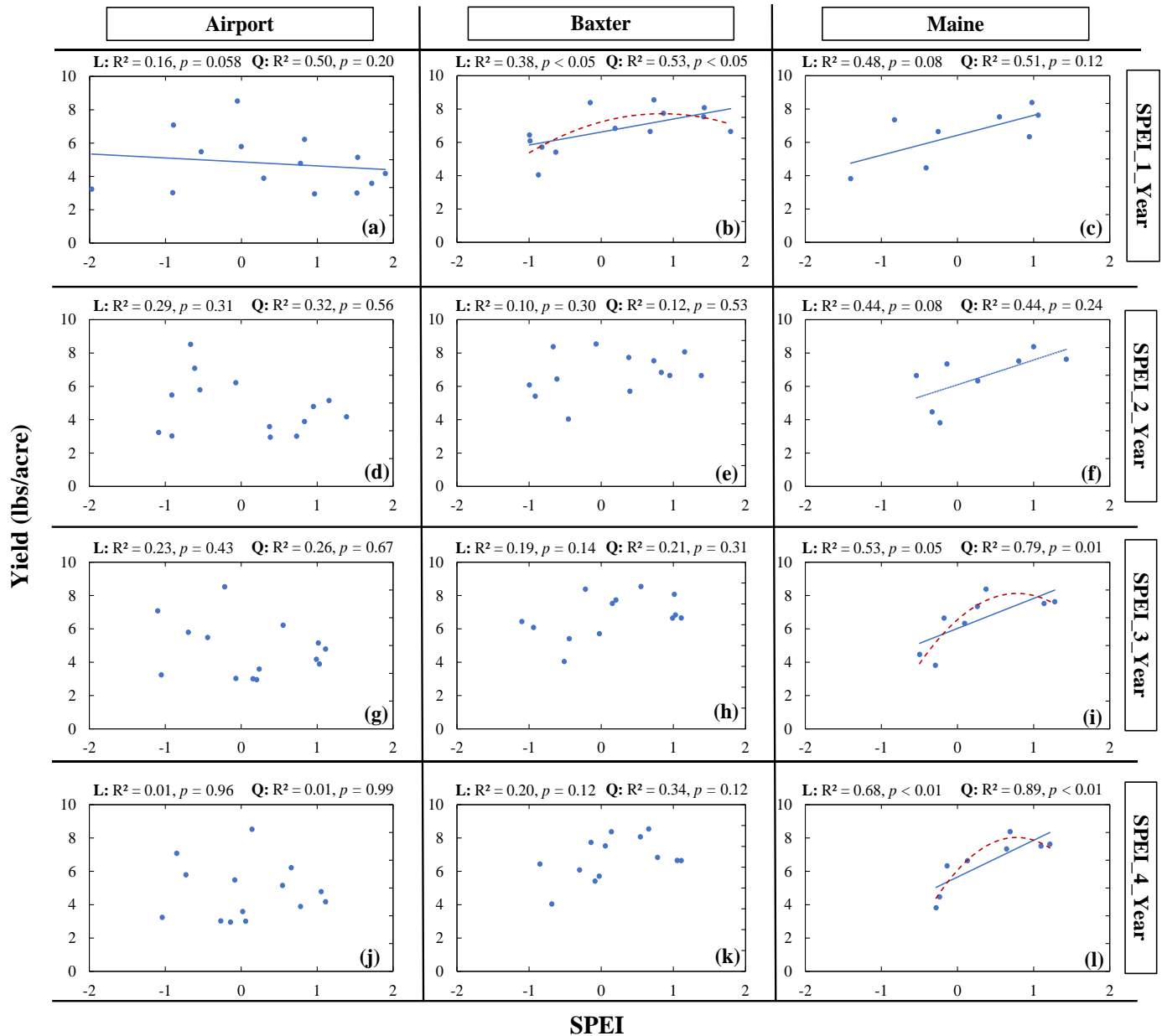


Figure 3.8. Average yield (lbs/acre) in a year for three different study zones: Airport, Baxter, and major wild blueberry (WB) production region in Maine in relation to (a), (b), (c) SPEI_1_Year ; (d), (e), (f) SPEI_2_Years (average SPEI of two consecutive years); (g), (h), (i) SPEI_3_Years (average SPEI of three consecutive years); (j), (k), (l) SPEI_4_Years (average of SPEI of four consecutive years). Here the SPEI data represents SPEI 6 of September. Here SPEI refers to SPEI_6 of September and it represents the SPEI (drought severity) of a growing season (April-September). A positive SPEI value represents wet condition where a negative SPEI value indicates dry condition. The numbers mentioned in the Y-axis was shortened by dividing the yield (lbs./acre) by 1000. The blue solid lines indicate significant ($p < 0.05$) or marginally significant ($p < 0.10$) linear relationships. The dashed red lines indicate significant ($p < 0.05$) or marginally significant ($p < 0.10$) quadratic relationships. The time-period of EVI and SPEI data was from 2000 to 2020.

3.4.4. Relationships between Vegetation indices and Productivity.

While comparing the impact of vegetation indices (EVI and NDVI) on the yield of irrigated Airport field and non-irrigated Baxter field, no significant relationship was found between the yield and growing season EVI and NDVI for the Airport and Baxter wild blueberry fields during the prune and crop year (Figure 3.9 and 3.10). The only significant correlation was found between mean EVI of prune year and crop year for Airport field and its yield when fitted with the quadratic relationship ($R^2 = 0.65$, $p = 0.03$), whereas correlation between mean NDVI of prune year and crop year for Airport field and its yield was non-significant.

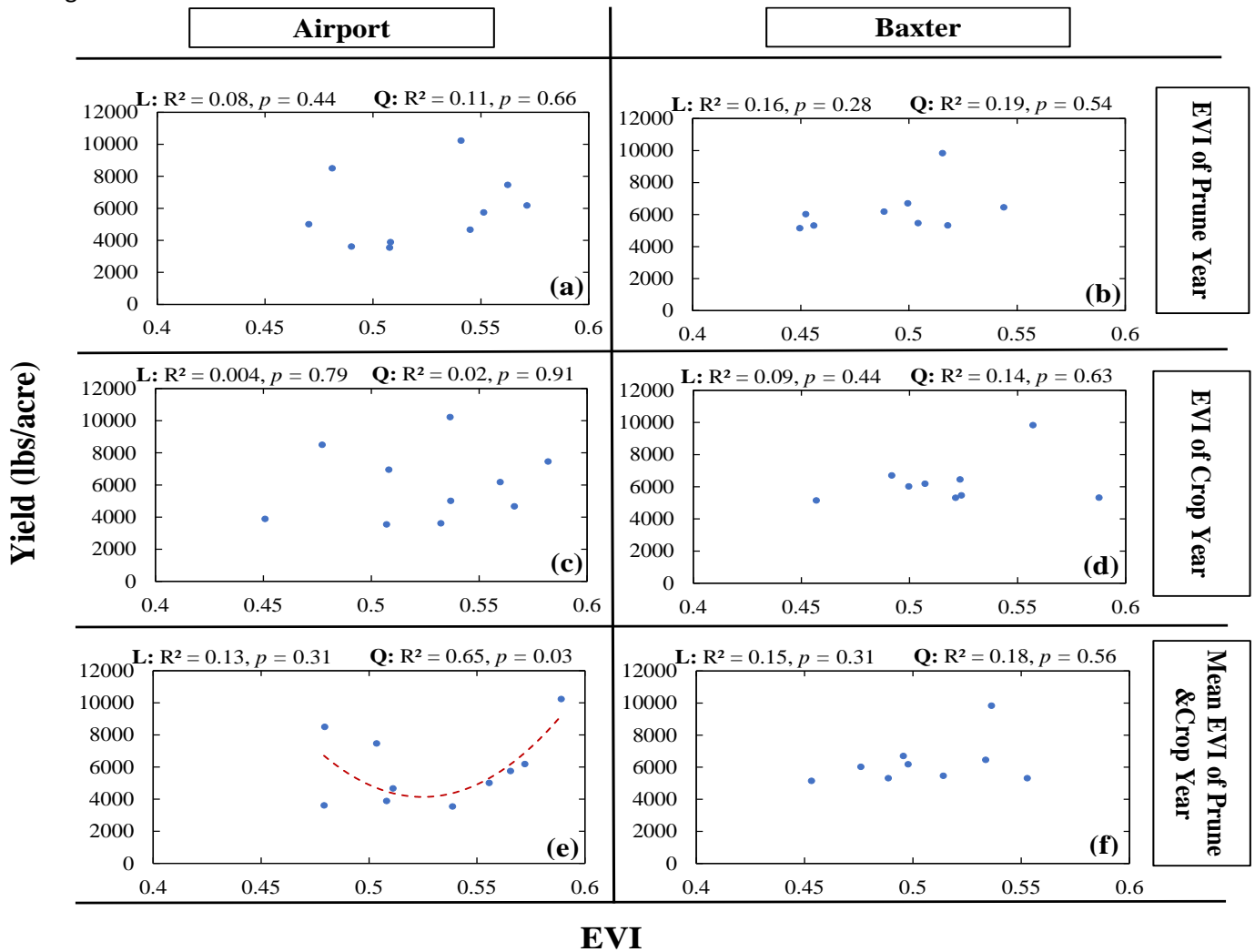


Figure 3.9. Relationship between wild blueberry yield (lbs/acre) and their average enhanced vegetation index (EVI) of (a,b) Prune year, (c,d) Crop year, and (e, f) average of Prune & Crop year from Airport (irrigated) and Baxter (non-irrigated) fields in Deblois, Maine.

