

MULTI-ENVIRONMENT EVALUATION OF WINTER PEA GENOTYPES FOR WINTER  
SURVIVAL AND YIELD STABILITY

by

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## DEDICATION

To my beloved mother, father, and sister whose love and guidance have been my constant light, I am eternally grateful. Your unwavering belief in my abilities and your endless support have shaped the person I am today. Thank you for always believing in me and encouraging my academic pursuits, helping me to make this journey smoother and more joyful. To my friends, Pragya and Shreejana, who provided their constant encouragement and support, for that I will always be grateful.

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## ABSTRACT

Winter pea can be grown as a rotational crop for soil moisture conservation and nutrient recycling in the wheat-growing region of Montana. Development of winter hardy cultivars would increase seed yield and expand the area of adaptation of this crop. Harsh winter conditions present a significant challenge to the production of winter peas. The objective of this study was to screen pea germplasm and breeding lines for winter survival and identify genotypes with good winter hardiness for future crop production. Field trials were conducted to evaluate genotypes at Bozeman, Havre, Huntley, and Moccasin, MT in 2021, 2022, and 2023. These lines included elite winter cultivars and several checks. Winter hardiness was evaluated as the percentage of surviving plants and by agronomic performance including yield. Genotypes were evaluated based on the GGE biplot method. This analysis captured multiple variables including yield, protein content, seed size, and their overall stability across multiple years and locations of study to aid in selecting lines. Differential winter survival was observed across locations and years. Higher winter survival was seen in Bozeman and Havre. Few lines were identified as having high seed yield and stable production over years and locations. Breeding lines had higher mean yield with few good lines having stable production of greater than 2500 kg/ha. Germplasm lines showed better winter survival than breeding lines. Protein content ranged from 20% to 31%. Larger seeds were observed in Moccasin, whereas Havre had the highest protein content. Mega-environment differentiation helped to select specific genotypes based on the trait of interest for a particular environment. Several European and US lines used in the experiments having high winter hardiness record performed better for seed yield and resistance to stress. The lines identified as having high levels of cold tolerance can be used as a prospective genetic resource in pea breeding programs. Genotypes having high and stable seed yield can be considered for release as a variety and made available to producers.

## INTRODUCTION AND LITERATURE REVIEW

### Introduction

The pea (*Pisum sativum* L.) is an important crop of the Leguminosae family that has been cultivated and consumed by humans for thousands of years. It is one of the earliest domesticated crops with archaeological evidence suggesting their cultivation as early as 8000-9000 years ago in the Middle East (Zohary et al., 2012).

Peas are a rich source of essential nutrients, including protein, dietary fiber, vitamins, and minerals, making them an important component of diets worldwide (Hacisalihoglu et al., 2021). Peas are cultivated globally, with major production regions including Europe, Asia, and North America (Carlson-Nilsson et al., 2021). They are grown for both human consumption and animal feed, contributing significantly to food security and livestock nutrition. Leguminous crops like peas fix atmospheric nitrogen through a symbiotic relationship with rhizobia bacteria and reduce the need for synthetic nitrogen fertilizers and improve soil fertility (Kebede, 2021). Incorporating peas into a cereal-based cropping system has become crucial due to their ability to serve as green manure, restoring fixed nitrogen and various nutrients back into the soil (Ghosh et al., 2007).

Dry pea production in the United States increased by almost 14% from 2022 to 2023 (USDA, 2023). In the same year, Montana experienced a 27% and 10% increase in dry pea production and area planted, respectively. Due to the harsh winter conditions, pea production in Montana is almost exclusively spring-sown. Despite the agronomic benefits the fall-sown winter pea offers, the acreage is very limited. Winter pea accounts for only about 2% of total pea acreage in Montana (USDA, 2023). Currently, winter pea is grown on about 5,000 hectares in the United States for seed production, of which about 2,000 hectares are in Montana (USDA, 2023).

However, most of the winter peas grown in the mild winter areas like North Carolina, Idaho and Oregon are used as cover crop, forage or green manure (McGee et al., 2017).