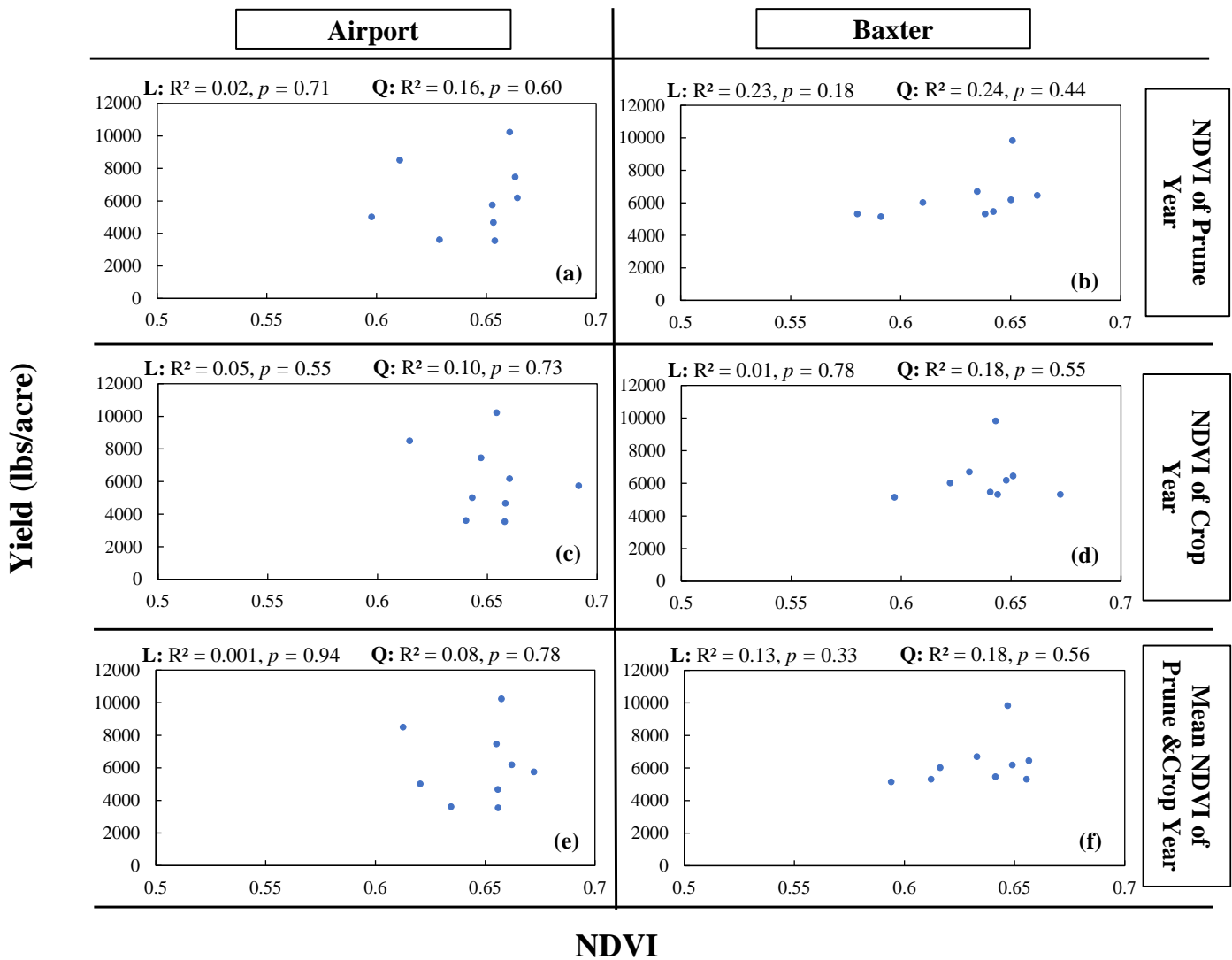


Figure 3.10. Relationship between wild blueberry yield (lbs/acre) and their average normalized difference vegetation index (NDVI) of (a,b) Prune year, (c,d) Crop year, and (e, f) average of Prune & Crop year from Airport (irrigated) and Baxter (non-irrigated) fields in Deblois, Maine.

3.5. Discussion

Our study revealed that, despite significant warming in the past century, there were no significant changes in drought patterns and drought impacts on the wild blueberry fields of Maine in the past 71 years. We also found that water conditions (dry or wet as indicated by SPEI) in the growing season have significant impacts on wild blueberry vegetation vigor (as indicated by NDVI and EVI) and production. The long-term water conditions (long-term average SPEI) have substantial significant impacts on wild

blueberry crop vegetation vigor (vegetation index: NDVI and EVI) as well as their production (yield) in Maine rather than the water conditions (SPEI) of the current growing season. The impact of water conditions on vegetation indices was more consistent and significant compared to the impact on yield. Interestingly, we also found that water conditions of the early growing season (April-June) might decide the fate of crop vegetation vigor and production of wild blueberry later in the season (July-August). We further found that, in terms of vegetation status, water conditions had little impact on the irrigated field. Water conditions indicated by SPEI had no impact on the yield of the irrigated field, suggesting irrigation effectively alleviated the impact of water deficits on yield of wild blueberries. Based on our analyses, we also found that satellite-based vegetation indices (NDVI and EVI) cannot be used to predict wild blueberry crop production. However, several previous studies found significant correlations between high-resolution spectroradiometer based vegetation indices and yield in different crops i.e., maize, wheat, and soybean (Bolton & Friedl, 2013; Marti et al., 2007; Teal et al., 2006). It could be because the yield of the wild blueberry system is more determined by other factors such as pollination rather than vegetation vigor. Also, further research could be carried out to test using drone based high-resolution data to predict yield of wild blueberries.

The absence of an increasing trend of drought index SPEI in the wild blueberry fields could be associated with a lack of significant change in precipitation patterns during the growing season (Belayneh et al., 2014; Potop et al., 2012; Tasnim et al., 2021). Although the atmospheric temperatures increased significantly in this region and wild blueberry fields in the past century (Arguez et al., 2010; Fernandez et al., 2020; Tasnim et al., 2021; Wolfe et al., 2018), the warming pattern and thus increased evapotranspiration (Tasnim et al., 2021) has not resulted in a significant increase in drought impact. The studied fields are in a temperate climate region, and they experience relatively low temperatures. The increase in evapotranspiration due to warming in this region possibly has not pushed the ecosystems here into the range of severe water deficits.

The water conditions of a relatively long period (SPEI of more than 11-12 months) showed significant and substantial impacts on vegetation vigor and the yield of wild blueberry crops. This could be because wild blueberries are a crop with a large perennial underground stem systems called rhizomes which can store sugar and nutrients (Bell, 2009; Drummond et al., 2009; Glass, 2001; Murray et al., 2002), and their health and yield could be mostly determined by sugar accumulation of previous years and not only that of the current growing season. Although the aboveground part of the wild blueberries is pruned to the ground every two years, the belowground rhizome and roots remain for a long time and the sugar storage underground could have a significant impact on the effect of precipitation on the crop in a long term (Easterling et al., 2007; Keyantash & Dracup, 2002). The wild blueberry crop requires only an inch of water per week (Hunt et al., 2009) and is regarded as a drought-resistant crop (Glass, 2001; Murray et al., 2002). This could be because of their large water and sugar storages in their underground tissues. The underground storage may weaken the effect of current year water conditions on crop health and yield.

Water conditions quantified by SPEI calculated based on the climate variables (temperature, precipitation and potential SPEI) certainly affects the vegetation status and vigor (NDVI and EVI) of wild blueberries. Vegetation greenness or vigor indicated by NDVI and EVI during both prune and crop years is affected by atmospheric temperature and precipitation during the growing period (Tasnim et al., 2021). Furthermore, the air temperature and precipitation directly affect the soil temperature and moisture availability (Tasnim et al., 2020). Soil temperature and moisture availability affect the nitrogen uptake in plants as well as their photosynthetic capacity (Wright et al., 2004), which consequently determines the growth, development, and yield of crops. However, neither drought index (SPEI) nor the vegetation indices (EVI and NDVI) are good predictors of the yield of wild blueberries. In fact, a previous study on the wild blueberries in Eastern Canada also did not find any correlations between the climate variables of that region with wild blueberry yield (Hall et al., 1982). This could be because of the fact that, besides the climatic condition wild blueberry yield during the crop year is affected by many other important factors

(Drummond, 2019a) such as – pollination, resources (water and nutrients) availability during fruit set and maturation, diseases and pathogens, etc. Though it has been found that vegetation indices are strongly correlated with yield in some crops (Bolton & Friedl, 2013), it might be a different story for wild blueberries. As vegetation indices represent the vegetation status, they might be useful to predict the vegetation health. Vegetation indices also correlates with leaf chlorophyll content which represents the nutritional quality of leaf tissue as well as photosynthesis capacity (Qu & Drummond, 2018) and might be related to number of developed flower buds (Obsie et al., 2020) of wild blueberries during crop year. Further, in order to turn the flower buds into final fruit set and yield, successful pollination is required (Drummond, 2019a; Qu et al., 2021; Samaniego et al., 2018) which is important and crucial to determine the final yield.

Water conditions (indicated by SPEI) of the early growing season (April-June) have a larger impact on vegetation status and yield of wild blueberry crops, compared to that of the later growing season (July-September) season. This could be related to pollination. Precipitation intensity and frequencies along with temperature and wind velocity during the pollination period (April-May) in crop year would affect the bee pollination, which significantly affects wild blueberry yield in July and August (Drummond, 2019a; Qu et al., 2021; Samaniego et al., 2018). Also, the availability of the resources such as- soil moisture and nutrients (Drummond, 2019a), determined by the precipitation and temperature (Hunt et al., 2009) during fruit set and maturation (May-June) right after the pollination period ends, decides the fate of the final fruit production (July-August) (Drummond, 2019a).

Increased natural water availability (as indicated by increased SPEI value) had a negative impact (marginally significant; $p = 0.058$) on the yield of the irrigated Airport field, compared to a positive relationship for the non-irrigated Baxter field despite being in the same location with same management practices (except irrigation). The positive correlation between SPEI and the yield of non-irrigated Baxter field suggests the importance of water conditions in determining yield and the need for optimum

irrigation practices to alleviate the impact of drought. The negative correlation between SPEI and yield in the irrigated field suggests that in years with high natural precipitation, irrigation may have a negative impact on yield for wild blueberries because they need only 1 inch of water per week (Hunt et al., 2009). Current uniform irrigation management practices without considering spatial variability in soil moisture retention capability may result in over irrigation in some parts of the field thus reduce yield. The fields of the major wild blueberry region (which are mostly non-irrigated) show similar patterns as the non-irrigated Baxter field. Thus, both water deficits and over-irrigation negatively impact the yield. Thus, it suggests that the introduction precision irrigation management practices might be useful to enhance the production of wild blueberries. The irrigation decoupled the relationship between SPEI and vegetation indices (EVI & NDVI), suggesting the positive effect of irrigation in mitigating drought effects. Meanwhile, the quadratic relationships between SPEI and vegetation indices as well as SPEI and yield suggest that when the optimum precipitation or water supply is reached, further increase in water supply may have a negative effect on crop vigor and yield. Similar results were also reported between EVI and precipitation from the wild blueberry fields in Downeast, Maine (Tasnim et al., 2021). Hence, no overall significant differences in vegetation indices or yield were observed between the irrigated Airport field and non-irrigated Baxter field, but in drought years (e.g., 2003), the yield and EVI of the irrigated field were higher than that of the non-irrigated field.

3.6. Conclusions

Overall, our study suggests that though temperature has been increasing significantly in the major Wild Blueberry production region of Maine, drought has not been increasing significantly over the last 71 years. However, accelerated warming and a projected decrease in soil water content may cause an increase in drought impact on agricultural systems in the future (Samaniego et al., 2018). The drought severity quantified by drought index SPEI had a stronger impact on vegetation status of the non-irrigated

fields compared to the irrigated field, and short-term SPEI was positively related to the yield of the non-irrigated field whereas negatively related for irrigated field. This suggests that both water deficit and over water supply than the optimum requirement of the wild blueberry system can negatively impact their production. However, maintaining optimum soil moisture is a challenge due to high spatial variability in soil water retention capacities. Therefore, developing a precision irrigation system could be an efficient way to mitigate the effects of water deficits. Interestingly, we found that long-term rather than short-term water conditions determine the vegetation vigor of wild blueberry crops. Thus, maintaining optimum water status for longer period is important for wild blueberries.

CHAPTER 4. USING UAV AND THERMAL-BASED REMOTE SENSING TO DETECT SPATIAL VARIATION IN WATER STRESS OF WILD BLUEBERRIES

4.1 Abstract

Wild blueberry fields are spatially heterogeneous in soil water retention capability and soil water content. The spatial heterogeneous nature of the soil water content along with crop physiological features make the crop water status more heterogeneous in wild blueberry fields. However, current conventional irrigation practices treat the field uniformly and irrigate uniformly. Uniform irrigation practices may result in over or under-application of water resources within a field that can negatively impact agricultural sustainability. The use of thermal-based crop water stress index (CWSI) has been studied in many crops in semi-arid regions and found as a cost-effective method in detecting real-time crop water status of large commercial fields remotely and non-destructively. But no previous studies have validated the usefulness of CWSI in a temperate crop like wild blueberries. Whether the predictive relationship changes in different crop development stages and changes with different reference temperatures is also unknown. In this multi-year study, a drone-based thermal sensor was used in 2019, 2020 and 2021 to validate different T_{wet} and T_{dry} reference-based CWSI in two large adjacent wild blueberry fields in Deblois, Maine, for estimating real-time leaf water status remotely. Different approach-based T_{wet} and T_{dry} references for estimating CWSI were tested. CWSI calculated with bio-indicator based T_{wet} and T_{dry} reference was found effective ($R^2 = 0.78$; $p < 0.05$) in detecting leaf water potential (LWP) in 2021, whereas the statistical T_{wet} and T_{dry} reference-based approach was less effective in 2019 ($R^2 = 0.37$; $p < 0.05$), 2020 ($R^2 = 0.34$; $p < 0.05$) and 2021 ($R^2 = 0.42$; $p < 0.05$). When we used flight as a factor in the linear mixed model relating CWSI to LWP, we found that different flights/crop developmental stages have a significant ($p < 0.05$) impact on CWSI-LWP models. CWSI-LWP model-based crop water status maps were also generated to show the water status variability in fields. Our findings support the presence of high variability in the crop water status of

wild blueberry fields. Our research also showed that drone-based thermal sensors can detect real-time crop water status within the field, with the CWSI calculated from bio-indicator based references being more reliable. Our results could be used for precision irrigation to increase the overall water use efficiency and profitability of wild blueberry farms.

4.2 Introduction

In commercial fields, wild blueberries are grown in semi-natural systems, which have been developed from naturally occurring native wild blueberry plants in forest understory but managed by growers (Yarborough, 2012). Fields of wild lowbush blueberries have high genetic diversity, with numerous genotypes coexisting in each field. Wild blueberry reproduces by cross-pollination and naturally propagated from seed, giving rise to a genetically distinct plant called genotype, which varies from each other. Due to that, considerable spatial heterogeneity in plant structural and functional traits is present. Along with that, several external factors may also vary within the field, and soil property is one of them. On a farm, there is significant spatial variation in soil physical and chemical properties within and between fields as well as from one year to the next (Farooque et al., 2012). Spatial variability of soil properties makes soil water retention capability and soil water content spatially heterogenous within a field. This high variability of soil properties and plant physiology makes water stress of wild blueberry genotypes within a field more heterogeneous. Irrigation is one of the major factors that drive crop health, development, and yield. Soil water deficits can cause stomatal closure, and decreases in photosynthetic rate and transpiration (Limpus, 2009). Overall, water stress can adversely affect the physiology and thus development and growth of crops, resulting in reduced growth and overall production (Olanike & Chandra, 2014; Rossini et al., 2013).

Wild blueberries are considered relatively adapted to drought stress compared to other cultivated crops (Murray et al., 2002). However, one inch (2.54 cm) of water per week is recommended for optimal

plant growth for wild blueberries (Yarborough, 2004a). Although moderate levels of drought stress have no impact on the growth, development, and production of wild blueberries (Murray et al., 2002), with effective irrigation, a total of 43% increase in yield can be obtained (Yarborough, 2004a). When soil moisture is limiting, effective irrigation can reduce crop failure by improving berry yield and quality thus increasing net profitability (Yarborough, 2012). During prune years, reduced precipitation and dry conditions can result in shorter stems with fewer buds (Hunt et al., 2009). In crop years, dry conditions can reduce berry quality and yield by preferentially directing its resources into vegetative growth at the expense of berry production (Hunt et al., 2009). When rainfall is limited, effective irrigation can provide the conditions for growing longer stems with more flower buds in the prune year and higher berry quality and yield in the crop year (Hunt et al., 2009). Insufficient water supply can lead to reduced plant growth and reduced yield, and irrigating excess water can drain pesticides and plant nutrients out of the root zone into ground or surface water. Thereby, excess irrigation increases expenses and decreases efficiency (Yarborough, 2012). Effective and precise irrigation management can provide a suitable soil environment for plant growth and yield, ultimately maximizing the water-use efficiency (Pascale et al., 2011). An efficient irrigation system requires continuous measurement of crop water status and soil moisture content to effectively forecast crop water needs in real-time and allow the possibility of providing just enough water to meet those needs at the right time (Osroosh et al., 2015). Due to genetic diversity among the genotypes as well as spatial variability of soil properties, crop water use and soil water retention capability can be drastically different within a field (Farooque et al., 2012). But current conventional irrigation practices treat the field and provide irrigation uniformly without considering the spatial variability of soil properties, genotype-specific water needs, and crop growth conditions. These uniform irrigation practices may result in over or under-application of water resources for specific genotypes. Over or under irrigation can affect physiological processes and increase fungal diseases, reducing overall production and net profitability. Also, water resources are limited and therefore it is important to

maximize their utilization. Thus, there is a need for an effective platform to quantify spatial variability in crop water stress within wild blueberry fields to increase water use efficiency and increase yield.

It is now possible to incorporate remote sensing technologies for better management of crops due to recent advancements in data acquisition from airborne and satellite-based platforms (Waheed et al., 2006). For crops, remote sensing technique refers to non-contact measurements of reflected/emitted electromagnetic radiation to indicate or predict soil physical properties or crop structural and physiological features (Mulla, 2013). Remote sensing data consisting of reflectance from different wavelengths can be interpreted for the detection of plant parameters such as crop health, crop cover, disease pressure, and crop water stress and are useful for operations such as yield prediction, pesticide application, fertilization and irrigation management. A prerequisite of precise crop farm management is the knowledge of spatial and temporal variability of crop physiological status as well as spatial variation of soil properties. The advancement of thermal, multispectral, and hyperspectral sensors made it possible to collect a large amount of remote sensing data in a cost-effective way but with higher spectral, spatial, and temporal resolutions (Liaghat & Balasundram, 2010). UAVs (Unmanned Aerial Vehicle) are unique in offering high-quality remote sensing data at the required spatial and temporal scale (Maes & Steppe, 2019). By detecting the temporal-spatial heterogeneity of crop water status, irrigation could be intelligently controlled in wild blueberry fields.

Accurate estimation of plant water status is significant in making management decisions for irrigation to reduce crop water stress and sustain crop production (Jones et al., 2004). Crop water stress or status is a complex phenomenon involving many factors. Plant water status is the combined effects of soil moisture status, atmospheric vapor pressure deficit (VPD), hydraulic resistance in xylem, and uptake capacity of water via roots (Ihuoma & Madramootoo, 2017). Effective measurement of plant water status is the key to precise irrigation for the optimization of crop yield. There are numerous conventional methods of quantifying field and plant water status for irrigation scheduling and can be grouped into three

main categories: (i) Soil-based, (ii) Weather-based, and (iii) Plant-based (Hillel, 2013). Soil-based approaches are soil moisture measurements, weather-based approaches are the use of evapotranspiration models and soil water balance models, and plant-based approaches are measurements of stomatal conductance, relative plant water content, and leaf water potential (Davis & Dukes, 2010; Hillel, 2013; Jones et al., 2004; Jones, 1999; Naor, 2000). Regular sampling of soil to measure the soil moisture has been used to assess the water status of the plant root zone (Gonzalez-Dugo et al., 2014). This method involves taking only a few measurements, assuming that soil water-holding capacity is uniform within the entire field (Clarke, 1997). This method is also quite time-consuming and assumes that all plants in a field transpire at the same rate (Ihuoma & Madramootoo., 2017), making this method ineffective for heterogeneous wild blueberry fields. Evapotranspiration models also assume the field is uniform in terms of vegetation cover and soil properties (Ihuoma & Madramootoo., 2017). Soil water balance-based models use the soil-water balance equation to predict soil moisture content indirectly, which requires estimated data of evaporation, rainfall events, and irrigation events (Ihuoma & Madramootoo., 2017). Although simple to use, this method is not very reliable and needs to be calibrated using actual field soil measurements. Stomatal conductance is an indirect indicator of plant water status based on stomatal opening, which can accurately represent plant water status but is very labor-intensive and not suitable for continuous monitoring and large commercial fields (Ihuoma & Madramootoo., 2017). Plant relative water content measurement is a direct and simple method for detecting leaf water status, but it is a destructive, laborious, time-consuming method not suitable for continuous monitoring of large commercial fields. Leaf water potential measurement is a widely accepted technique for the direct detection of leaf water status. Leaf water potential is the chemical potential of water, a direct measure of plant water status, which indicates the tension in the xylem conduits, and the force that causes water movement (Kramer & Boyer, 1995; Robbins & Dinneny, 2015). Leaf water potential can accurately indicate the current water status in the crop, but its measurement is usually a slow and destructive process and is

not suitable for estimating the water status of a large field with heterogeneous soil properties (Ihuoma & Madramootoo., 2017). Most of the current conventional methods of detecting plant water status for irrigation scheduling have some limitations for large-commercial scale frequent monitoring such as large wild blueberry fields with high spatial heterogeneity. Thus, there is a need for a method that can non-destructively assess the plant water status of the wild blueberry crops in real-time, considering the spatial heterogeneity of soil properties and crop physiology within a field.

Plant water status and canopy temperature are directly correlated, and canopy temperatures can represent plant water stress (Gonzalez-Dugo et al., 2014). Plant canopy temperature increases when plants absorb solar radiation as energy, but the canopy temperature cools down when the process of transpiration uses the energy. When water is scarce, a plant slows its transpiration by stomatal regulation, which raises the temperature of its leaves relative to unstressed, well-watered plants (Gonzalez-Dugo et al., 2014). But one single canopy temperature-based crop water stress model cannot be efficiently used in different weather conditions and crop developmental stages due to its dependency on environmental factors including the air temperature and vapor pressure deficit (Gonzalez-Dugo et al., 2014). Canopy temperature-based indices considering the surrounding real-time weather conditions are strongly related to plant water status, such as relative water content of crop and midday leaf water potential (Colaizzi et al., 2012). Based on this idea, canopy temperature products from thermal imagery have been widely used in multiple crops to detect water stress for irrigation scheduling. Recent developments in remote thermal imaging technology provide the opportunity to collect spatial data on crop surface temperature and make it easier to track canopy temperature variations across wide areas (Cohen et al., 2005).

Different indices have been developed based on the canopy temperature. Among the indices, crop water stress index (CWSI), developed by Idso et al. (1981), has been widely used. CWSI is used to map the in-field variations of crop water stress. , CWSI based on thermal imaging has evolved into a reliable index for several crop systems (Gonzalez-Dugo et al., 2014; Cohen et al., 2017). CWSI is the

difference in canopy leaf temperature between the dry references and the wet reference under atmospheric conditions. In ambient settings, it can be calculated by equation (1).

$$CWSI = \frac{T_{canopy} - T_{wet}}{T_{dry} - T_{wet}} \dots\dots\dots(1)$$

Where T_{canopy} refers to the leaf canopy temperature; T_{wet} is the lower reference temperature corresponding to the temperature of a fully transpiring leaf and T_{dry} is the upper baseline corresponding to the canopy temperature of a fully non-transpiring leaf. Wet and Dry baselines vary in different crops as well as with the agroclimatic conditions in which the crop is being grown (Idso et al., 1981). Thermal infrared sensors are now commercially available, and UAVs with thermal infrared cameras can now be used to detect water stress and plan irrigation (Maes & Steppe, 2019). CWSI based on thermal camera/sensors has been studied in many crops in semi-arid regions and found to be a cost-effective method for large commercial fields. This method is also remotely and non-destructively. However, no previous studies have been done on validating the usefulness of CWSI in temperate crop systems like wild blueberries. Also, the reliability of using different type of references for calculating CWSI, and whether the relationship changes across different crop development stages have not been well-tested. To do so, the objectives of this chapter were to:

- 1) Validate different dry and wet reference based CWSI in predicting crop LWP in large heterogenous commercial wild blueberry fields;
- 2) Test whether the relationship between CWSI and LWP changes over different crop development stages;
- 3) Quantify the spatial variation of water status within wild blueberry fields using the CWSI-LWP model.