Winter peas planted in the early autumn confer most of the advantages of spring-planted peas in addition to more benefits. Fall planting would avoid fieldwork in wet spring conditions and the plants develop earlier and escape summer heat and drought. The addition of winter peas in the crop rotation would replace the winter fallow period, thus providing soil stability and aggregation, and additional nitrogen by atmospheric nitrogen fixation. This reduces the use of fertilizer requirements not only for the peas but also for the succeeding cereal crop. Incorporation of winter peas in the crop rotation will help break disease and weed cycles and reduce the occurrence of diseases (McGee et al., 2017). While it is not evident that the winter pea outperforms the spring pea in production under all conditions, Chen et al., (2012) observed an increase in wheat yield following winter pea in Montana indicating a potential advantage to the cropping system.

There are very few winter pea cultivars that have been released and even fewer of those can survive in Montana growing conditions. The advancement of winter hardy pea breeding faces a unique challenge, primarily from the necessity of maintaining consistent winter conditions for rigorous selection processes. These conditions are essential to identify and develop winter-hardy pea lines. The use of alternative screening methods with controlled environments, such as freezing chambers have been used to simulate winter conditions (Fiebelkorn, 2013). However, these controlled environments may not capture all the nuances of field conditions. Developing winter pea varieties that can withstand these colder temperatures would allow for the

expansion of field pea production in the Northern Great Plains (NGP) and the Pacific Northwest (PNW).

A better understanding of environmental factors and agronomic practices that control winter hardiness in peas is needed to obtain winter hardy pea varieties. Delayed planting date usually correlates with lower yield of winter peas (Chen et al., 2006; Knott & Belcher, 1998). It is advisable to plant in early September in the NGP to allow for good stand establishment and provide enough acclimation time to survive the harsh winter. It is still unclear whether a higher seeding rate would compensate for the winterkill. The higher plant density normally hinders branching and results in poor plant growth (Knott & Belcher, 1998). To better manage winter hardiness in peas, greater understanding of the underlying genes is necessary. Lejeune-Hénaut et al. (2008) reported a flowering locus, *Hr*, that delays floral initiation during winter conditions and identified six quantitative trait loci (QTL) responsible for frost tolerance. The presence of consistent QTLs can be used for marker-assisted selection (MAS) and be used to reduce the time to develop a new variety.

### Objectives

The goal of this project was to improve pea production through the development of improved winter pea varieties for producers. This research aimed to screen pea genotypes for winter survival at four locations in Montana and identify genotypes with greater winter hardiness for future crop production. This will also help better understand the underlying environmental factors and the inter-relationship of those factors as they impact the production of winter pea.

## Literature Review

### Origin and Domestication

Domestication of pea (*Pisum sativum* L.) started in the Fertile Crescent that includes present day Turkey, Syria, Iraq, Israel, and Lebanon in 7000-6000 B.C. (Zohary & Hopf, 1973). The origin is further expanded to the area of the Mediterranean area around Southern Europe, and later to Northern and Western Europe (Smýkal et al., 2012). Reports suggest that the domestication of different pulse crops including pea occurred along with the domestication of wheat (*Triticum aestivum* L.) and barley (*Hordeum vulgare* L.) (Zohary & Hopf, 1973). Pulse crops are an important part of the agriculture system developed in the Fertile Crescent, contributing to crop rotation, soil fertility, and dietary diversity, thus helping to develop a permanent settlement in that area.

*Pisum* is a member of the family Fabaceae with subfamily Papilionaceae, and tribe Viciae. *Pisum* is comprised of two species, *Pisum fulvum* Sibth. and Sm. and *Pisum sativum* L. *Pisum fulvum* is comprised of relatively small plants with yellow-brownish flowers and tiny pods (Muehlbauer, 1992). *Pisum sativum* is divided into the subspecies *sativum*, *elatius*, *humile*, *arvense*, and *hortense*. The subspecies *elatius*, has contributed genetic diversity to cultivated pea through both natural and human-facilitated hybridization, providing genes for disease resistance and environmental adaptability. *Pisum abyssinicum* was once used to describe a separate type that is native to Ethiopia but is now considered a subspecies of *P. sativum* (Smartt, 1990).