4.3 Methods

4.3.1 Study Site

The study was conducted on two adjacent commercial blueberry fields owned by Jasper Wyman and Sons in Deblois, Maine (Longitude: -68.0001° N, Latitude: 44.7350° W). Deblois is located in Washington county, the largest wild blueberry-producing county in Maine. The soil type of these study sites is sandy loam, which is well-drained acidic soil. Deblois is located in the Downeast coastal region of Maine, which has a four-season climate with an average low of -10.6 °C and a high of 24.2 °C, and monthly average precipitation low of 85.1 mm and high of 136.4 mm. These commercial crop fields contain many different genotypes of wild blueberry plants growing within a particular field. Due to this pattern, 30 plots (genotypes) in 2019 and 20 plots in 2020 and 2021 were systematically selected from the area for field measurement to cover a high range of phenotypic traits and the entire field. The study site had two adjacent fields (Figure 4.1), irrigated and non-irrigated (Irrigated: Airport and Non-irrigated: Baxter). The area of the Airport field is 77 acres (0.31 km²), and the Baxter field is 39 acres (0.16 km²). These two fields were selected to understand the effectiveness of current water management practices in a single area.

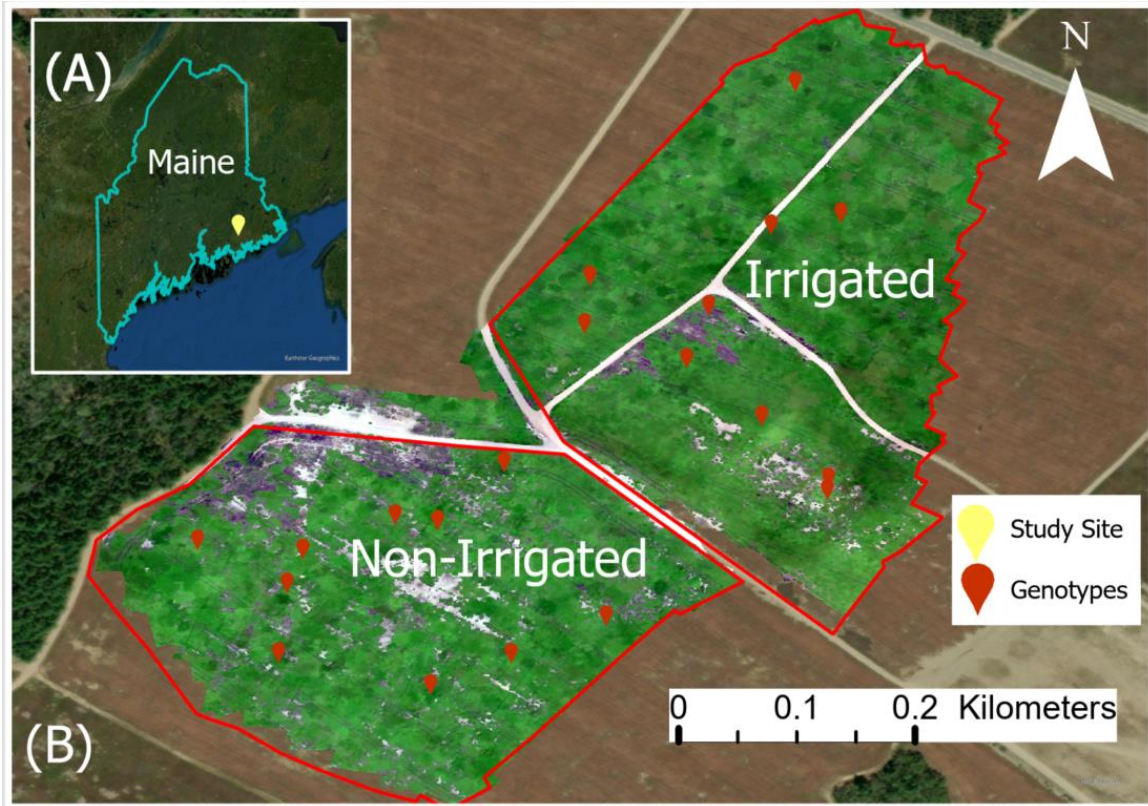


Figure 4.1: Location of the study site. (A) The yellow dot indicates the study site location in Jasper Wyman & Sons blueberry farm in Deblois, Maine. (B) Red dots indicate genotypes at irrigated Airport and non-irrigated Baxter wild blueberry field (red boundary line) for on-ground and drone data collection.

4.3.2 Remote Sensing Data: UAV Platform and Sensors

A custom-built UAV was used as a platform for the Micasense Altum (Mica Sense, Seattle, WA, USA) sensor, which was used in this study to acquire thermal images (Figure 4.2). The MicaSense Altum can capture six bands - blue, green, red, red edge, near-infrared, and thermal. These bands are concurrently captured, which makes it simpler to align and use the data for analytics. Only the data from the infrared thermal sensor was used in this study.



Figure 4.2. UAV platform and sensors used in acquiring multispectral and thermal images. (A) UAV platform; (B) Altum MicaSense sensor. The figures of the UAV platform and Altum sensor were acquired from micasense.com.

The resolution of the Altum thermal infrared sensor is 160 x 120 at a 1.77 mm focal length. It can capture thermal images at 81cm per pixel from a 120m distance. The pixel resolution size is well enough to capture the reflectance properties of individual clones. The UAV and Altum sensor setup were used due to their low-cost but high-performance photogrammetry across various disciplines. This type of commercial sensors have been proven to be particularly suitable for UAV-based remote sensing research in agriculture (Dugdale et al., 2019; Gu et al., 2021; Long et al., 2018).

4.3.3 UAV Image Acquisition

The image acquisitions were conducted three times on three major crop developmental phases in the summer of 2019, three times on three vegetative leaf developmental phases in the summer of 2020, and again three times in three major crop developmental phases in the summer of 2021 (Table 4.1). The dates were chosen based on the developmental phases of wild blueberries. Image acquisition and field ground data collection dates were done on sunny days with favorable conditions for UAV flights to avoid any errors related to cloud and wind. The flight plan for image collection was designed and executed using the Pix4D Planner software (Pix4D S.A., Switzerland). Flight height was approximately 120 m, resulting in 81 cm per pixel image resolutions for the thermal images. These image acquisitions were done from 12:00–13:00 (solar time). A Downwelling Light Sensor (DLS) was mounted on the top of the drone for the Altum sensor to measure incoming irradiance in the five individual bands to aid in acquiring imagery with accurate reflectance in changing illumination conditions. Before and after each UAV flight, spectral calibration was conducted by acquiring an image of a Calibrated Reflectance Panel (CRP) by Micasense. The CRP can accurately represent light conditions during the flight (Aasen et al., 2018).

4.3.4 Image Processing and Analysis

The UAV imagery was processed using the Agisoft Metashape software 1.6.2 (Agisoft LLC., St. Petersburg, Russia) with the following processing stages:

1. Adding images using the multi-camera system for multispectral cameras.

2. Reflectance calibration of the images.
3. Camera alignment and optimization.
4. Surface reconstruction (dense cloud, mesh, digital elevation).
5. And finally ortho mosaic generation.

The final thermal images (Orthomosaic tiff) files were then transferred to ArcGIS Pro (Version 2.7, Esri, Redlands, CA, USA) for further analysis. CWSI was then calculated using the Raster Calculator tool in ArcGIS Pro. A shapefile was created by digitizing the ground sampled genotypes in ArcGIS Pro with the help of GPS coordinates. The clone-specific index values were extracted from these images using the Spatial Analyst tool in ArcGIS Pro.

4.3.5 Ground Sampling

Ground measurements were taken over the two fields at the same time of flights (three times per year, Table 4.1). These represent important development stages of wild blueberries throughout the summer growing season over the two-year crop cycle. We collected data over the different developmental stages to determine the temporal variation in water status and spectral response over the seasonal cycle of blueberry growth due to these variations in water status. 30 ground sample measurements (15 genotypes on each field) in 2019 and 20 ground sample measurements (10 genotypes for each field) selected in 2020 and 2021 by an arbitrarily sampling design and based on genotypes morphological distinction. One wild blueberry stem was randomly selected from each treatment area (per genotype) at midday between 12.00-13.00-hour solar time to measure midday leaf water potential. The samples were stored in plastic airtight Ziploc bags with some water-soaked tissue to maintain the moisture in coolers and measured within two-three hours. The measurements were taken using a leaf pressure chamber (PMS Inc., Albany, OR, USA). Canopy leaf temperature was measured using a Fluke 62 Max+ hand-held infrared thermometer (Fluke Corporation, Everett, WA, USA) from randomly selected four leaves of each genotype. Soil water content was measured by a Fieldscout TDR 150 Soil Moisture Meter (Spectrum

Technologies Inc., Aurora, IL, USA) from four random places within each of the genotypes of Jasper Wyman & Sons blueberry farm.

4.3.6 Climatic Data Acquisition

In this study, climatic variables such as standard precipitation and evapotranspiration index (SPEI), the mean precipitation, and mean air temperature of the last 14 consecutive days before the flight dates were acquired and used to understand the water and climatic conditions. SPEI is an index to indicate drought and calculated based on precipitation, temperature, and potential evapotranspiration. A positive SPEI value represents a wet condition whereas a negative SPEI value indicates a dry condition. SPEI data was collected from the open access database “SPEI Global Drought Monitor” (<https://spei.csic.es>); accessed on 10 March 2022) in netcdf format. The netcdf files containing the SPEI data were transferred to ArcGIS Pro Version 2.7 (ESRI, Redlands, California, USA) to acquire SPEI data for the study site. The dataset of climate variables such as total precipitation and mean temperature were acquired from the Prism database using the google earth engine-based platform “Climate Engine” (<https://climate.engine.appspot.com/climateEngine>).

Table 4.1: Dates of UAV flight and ground sampling in accordance with wild blueberry crop developmental stages in the years 2019, 2020 and 2021 along with the mean precipitation, mean air temperature and standard precipitation and evapotranspiration index (SPEI) of the last consecutive 14 days before the flight dates. A positive SPEI value represents a wet condition whereas a negative SPEI value indicates a dry condition.