Pea can be grown in a broad range of climatic conditions. It is typically grown in the cooler and semi-arid areas of the world. There are reports of successful pea cultivation from temperate to the Arctic regions (Carlson-Nilsson et al., 2021). As more pea varieties are

developed with resistance to more abiotic and biotic stresses, the geographic regions suitable for pea cultivation are expanding. Plants combat heat stress by boosting their ability to photosynthesize and transport electrons, while also increasing their antioxidant activity through enzymes like peroxidase, superoxide dismutase, and catalase

### Pea Production

Field pea is widely cultivated for both human consumption and livestock feed globally. In 2021, a total of 7.0 million hectares of field pea were harvested globally. The top producers consist of Canada, Russia, China, and India (FAOSTAT, 2023). Cultivated land acreage for field peas has been steady for the past 20 years on a global scale.

Dry pea production in the U.S. has fluctuated over the years with area dedicated to dry peas remaining steady in recent years. Between 2019 and 2023, the area sown to dry peas in the United States was about 400,000 hectares (USDA, 2023). Montana, North Dakota, and Washington are the leading states for pea production (USDA, 2023). In 2022, Montana had the highest area harvested and production with 206,390 hectares and 277,228 metric tons, respectively, followed by North Dakota with 84,984 hectares and 140,824 metric tons. Winter pea has even less acreage than spring peas. Most of the winter pea hectareage is for cover cropping, and only a small portion of winter pea is currently used for the seed production. Austrian winter pea dedicated to seed production is planted in about 5000 hectares in the U.S. (USDA, 2023).

Field pea production is very small compared to global cereal production. Grain legumes and peas are only a minor part of most diets, and legume crops are underutilized. The low expansion of pulses is due to lower and uncertain yields, which are caused by their higher

susceptibility to environmental stresses as compared to cereal crops (Foyer et al., 2016). Due to low expansion and slower growth in production, the price of pulses is higher than cereals (Joshi & Rao, 2017). Pulse crop yield growth rate is also much lower than that of cereals. Globally, cereal yields increased by 3-fold in the last 50 years while pulses increased their yield by barely 60% (S. Y. Yang, 2018). Cereal yield increased both in developed and underdeveloped countries while pulse yield increased mainly in the developed countries. After the late 1990s, North America saw a greater expansion of pulse crops than any other region (Kumar & Parthasarathy, 2016). Legume breeding programs are relatively small, often forcing breeders to focus on a limited number of traits and crops. This has resulted in slower progress in improving legume varieties compared to cereals (Rubiales et al., 2021).

### Uses and Market Classes

Pea is primarily used for human food and livestock feed. It is commonly used throughout the world in human diets. It is also used as a cover crop in soil improvement and industrial uses. Mature peas are also used as animal feed, particularly when the quality standard for human consumption is not met (Stein et al., 2016). Peas contain high levels of carbohydrates and total digestible nutrients. The presence of high digestible nutrients makes them a great livestock feed (Schatz & Endres, 2003). Peas also provide various mineral nutrients like calcium, iron, potassium, phosphorous, sodium (Muehlbauer & McPhee, 1997), and selenium (Thavarajah et al., 2010).

Field pea market grades are determined based on important visual traits of the seeds, such as their shape, size, and the color of the seed coat and cotyledon. Peas are consumed in both green and mature forms. Fresh green types include freezing and canning. Dry seed types include

green and yellow cotyledon peas. A wide range of market grades of field peas is determined by the general appearance. Color is the major component that drives the market. Most of the time, the color is determined by the color of the cotyledon, not the seed coat. Green and yellow are the two main types of field peas produced. The most widely grown spring dry peas, including both green and yellow types, are planted in the spring and harvested in the summer. The U.S. market is more inclined towards green peas, while India is a big consumer of yellow peas (Janzen et al., n.d.). The edible pod types are harvested when the pods are young and succulent. These are classified into snow peas and snap peas. Snow peas are harvested when the seeds are very small, and the pods are flat whereas snap peas are consumed when the pods are more mature (Smykal, 2019). Marrowfat peas are used to produce dried snacks like wasabi peas and are large, irregularly shaped peas with a distinct appearance. Austrian winter pea is a market class that is planted in the fall and harvested the following summer. According to the Food and Agriculture Organization of the United Nations, in 2021, fresh green peas accounted for 62% of global pea production, while dry peas accounted for 38% (FAOSTAT, 2022).