2019	Flight 1	Flight 2	Flight 3
	07/03/2019	07/25/2019	08/14/2019
Developmental Stage	Green Fruit	Color Break	Mature Fruit
Precipitation	10.16	2.93	2.54
Temperature	15.45	18.15	19.68
SPEI	1.45	0.17	-0.44
Conditions	(Very Wet)	(Slightly Wet)	(Moderate Dry)
2020	Flight 1	Flight 2	Flight 3
	07/09/2020	08/04/2020	08/26/2020
Developmental Stage	Leaf Development	Full Mature Leaves	Leaf Senescence
Precipitation	3.96	2.60	0.69
Temperature	19.51	21.7	12.37
SPEI	0.17	-0.32	-0.54
Conditions	(Slightly Wet)	(Moderate Dry)	(Moderate Dry)
2021	Flight 1	Flight 2	Flight 3
	06/18/2021	07/01/2021	07/15/2021
Developmental Stage	Green Fruit	Color Break	Early Mature Fruit
Precipitation	1.14	2.93	7.02
Temperature	19.25	20.7	17.79
SPEI	-1.45	-0.24	1.69
Conditions	(Very Dry)	(Slightly Dry)	(Wet)

4.3.7. CWSI-LWP Regression Models

To examine the relationship between CWSI and Leaf water potential (LWP), the mean temperature values of individual clones were extracted from the UAV-based thermal imagery. In order to develop models for leaf water potential (LWP) prediction from crop water stress index (CWSI), these values were used. Calculations of the CWSI were done according to (Jackson et al., 1981), where T_{canopy} is the thermal image-based actual canopy temperature, T_{wet} is the lower boundary temperature corresponding to a fully transpiring leaf with open stomata, and T_{dry} is the upper boundary temperature corresponding to a non-transpiring leaf with closed stomata, respectively (Eq 1).

$$CWSI = \frac{T_{canopy} - T_{wet}}{T_{dry} - T_{wet}} \dots\dots\dots(1)$$

Meron et al. created an empirical technique of temperature extraction of crop canopy from remotely sensed thermal imagery for crop field with some exposed soil (Meron et al., 2010). This technique is based on two empirical assumptions. The first assumption is that in a thermal image, pixels connected to the canopy are different from those related to the soil and other objects, which can be distinguished by higher and lower limits related to the temperature of air using this equation (Eq 2):

$$(T_{air} - 10) < T_{cr} < (T_{air} + 7) \dots\dots\dots(2)$$

In equation (2) T_{air} represents temperature (°C) of air and T_{cr} represents thermal canopy pixels in a thermal imagery. The second assumption is that the temperature of canopy is associated to the mean temperature of the coldest 33% of canopy pixels. T_{canopy} , can be calculated using equation (Eq 3):

$$T_{canopy} = \frac{\sum_{i=1}^{0.33n} T_{cr_i} * f_i}{\sum_{i=1}^{0.33n} f_i} \dots\dots\dots(3)$$

Where T_{canopy} ($^{\circ}\text{C}$) represents the canopy temperature, the total number of pixels in canopy temperature 33 % class histogram is denoted by f , and n is the total number of pixels after discarding non-crop related pixels such as soils (Eq. 2). In this study, the canopy temperature pixel was obtained from the thermal imagery following these assumptions using ArcGIS Pro software.

Various T_{wet} and T_{dry} baseline forms relating to the temperature of leaf canopy with full transpiration (open stomata) and no transpiration (closed stomata) have been developed and applied for calculating CWSI and water stress mapping. Because empirical T_{dry} and T_{wet} baselines depend on vapor pressure deficit (VPD), determination of T_{dry} and T_{wet} requires long-term monitoring of canopy temperature (Cohen et al., 2017; Jackson et al., 1981). This long-term monitoring makes it unsuitable for field application. Several studies have found that theoretical wet-baselines are comparable or less effective compared to empirical baselines (Alchanatis et al., 2010; Cohen et al., 2005). Recently, two approaches, the statistical approach and the measured bio-indicator approach were suggested and successfully used for determining the T_{wet} and T_{dry} baselines (Cohen et al., 2017). Bio-indicator based T_{wet} and T_{dry} baselines are based on the actual canopy temperature measurements relating to the temperature of leaf canopy with full transpiration (open stomata) and no transpiration (closed stomata). In this study, the statistical T_{wet} and empirical T_{dry} reference approaches were taken in 2019 and 2020. The statistical approach, in general, considers the average canopy temperature of the cooler 5-10% of pixels as the T_{wet} reference. Clawson et al. (1989) proposed this approach suggesting that the coldest leaf temperatures of a certain crop in a specific area might be taken to constitute the T_{wet} reference temperature for CWSI computation (Clawson et al., 1989). The empirical approach was taken for calculating T_{dry} by adding 5°C with air temperature: $T_{dry} = T_{air} + 5^{\circ}\text{C}$, and a statistical approach was taken for T_{wet} calculation in 2019 and 2020. Air temperature data was taken from a field-based meteorological station. Bio-indicator based T_{wet} and T_{dry} reference approach was taken in 2021. Bio-indicator based T_{wet} base temperature for each flight date was measured by a Fluke 62 Max+ handheld infrared thermometer (Fluke Corporation, Everett, WA,

USA) from three leaves from three arbitrarily selected stems from three arbitrarily selected genotype, wetted on both sides using a sprayer. The wetter solution was made by mixing detergent with water. Bio-indicator based T_{dry} base temperatures for each flight date were measured by the infrared thermometer from three leaves of three randomly selected stems from three different arbitrarily selected genotypes. These leaves were treated with a thin layer of petroleum jelly to both upper and lower surfaces to prevent transpiration (Jones, 1999). The bio-indicator based wet and dry temperature values were then averaged for each date before being used in the CWSI calculations.

4.3.8 Statistical Analysis

In this study, RStudio software (RStudio, PBC, Vienna, Austria) was used for statistical analysis. To determine differences in water condition-related variables (SWC, LWP, and CWSI) between the two fields and genotypes within fields, we used a mixed model with the field as the fixed effect and the genotype nested within the field as the random effect ($\alpha = 0.05$). This approach was suggested for split-plot designs (genotypes within fields) (Harrison et al., 2018). We determined the statistical significance of the linear regression (in the form of $a + bx$) relationship using the coefficient of determination and its significance (α) at $p < 0.05$. We used the “lme4” package in R to test the effect of flight (crop developmental stage) on the LWP-CWSI models by using a linear mixed model and putting flight as a random factor. The effect of different flights and fields on water condition-related variables SWC, LWP, and CWSI was tested using a Generalized Linear Model in R.

4.4 Results

4.4.1. Variation In Soil and Crop Water Conditions

Table 4.2: Comparison in water conditions between irrigated (Airport) and non-irrigated (Baxter) wild blueberry fields in the minimum (Min), maximum (Max), mean, and standard deviation (SD) of soil water content (SWC), leaf water potential (LWP), crop water stress index (CWSI) in three different flights during the 2019 crop season. Water conditions classification of the respective flight was done based on the standard precipitation and evapotranspiration index (SPEI 14D), which determines the water condition based on precipitation, temperature, and potential evapotranspiration conditions of the consecutive 14 days before the flight dates.

2019		Flight 1			Flight 2			Flight 3		
Water Condition		Very Wet			Slight Wet			Moderately Dry		
		SWC (%)	LWP (Mpa)	CWSI (#)	SWC (%)	LWP (Mpa)	CWSI (#)	SWC (%)	LWP (Mpa)	CWSI (#)
Airport (Irrigated)	Min	31.83	-1.19	0.13	14.9	-1.86	0.22	16.93	-1.71	0.28
	Max	39.03	-0.80	0.32	32.2	-0.99	0.47	34.67	-1.29	0.61
	Mean	35.61	-1.02	0.26	20.65	-1.31	0.31	25.62	-1.44	0.45
	SD	3.15	0.13	.06	5.01	0.24	0.08	3.80	0.23	0.10
	Difference Within Field	Yes ($p < 0.05$)	NA ¹	NA ¹	Yes ($p < 0.05$)	NA ¹	NA ¹	Yes ($p < 0.05$)	NA ¹	NA ¹
Baxter (Non-irrigated)	Min	36.66	-1.25	0.13	24.33	-1.70	0.20	11.08	-1.77	0.34
	Max	45.3	-0.69	0.53	34.33	-0.99	0.48	18.60	-1.22	0.77
	Mean	42.44	-1.05	0.34	28.21	-1.30	0.36	15.21	-1.55	0.56
	SD	2.49	0.19	0.14	2.95	0.20	0.08	6.19	0.22	0.13
	Difference Within Field	No ($p = 0.26$)	NA ¹	NA ¹	No ($p = 0.07$)	NA ¹	NA ¹	Yes ($p < 0.05$)	NA ¹	NA ¹
Mean (Both fields)	39.71	-1.03	0.30	24.62	-1.30	0.33	19.84	-1.50	0.51	
Difference Between Fields	No ($p = 0.67$)	No ($p = 0.75$)	No ($p = 0.28$)	No ($p = 0.12$)	No ($p = 0.91$)	No ($p = 0.22$)	Yes ($p < 0.05$)	Yes ($p < 0.05$)	Yes ($p < 0.05$)	

¹ no sub-sample contribution

We found variation in the SWC, LWP and CWSI among genotypes of the fields, between the fields as well as among different flights in 2019 (Table 4.2). The SWC varied by 1.22-fold in the irrigated field and varied by 1.23-fold in the non-irrigated field in flight 1. Variation within the field was statistically significant ($p < 0.05$) for the irrigated Airport field whereas non-significant for the non-irrigated Baxter field for flight 1. The water condition of the last 14 consecutive days before flight 1 was very wet (SPEI 14D: 1.45). The LWP varied by 1.48-fold and 1.02-fold in the irrigated and non-irrigated fields, respectively, in flight 1. The CWSI varied by 2.46 and 4.07 -fold in the irrigated and non-irrigated fields, respectively, in flight 1.

In flight 2, the SWC varied by 2.16 -fold in the irrigated field and varied by 1.41-fold in the non-irrigated field. Variation within the field was statistically significant ($p < 0.05$) for the irrigated Airport field, whereas non-significant for the non-irrigated Baxter field. The water condition of the last 14 consecutive days before flight 2 was slightly wet (SPEI 14D: 0.17). The LWP varied by 1.87 and 1.71-fold in the irrigated and non-irrigated fields, respectively, in flight 2. The CWSI varied by 2.13 and 2.4-fold in the irrigated and non-irrigated fields, respectively, in flight 2. In flight 3, the SWC varied by 2.04-fold in the irrigated field and varied by 1.67-fold in the non-irrigated field. Variation within the field was statistically significant ($p < 0.05$) for both irrigated Airport field and non-irrigated Baxter field. The water condition of the last consecutive 14 days before flight 3 was moderately dry (SPEI 14D: -0.44). The LWP varied by 1.32 and 1.45-fold in the irrigated and non-irrigated fields, respectively, in flight 3. The CWSI varied by 2.17 and 2.26-fold in the irrigated and non-irrigated fields, respectively, in flight 3. In 2019, the difference in SWC within the field was significant for all flights in irrigated Airport field but only in flight 3 for the non-irrigated Baxter field.

In flight 1, on which the water condition of the last 14 days was very wet (SPEI 14D: 1.45), the mean SWC was 1.19-fold higher in the irrigated field compared to the non-irrigated whereas the mean LWP was 2% lower (lower value indicates high water stress) and mean CWSI was 1.30-fold higher in the

non-irrigated field compared to the irrigated one. In flight 2, on which the water condition of the last 14 days was slightly wet (SPEI 14D: 0.17), the mean SWC was 1.36-fold higher in the irrigated field compared to the non-irrigated whereas the mean LWP was 1% lower and mean CWSI was 1.16 -fold higher in the non-irrigated field compared to the irrigated one. In flight 3, the water condition of the last 14 days was moderately dry (SPEI 14D: -0.44), and the mean SWC was 1.68-fold higher in the irrigated field compared to the non-irrigated whereas the mean LWP was 7% lower and mean CWSI was 1.24-fold higher in the non-irrigated field compared to the irrigated one. Variation was also observed among different flights, with the highest mean SWC and LWP recorded in flight 1 (very wet) on the irrigated field and lowest in the non-irrigated field on flight 3 (moderately dry), and mean CWSI was observed highest in flight 3 and lowest in flight 1. Despite differences in mean SWC, LWP and CWSI, the variations between the fields in terms of all water condition-related factors (SWC, LWP and CWSI) were statistically significant only during flight 3 of 2019, on which the water condition of the last 14 days was moderately dry.

Table 4.3: Comparison in water conditions between irrigated (Airport) and non-irrigated (Baxter) wild blueberry fields in the minimum (Min), maximum (Max), mean, and standard deviation (SD) of soil water content (SWC), leaf water potential (LWP), crop water stress index (CWSI) in three different flights during the 2020 crop season. Water conditions classification of the respective flight was done based on the standard precipitation and evapotranspiration index (SPEI 14D), which determines the water condition based on precipitation, temperature, and potential evapotranspiration conditions of the consecutive 14 days before the flight dates.