### Agronomic Benefits

Peas are nutritious and versatile legumes that offer many agronomic benefits. They do not require additional nitrogen fertilizer for growth which makes them an economic and environmentally friendly crop (Graham & Vance, 2003). Peas establish a symbiotic relationship with rhizobia bacteria and fix atmospheric nitrogen. *Rhizobium* is a genus of Gram-negative bacteria that fixes atmospheric nitrogen. One of the most important bacteria that colonize peas is *Rhizobium leguminosarum* biovar *viciae* offers the opportunity to reduce the use of synthetic fertilizers (Rennie & Dubetz, 1986). *Rhizobium leguminosarum* biovar *viciae* also forms a

symbiotic relationship with other crops like lentils and vetch, facilitating the conversion of atmospheric nitrogen into a form that the plants can readily absorb and utilize. Colonization is triggered by the flavonoids from plant root systems during nitrogen-deficit conditions by inducing transcription of nodulation genes in the bacterium that signals the host plant to develop nodules (Long, 1996).

The impact of peas on soil nitrogen levels provides significant benefits in sustainable agricultural practices. Peas contribute to soil health by leaving behind residual nitrogen. According to a research (Schatz & Endres, 2003), for 100 kg peas produced per hectare, there is an addition of 0.5 kg of residual nitrogen, available for the next planting cycle. This process enriches the soil, making peas a valuable crop in rotation systems for soil fertility enhancement and reduction of synthetic fertilizers.

Among legumes, field peas are recognized for their stable biological nitrogen fixation ability across different years. Studies, including those by Hossain et al., (2017) and Bruce (2018) highlight that peas can fix approximately 55 kg of nitrogen per hectare. This ability not only supports the pea crop by fulfilling a significant portion of their nitrogen requirement but also contributes to a sustainable farming ecosystem. Dhillon et al., (2022) reported that in Canadian environments, the nitrogen fixation capacity of peas ranges from 50% to 55% of the total nitrogen needed by the crop. This underscores the efficiency of peas in utilizing atmospheric nitrogen, reducing reliance on external nitrogen inputs, and promoting a more sustainable agricultural practice.

Crop rotations are crucial for maintaining soil moisture and nutrient levels as well as in controlling the development of pests and diseases (Krupinsky et al., 2002). The introduction of

legumes like peas in the cropping system improves plant nutrient uptake through mycorrhizal associations and decrease disease development (Lupwayi & Kennedy, 2007). Legumes also lower greenhouse gas emissions compared to fertilized monoculture practices (Lupwayi & Kennedy, 2007). A broadleaf crop like pea in the crop rotation with cereals helps to break the disease cycle of cereal pathogens. Growing wheat after pea reduces foliar disease of wheat compared with wheat monocropping (Bailey et al., 2001). The severity of wheat leaf blotch disease in wheat was found to be 20% less when pea was introduced in the crop rotation (Jalli et al., 2021).

A study by Chen et al., (2012), found the introduction of field peas in winter wheat-based cropping systems produced greater wheat grain yield and protein content at lower nitrogen input. Winter wheat yield in rotation with peas was found higher than a spring wheat-winter wheat system (Chen et al., 2012), thus improving the system's profitability. Another similar study in Nebraska showed that pea-wheat rotation systems resulted in greater net profit compared to a fallow-wheat system (Koeshall et al., 2022). A study also found the addition of field pea cover crops in the cropping system increased the production of subsequent Sudan grass (Cupina et al., 2011).

Inclusion of legumes in the cereal-based cropping system has been shown to improve the uptake of nitrogen and phosphorus in subsequent cereals (Yu et al., 2021). Nitrogen uptake can be increased by 49% in pea-wheat rotations compared to the wheat continuous cropping (Mesfin et al., 2023). Pea included in the rotation enhanced phosphorus uptake in a subsequent wheat crop (Nuruzzaman et al., 2005).