2020		Flight 1			Flight 2			Flight 3		
Water Condition		Slightly Wet			Moderately Dry			Moderately Dry		
		SWC (%)	LWP (Mpa)	CWSI (#)	SWC (%)	LWP (Mpa)	CWSI (#)	SWC (%)	LWP (Mpa)	CWSI (#)
Airport (Irrigated)	Min	20.56	-1.22	0.11	14.73	-1.28	0.33	24.83	-1.51	0.23
	Max	39.50	-0.86	0.45	45.46	-1.24	0.45	38.86	-1.11	0.87
	Mean	30.25	-1.06	0.30	29.56	-1.25	0.40	32.74	-1.32	0.54
	SD	5.71	0.18	0.14	9.19	0.02	0.06	5.48	0.15	0.28
	Difference Within Field	Yes ($p < 0.05$)	NA ¹	NA ¹	Yes ($p < 0.05$)	NA ¹	NA ¹	Yes ($p < 0.05$)	NA ¹	NA ¹
Baxter (Non-irrigated)	Min	17.7	-1.32	0.16	8.63	-1.69	0.37	14.43	-1.40	0.32
	Max	24.7	-0.84	0.58	18.23	-1.11	0.79	26.01	-1.57	0.95
	Mean	21.47	-1.07	0.35	12.51	-1.29	0.58	22.09	-1.51	0.66
	SD	2.70	0.20	0.16	3.04	0.23	0.15	3.73	0.07	0.24
	Difference Within Field	No ($p = 0.06$)	NA ¹	NA ¹	Yes ($p < 0.05$)	NA ¹	NA ¹	Yes ($p < 0.05$)	NA ¹	NA ¹
Mean (Both fields)	27.11	-1.06	0.32	21.03	-1.28	0.51	26.17	-1.41	0.60	
Difference Between Fields	Yes ($p < 0.05$)	No ($p = 0.89$)	No ($p = 0.57$)	Yes ($p < 0.05$)	No ($p = 0.89$)	Yes ($p < 0.05$)	Yes ($p < 0.05$)	Yes ($p < 0.05$)	No ($p = 0.48$)	

¹ no sub-sample contribution

We found variation in the SWC, LWP and CWSI among fields, between fields as well as among different flights in 2020 (Table 4.3). The SWC varied by 1.92-fold in the irrigated field and varied by 1.40-fold in the non-irrigated field in flight 1. Variation within the field was statistically significant ($p < 0.05$) for the irrigated Airport field, whereas non-significant for the non-irrigated Baxter field. The water condition

of the last consecutive 14 days before flight 1 was slightly wet (SPEI 14D: 0.17). In flight 1, the LWP varied by 1.41-fold and 1.57-fold in the irrigated and non-irrigated fields, respectively. The CWSI varied by 4.09 and 3.62 -fold in the irrigated and non-irrigated fields, respectively, in flight 1. In flight 2, the SWC varied by 3.08-fold in the irrigated field and varied by 2.11-fold in the non-irrigated field. Variation in SWC within the field was statistically significant ($p < 0.05$) for both irrigated Airport field and non-irrigated Baxter field for flight 2. The water condition of the last consecutive 14 days before flight 2 was moderately dry (SPEI 14D: - 0.32). The LWP varied by 1.03 and 1.52-fold and the CWSI varied by 1.36 and 2.13-fold in the irrigated and non-irrigated fields, respectively, in flight 2. In flight 3, the SWC varied by 1.56-fold in the irrigated field and varied by 1.80-fold in the non-irrigated field. Variation within the field was statistically significant ($p < 0.05$) for both irrigated Airport field and non-irrigated Baxter field for flight 3. The water condition of the last consecutive 14 days before flight 3 was moderately dry (SPEI 14D: -0.54). The LWP varied by 1.36 and 1.12-fold and the CWSI varied by 3.78 and 2.96-fold in the irrigated and non-irrigated fields, respectively, in flight 3.

In flight 1, on which the water condition of the last 14 days was slightly wet, the mean SWC was 1.40-fold higher in the irrigated field compared to the non-irrigated, whereas the mean LWP was 1% lower and mean CWSI was 1.16-fold higher in the non-irrigated field compared to the irrigated one. The difference in SWC was statistically significant ($p < 0.05$) for flight 1, whereas the difference was not significant for LWP and CWSI. In flight 2, on which the water condition of the last 14 days was moderately dry, the mean SWC was 2.36-fold higher in the irrigated field compared to the non-irrigated, whereas the mean LWP was 3% lower and mean CWSI was 1.45 -fold higher in the non-irrigated field compared to the irrigated one. The difference in SWC and CWSI was statistically significant ($p < 0.05$), whereas the difference was not significant for LWP. In flight 3, on which the water condition of the last 14 days was moderately dry, the mean SWC was 1.48-fold higher in the irrigated compared to the non-irrigated field, whereas the mean LWP was 17% lower and mean CWSI was 1.22-fold higher in the non-irrigated field

compared to the irrigated one. The difference in SWC, LWP between fields was statistically significant ($p < 0.05$) but not the CWSI. Variation was also observed among different flights, with the highest mean SWC and LWP recorded in flight 1 (very wet) on the irrigated field and lowest SWC recorded in flight 2 (moderately dry) and the lowest LWP (moderately dry) was recorded in flight 3 and for the non-irrigated field. And the mean CWSI was observed highest in flight 3 (moderately dry) for the non-irrigated field and lowest in flight 1 for the irrigated field (very wet).

Table 4.4: Comparison in water conditions between irrigated (Airport) and non-irrigated (Baxter) wild blueberry fields in the minimum (Min), maximum (Max), mean, and standard deviation (SD) of soil water content (SWC), leaf water potential (LWP), crop water stress index (CWSI) in three different flights during the 2021 crop season. Water conditions classification of the respective flight was done based on the standard precipitation and evapotranspiration index (SPEI 14D), which determines the water condition based on precipitation, temperature, and potential evapotranspiration conditions of the consecutive 14 days before the flight dates.

2021		Flight 1			Flight 2			Flight 3		
Water Condition		Very Dry			Slight Dry			Very Wet		
		SWC (%)	LWP (Mpa)	CWSI (#)	SWC (%)	LWP (Mpa)	CWSI (#)	SWC (%)	LWP (Mpa)	CWSI (#)
Airport (Irrigated)	Min	12.55	-1.37	0.40	17.43	-1.27	0.29	20.58	-1.21	0.21
	Max	23.85	-1.08	0.65	28.77	-0.90	0.43	31.16	-0.83	0.46
	Mean	19.70	-1.26	0.49	20.89	-1.11	0.37	24.71	-1.01	0.31
	SD	3.74	0.09	0.07	3.10	0.11	0.05	4.87	0.09	0.08
	Difference Within Field	Yes ($p < 0.05$)	NA ¹	NA ¹	Yes ($p < 0.05$)	NA ¹	NA ¹	Yes ($p < 0.05$)	NA ¹	NA ¹
Baxter (Non-irrigated)	Min	9.77	-1.71	0.41	13.45	-1.35	0.32	20.23	-1.19	0.25
	Max	18.65	-1.13	0.79	21.86	-1.03	0.57	25.76	-1.01	0.40
	Mean	14.98	-1.46	0.58	18.22	-1.21	0.43	23.10	-1.07	0.34
	SD	2.56	0.17	0.12	2.64	0.09	0.07	2.05	.06	0.05
	Difference Within Field	Yes ($p < 0.05$)	NA ¹	NA ¹	Yes ($p < 0.05$)	NA ¹	NA ¹	Yes ($p < 0.05$)	NA ¹	NA ¹
Mean (Both fields)	17.33	-1.36	0.54	19.55	-1.14	0.40	28.15	-1.04	0.32	
Difference Between Fields	Yes ($p < 0.05$)	Yes ($p < 0.05$)	Yes ($p < 0.05$)	Yes ($p < 0.05$)	Yes ($p < 0.05$)	Yes ($p < 0.05$)	No ($p = 0.10$)	No ($p = 0.11$)	No ($p = 0.26$)	

¹ no sub-sample contribution

We found variation in the crop and soil water conditions (SWC, LWP and CWSI) among fields, between fields as well as among different flights in 2021 (Table 4.4). The SWC varied by 1.90-fold for both

irrigated and non-irrigated fields in flight 1. The water condition of the last consecutive 14 days before flight 1 was very dry (SPEI 14D: -1.45). The LWP varied by 1.26 and 1.51-fold, whereas the CWSI varied by 1.6 and 1.92-fold in the irrigated and non-irrigated fields, respectively, in flight 1. In flight 2, the SWC varied by 1.65-fold in the irrigated field and varied by 1.62-fold, in the non-irrigated field. The water condition of the last consecutive 14 days before flight 2 was slightly dry (SPEI 14D: -0.24). The LWP varied by 1.41 and 1.31-fold where CWSI varied by 1.48 and 1.78-fold in the irrigated and non-irrigated fields, respectively, in flight 2. In flight 3, the SWC varied by 1.76-fold in the irrigated field and varied by 1.27-fold in the non-irrigated field. The water condition of the last consecutive 14 days before flight 3 was very wet (SPEI 14D: 1.45). The LWP varied by 1.45 and 1.17-fold in the irrigated and non-irrigated field whereas the CWSI varied by 2.19 and 1.6-fold in the irrigated and non-irrigated field, respectively, in flight 3. Variation in SWC within the field was statistically significant ($p < 0.05$) for both irrigated Airport field and non-irrigated Baxter field for all flights in 2021.

In flight 1, on which the water condition of the last 14 days was very dry, the mean SWC was significantly ($p < 0.05$) higher (1.31-fold) in the irrigated field compared to the non-irrigated whereas the mean LWP was significantly ($p < 0.05$) lower (15%) and mean CWSI was significantly ($p < 0.05$) and 1.18-fold higher in the non-irrigated field compared to the irrigated one. In flight 2, on which the water condition of the last 14 days was slightly dry, the mean SWC was 1.14-fold higher in the irrigated field compared to the non-irrigated whereas the mean LWP was significantly ($p < 0.05$) and 12% lower and mean CWSI was significantly ($p < 0.05$) and 1.16 -fold higher in the non-irrigated field compared to the irrigated one. In flight 3, on which the water condition of the last 14 days was very wet, the mean SWC was 1.06-fold higher in the irrigated field compared to the non-irrigated, whereas the mean LWP was 5% lower and mean CWSI was 1.10-fold higher in the non-irrigated field compared to the irrigated one. The differences in water conditions (SWC, LWP and CWSI) were statistically non-significant for flight 3. Like 2019 and 2020, we observed variation among different flights, with the highest mean SWC (24.71%) and

LWP (-1.01 MPa) recorded in flight 3 (very wet) on the irrigated field and the lowest mean SWC (14.98 %) and lowest mean LWP (-1.46 MPa) was recorded in flight 1 (very dry) and for the non-irrigated field. And the mean CWSI was observed highest in flight 1 for the non-irrigated field and lowest in flight 3 for the irrigated field.

Table 4.5 : Statistical differences in water condition-related variables including soil water content (SWC), leaf water potential (LWP), crop water stress index (CWSI) between irrigated (Airport) and non-irrigated (Baxter) wild blueberry fields and among different flights during 2019, 2020 and 2021. Statistical significance was tested with the linear mixed model with 95% confidence level.

Year	2019			2020			2021		
	SWC (%)	LWP (Mpa)	CWSI (#)	SWC (%)	LWP (Mpa)	CWSI (#)	SWC (%)	LWP (Mpa)	CWSI (#)
Field (Irrigated/ Non-irrigated)	0.17	0.73	<0.05	<0.001	0.17	0.10	<0.01	<0.001	<0.01
Flight (Developmental Stages)	<0.001	<0.001	<0.001	<0.01	<0.001	<0.05	<0.001	<0.001	<0.001

Variations in water condition-related traits (SWC, LWP, and CWSI) were determined mainly by the differences in flight date rather than the differences in field conditions (irrigated/non-irrigated) which was a relatively wet year (Table 4.5). In 2020 which was a slightly dry year, variation in water condition-related traits was also determined mainly by the differences in flight date rather than the differences in field conditions (irrigated/non-irrigated). 2021 was a dry year, and the variation in water condition-related traits was determined by both the differences in flight dates and the differences in field conditions.

4.4.2. UAV Thermal Sensor Based Remotely Sensed Canopy Temperature in Relation to Ground-Measured Leaf Temperature

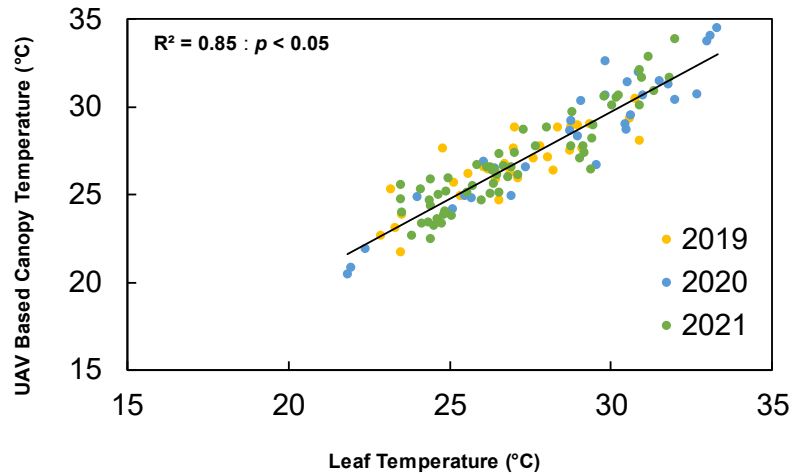


Figure 4.3: Unmanned aerial vehicle-based remotely detected canopy temperature (°C) in relation to handheld IR thermometer-based leaf temperature (°C) in three different years (2019, 2020 and 2021) for the irrigated Airport and non-irrigated Baxter field in Wyman’s blueberry field.

A significant and positive linear relationship ($R^2 = 0.85$, $p < 0.05$) was found between the UAV-based remotely detected canopy temperature (33% percentile of the coldest temperature) of the genotypes and the average leaf temperature derived from the handheld IR thermometer across dates, fields, and years of the irrigated and non-irrigated fields in 2019, 2020 and 2021 (Figure 4.3). The canopy temperature of the sampled genotypes varied from 20 to 35 °C.

4.4.3. Relationships Between UAV Thermal Sensor Based Crop Water Stress Index and Midday Leaf Water Potential

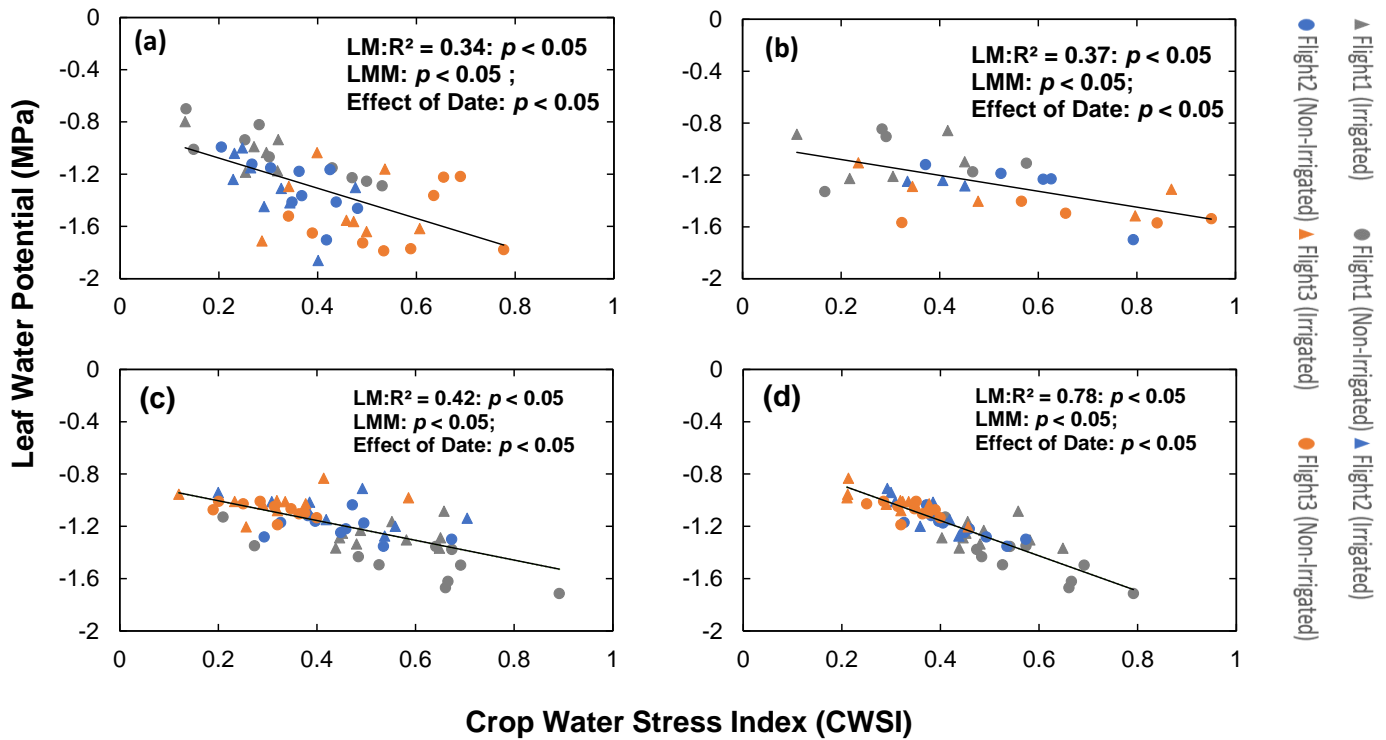


Figure 4.4: Field-measured Leaf water potential in relation to crop water stress index (CWSI) in 2019, 2020 and 2021. Statistical T_{wet} and empirical T_{dry} reference-based approach was taken to calculate CWSI in 2019 (a) and 2020(b), whereas in 2021, both statistical (c) and bio-indicator based (d) T_{wet} and T_{dry} reference approach were used. CWSI values are in the range of 0–1 with a higher value of CWSI indicating high water stress. LM represents linear model and LMM represents linear mixed model with flight/crop developmental stage as a factor.

We found significant linear relationships between UAV-based crop water stress index and midday leaf water potential. But the performance of the CWSI calculated based on the statistical T_{wet} and empirical T_{dry} reference approach was inferior to the CWSI calculated based on bio-indicator reference T_{wet} and T_{dry} in terms of predicting leaf water potential. We found significant and positive linear relationships between CWSI and LWP based on statistical T_{wet} and empirical T_{dry} reference approach the but lower coefficient of determination (Figure 4.4a : $R^2 = 0.34$, $p < 0.05$) and (Figure 4.4b : $R^2 = 0.37$, $p < 0.05$) for predicting LWP in 2019 and 2020, respectively. In 2021, both statistical and bio-indicator approaches were tested. The performance of the bio-indicator approach (Figure 4.4d : $R^2 = 0.42$; $p < 0.05$) was superior compared to

the statistical T_{wet} and empirical T_{dry} based calculated CWSI (Figure 4.4c: $R^2 = 0.78$: $p < 0.05$). When we used flight/ crop developmental stage as a random factor for a fitted linear mixed model, we also found the CWSI-LWP models were significant ($p < 0.05$) across years (2019, 2020 and 2021) though the effect of the different dates/crop developmental stages was on the LWP-CWSI models were also significant across years.

The performance of the bio-indicator based CWSI was also found to be better in differentiating differences in the water stress between irrigated and non-irrigated fields, especially when the water condition is dry. As we found a mean LWP (more negative) of -1.46 MPa (ranging from -1.71 to -1.13 to MPa) and mean higher CWSI of 0.58 (ranging from 0.41 to 0.79) for the non-irrigated field in flight 1 of 2021 which was very dry, whereas, for the irrigated field, the average leaf water potential was -1.26 MPa (ranging from -1.37 to -1.08 MPa) with an average CWSI of 0.49 (ranging from 0.49 to 0.65).

4.4.4. Leaf Water Potential Variability Maps

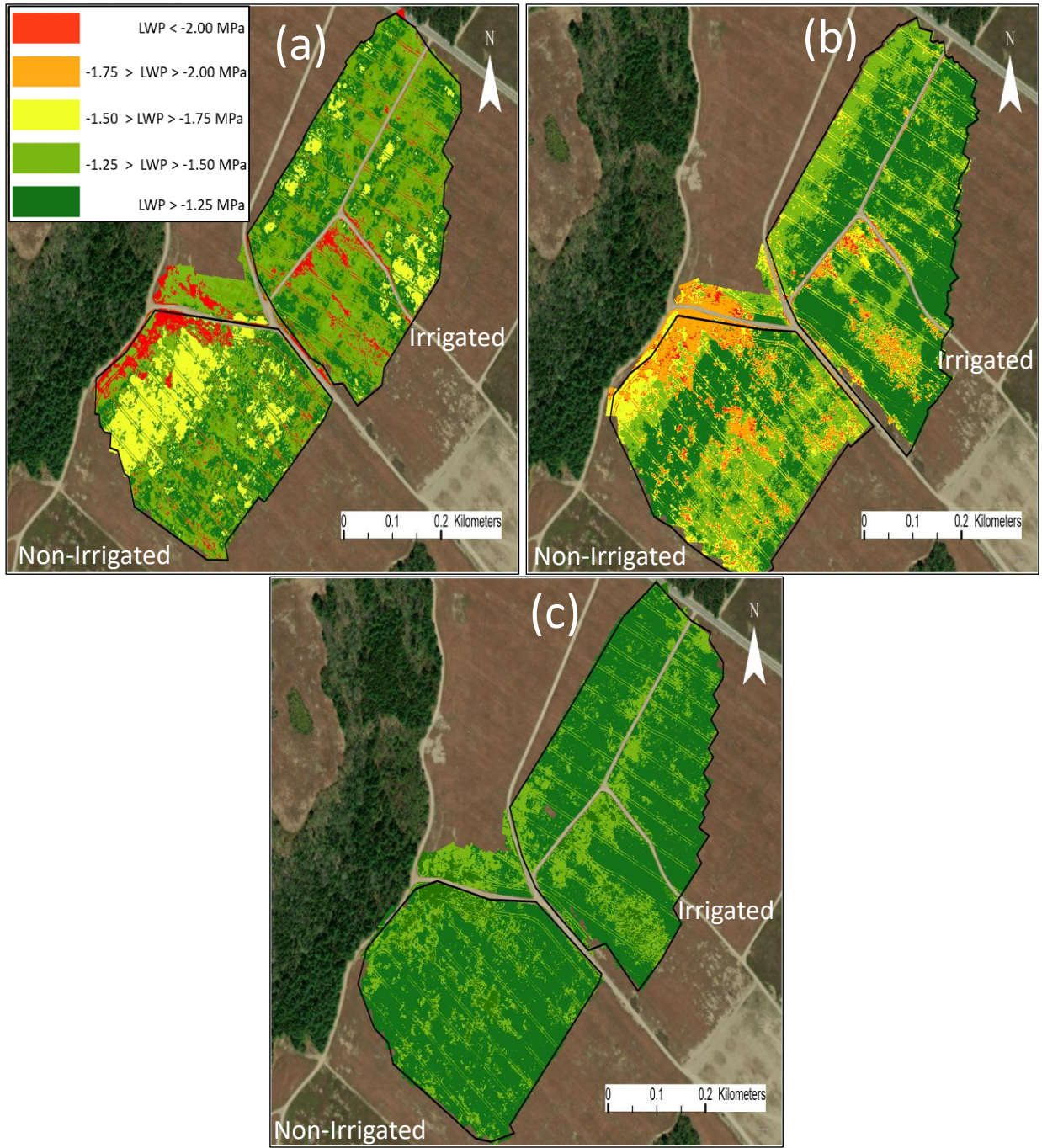


Figure 4.5: Predicted leaf water potential (LWP) variability map of adjacent irrigated (Airport) and non-irrigated (Baxter) fields in (a) flight 1, (b) flight 2, and (c) flight 3 of 2021. Calculated LWP was derived from the LWP-CWSI model.

In 2021, the water condition before the last 14 days of the first flight was very dry (SPEI 14D: -1.45), and in the predicted LWP variability map, we found high variability in LWP within the irrigated and

non-irrigated fields as well as between the irrigated and non-irrigated fields (Figure 4.5a). Though most of the part of the irrigated and non-irrigated field was without any water stress (Light Green: $-1.25 > \text{LWP} > -1.50$ MPa), there is a huge portion of the non-irrigated field, and some part of the irrigated field was in low water stress (Yellow: $-1.50 > \text{LWP} > -1.75$ MPa). We also found that some small parts of the irrigated and non-irrigated fields were in severe water stress (Red: $-2.00 < \text{LWP}$), which might be resulted from the unavailability of natural precipitation for a long time.

The water condition before the last 14 days of the second flight was slightly dry (SPEI 14D: -0.24) and in the LWP variability map, we can also see variability in LWP within the irrigated and non-irrigated field as well as between the irrigated and non-irrigated fields (Figure 4.5b). We found most of the part of the irrigated field was well watered (Light Green: $-1.25 > \text{LWP} > -1.50$ MPa) and over watered (Dark green: $\text{LWP} > -1.25$ MPa). A very few portions of the irrigated field were in low water stress (Yellow: $-1.50 > \text{LWP} > -1.75$ MPa) and in moderate water stress (Orange: $-1.75 > \text{LWP} > -2.00$ MPa). In the non-irrigated field, we found most of the part of the field was in optimum water condition and without any water stress (Light Green: $-1.25 > \text{LWP} > -1.50$ MPa) along with some low water stressed (Yellow: $-1.50 > \text{LWP} > -1.75$ MPa) area and moderately water stressed area (Orange: $-1.75 > \text{LWP} > -2.00$ MPa).

In flight 3 of 2021, the water condition before the last 14 days was Wet (SPEI 14d: 1.69). Though most of the part of the irrigated and non-irrigated field was in excess water condition Dark green: $\text{LWP} > -1.25$ MPa) and a very few portions of the irrigated field was in optimum water condition (Light Green: $-1.25 > \text{LWP} > -1.50$ MPa) (Figure 4.5c).

4.5 Discussion

4.5.1. High Spatial Variation In Soil and Crop Water Conditions

We found variation in soil water content (Table 4.2, 4.3, 4.4) within the irrigated and non-irrigated fields, which supports the presence of the spatial variability of soil water availability and soil water

retention capability within the wild blueberry field (Farooque et al., 2012). High variation in LWP within fields found in this study suggests the spatial variability of water availability and water need in wild blueberry genotypes within wild blueberry fields (Ihuoma & Madramootoo, 2017). The variation in soil water content between the irrigated Airport field and non-irrigated Baxter field suggests that the irrigation practice is effective. Overall higher LWP and lower CWSI in the irrigated field compared to the non-irrigated field also suggests that irrigation can provide water needs effectively. However, the variability of soil water content, LWP and CWSI within the irrigated field suggests that despite uniform irrigation management, the current irrigation system (Nelson Full-Circle Impact sprinklers) cannot provide uniform soil water content due to the variation in soil retention capability, which might be due to the spatial variability of soil properties in wild blueberry fields. We also found high variation in soil water content among different flights in different years, which is mainly influenced by the variation in natural water conditions and natural precipitations. This suggests high variation in crop water status can happen due to the unevenness of natural precipitation (Gu et al., 2021).

4.5.2. UAV Thermal Sensor Based Crop Water Stress Index Predicts Midday Leaf Water

Potential

The significant linear relationship between Infrared thermometer-based leaf canopy temperature and the UAV-based leaf canopy temperature confirms that UAV-based high-resolution thermal sensors can be successfully used to remotely detect canopy temperature for wild blueberries (Gonzalez-Dugo et al., 2014). When water is scarce, a plant slows its transpiration rate, which raises the temperature of its leaves relative to unstressed, well-watered plants (Gonzalez-Dugo et al., 2014). However, we found that the statistical T_{wet} and empirical T_{dry} based CWSI was inferior to the bio-based calculated CWSI in predicting leaf water potential in wild blueberries. The performance of the CWSI calculated from the bio-indicator based T_{wet} and T_{dry} was also better in differentiating the difference in water stress between irrigated and non-irrigated fields (Figure 4.4). CWSI calculated based on statistical T_{wet} and empirical T_{dry} reference has

been found effective in indicating crop water stress in arid or semi-arid conditions (Alchanatis et al., 2010; Gonzalez-Dugo et al., 2013; Rud et al., 2014), but we did not find it suitable for predicting water stress of wild blueberries in a humid summer weather with significant variability of weather conditions. The variability of the environment, including changes in temperature and vapor pressure deficit, can also affect the canopy temperature of fully transpiring leaves (T_{wet}) as well as fully non-transpiring leaves (T_{dry}) (Jones, 1999). Successful measurement of the leaf temperature of complete non-transpiring leaves as T_{dry} and full transpiring wet leaves as T_{wet} is very important for effective calculation of CWSI (Cohen et al., 2017). The use of canopy temperature of non-transpiring dry leaves as T_{dry} base and canopy temperature of real wet leaves as T_{wet} base was found effective for temperate humid regions (Jones, 1999), which aligns with our findings. Although CWSI was found to be a good index for water stress detection in our study, this index has some limitations. Though we found the CWSI-LWP models were significant ($p < 0.05$) across years (2019, 2020, and 2021) when we used flight/crop developmental stage as a random factor for the fitted linear mixed models, the effect of different dates/crop developmental stages was also significant. This suggests that the LWP-CWSI relationship differs among dates/development stages. CWSI also has some difficulty in its application for large areas with varying topography (Rahimzadeh-Bajgiran & Berg, 2016). If optical data are available in addition to canopy temperature, it is recommended to use indices such as water deficit index (WDI; a modified version of CWSI), soil adjusted vegetation index (SAVI), and transformed difference vegetation index (TDVI) for water stress detection based on the T_{canopy} /vegetation indices relationship concept as it provides a better estimation of stomata conductance, evapotranspiration and less sensitive to plant developmental stages (Rahimzadeh-Bajgiran & Berg, 2016). Future works are needed to test the effectiveness of these indices.

4.5.3. Spatial Variation in Leaf Water Potential, Its Impact and Recommendations

We found spatial variability in LWP within irrigated and non-irrigated fields, between irrigated and non-irrigated fields, as well as among three different flights of 2021, based on the predicted LWP maps constructed from the CWSI-LWP model (Figure 4.5). The variation between irrigated and non-irrigated fields was higher when natural precipitation was limited. A recent study found a significant reduction in stomatal conductance, photosynthesis, and transpiration rate before or at the turgor loss point (-2.00 MPa) (Pahadi, 2021). In mild drought conditions, the majority of plants may adjust their stomatal conductance to prevent low water potentials (Sperry et al., 2016). Turgor loss and xylem cavitation occur beyond a certain threshold of drought stress when plants are incapable of maintaining the balance between water loss and uptake (Mingeau et al., 2001). This can result in damage to the photosynthetic apparatus and xylem embolism, resulting in mortality (Pahadi, 2021). For pressure chamber measured LWPs, we did not find any genotypes reaching the suggested turgor loss point of -2.00 MPa. However, we found a large portion of the non-irrigated field and a small portion of the irrigated field showed LWP over -2.00 MPa (Flight1: 2021) and in the range of -1.75>LWP> -2.00 MP (Flight2: 2021) in the predicted LWP variability maps, which could potentially impact the major physiological processes of wild blueberries like stomatal conductance, photosynthesis, and transpiration. As physiological processes are sensitive to declining leaf water potentials, reduced biomass production and crop yield could happen. Moreover, the drought frequency in Maine is predicted to increase (Fernandez et al., 2020), which might affect the physiological processes of non-irrigated wild blueberry fields in the future, ultimately reducing yield and impacting the wild blueberry industry. Thus, we propose the introduction of precision agriculture including the precise management of water conditions to the wild blueberry fields by applying crop inputs at the right location with the right amount and at the right time to manage soil and water inputs efficiently and effectively. Precise water management can optimize water use, resulting in increased crop productivity (Harmon et al., 2005). Under conventional management, large wild blueberry farms get

irrigation applied uniformly. Introducing precision agriculture, wild blueberry fields can be divided into multiple management zones in a way that each zone receives inputs for management that are specifically tailored based on genotypes specific water use, vegetation density, soil properties, topography, and management history.

4.6 Conclusion

Global climate change is one of the major challenges for the agriculture sector. In recent years, short- and long-term drought has increased and is also predicted to increase further in the future, leading to decreased gross primary productivity, carbon storage, and yield of crops (D'Orangeville et al., 2018; McDowell & Allen, 2015). Increased drought conditions might affect the water conditions and increase the water stress of wild blueberries in the future, which can result in turgor loss and ultimately reduction in crop productivity. To be prepared for a future with more severe drought conditions, stakeholders of the wild blueberry industry should be prepared and adopt new technologies to monitor the crop water stress effectively. Effective irrigation technologies for wild blueberries should have the capability of monitoring spatial variability of the water status of a large field in real time to precisely manage the irrigation based on plant-specific needs. UAV-based thermal sensors are relatively cheap, and CWSI based on the UAV-based thermal sensor has the potential to effectively monitor crop water status remotely. By detecting the temporal-spatial heterogeneity of crop water status, irrigation can be intelligently controlled by adopting precision irrigation management systems to minimize crop water stress and maximize overall production.

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3. Barai, K., Tasnim, R., Hall, B., Rahimzadeh-Bajgiran, P., & Zhang, Y. J. (2021). Is Drought Increasing in Maine and Hurting Wild Blueberry Production?. *Climate*, 9(12), 178.
4. Chen, Y. J., Maenpuen, P., Zhang, Y. J., Barai, K., Katabuchi, M., Gao, H., ... & Zhang, J. L. (2021). Quantifying vulnerability to embolism in tropical trees and lianas using five methods: Can discrepancies be explained by xylem structural traits?. *New Phytologist*, 229(2), 805-819.

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BIOGRAPHY OF THE AUTHOR

Kallol Barai was born in Shoshikor, Bangladesh on November 14, 1995. He was raised in Barisal, Bangladesh and graduated from Torkey Bandar Victoria High School in 2010. He attended the Shahjalal University of Science and Technology and graduated in 2018 with a Bachelor's degree in Genetic Engineering and Biotechnology. He entered the MS in Botany and Plant Pathology program at The University of Maine in the fall of 2019. After receiving his degree, Kallol will be joining the same University for PhD in Plant Science. Kallol Barai is a candidate for the Master of Science degree in Botany and Plant Pathology from the University of Maine in August 2022.