Field pea used as a cover crop helps to reduce soil erosion, nitrogen leaching and conserves soil aggregation (Decker et al., 1994; Blanco-Canqui et al., 2013). It has been shown that the use of field peas as a cover crop decreased the number of harmful fungi like *Fusarium oxysporum* and *Rhizoctonia solani* (Jamiołkowska, 2006). Field peas grown as a cover crop helped to suppress weed growth and competition in the corn in reduced tillage conditions (Büchi et al., 2020) and provided additional soil nitrogen for organic potato production (Wilson et al., 2019). The use of winter peas as a cover crop also suppressed weed dry matter linearly with an increased seeding rate of the peas (Brennan et al., 2009). Spring planting normally results in soil compaction while using equipment in the wet field during the spring. Fall planting, on the other hand, reduces this risk, and plants develop rapidly.

When grown in rotation with cereals, field peas can reduce crop water use. Field peas grown in rotation with cereals reduced crop water use by 20-30% (Gufi et al., 2022). This finding is supported by research conducted in Western Canada, where increasing the frequency of field peas in the cropping system is found to reduce moisture requirement for the subsequent crop (Fleury, 2012).

Field peas, as high nitrogen fixers, contribute to short-term soil conditioning. The succulent stems of field peas break down easily, providing a quick source of available nitrogen. Field peas, as green manure, produce 2-3 tons of dry matter per acre and increase the microbial biomass phosphorous content by enhancing the P-related enzymatic activity (Piotrowska-Długosz & Wilczewski, 2020).

## Genomic Structure of Pea

The pea reference genome, based on the cultivated variety ‘Cameor’, was recently completed. The estimated size of the pea genome is approximately 4.45 gigabase pairs (Gb) making it one of the largest among land plants and within the upper range for the superrosid eudicots (Kreplak et al., 2019).

The pea genome is organized into seven pairs of chromosomes, with a total of 14 chromosomes ( $2n=2x=14$ ). The karyotype shows two sub-metacentric and five acrocentric chromosomes. The chromosomes in pea have extended primary constrictions, which are attributed to the presence of highly repeated sequences (Macas et al., 2007)). These sequences can be categorized into two main types: tandem repeats (linear) and interspersed repeats (satellite DNA) (Mehrotra & Goyal, 2014). Approximately 83% of the pea genome consists of repetitive sequences which include microsatellites (or simple sequence repeats, SSRs), transposable elements (TEs), retrotransposons, and repetitive DNA. The remaining 17% of the genome consists of unique sequences, which include genes, regulatory sequences, and non-repetitive DNA (Macas et al., 2007). Understanding the presence and nature of repetitive sequences in the pea genome is essential for various areas of plant biology research, including genome assembly, evolution, and understanding gene regulation (Sato et al., 2008).

Comparative genomics studies have revealed synteny and collinearity between the pea genome and related legume genomes, such as *Medicago truncatula* and *Lotus japonicus* (Sato et al., 2008). In the research of translational genomics of pea with *M. truncatula* using SNPs to draw a functional consensus map, Bordat et al. (2011)) found higher syntenic conservation with pea compared to other legumes. This finding indicates that *M. truncatula* is a suitable legume to

use in syntenic studies with pea. These comparisons help researchers understand the evolutionary relationships and conservation of genomic elements.

### Winter Hardiness

Winter hardiness is a crucial trait that directly impacts a crop survival and health in cold climates. It includes a range of physiological and biochemical adaptations that enable plants to withstand freezing stress including ice-encasement, snow molds, and environmental challenges associated with winter conditions (Mckersie & Leshem, 1994). The important aspects of these adaptations are mechanisms aimed at preserving cellular integrity, maintaining metabolic activity at minimal levels, and protecting the plant from the deleterious effects of freezing temperatures. Snow cover, soil and air temperature are other major components of winter survival (Eteve, 1985). It is a complex trait that is not clearly understood compared to other traits, however, progress has been made in the study of winter hardiness and the development of winter hardy crops.

Many temperate plants that overwinter will be more tolerant of a subsequent cold if they experience a low positive temperature period, the process called cold acclimation. This ability to endure cold or freezing temperatures shows biochemical, structural, and physiological changes that result in improved defense against the consequences of frost damage (Thomashow, 1999). Acclimation is the process where plants develop freezing tolerance when exposed to chilling temperatures (Levitt, 1980). Freezing tolerance in plants is a major trait that determines the ability of winter survival (D. B. Fowler et al., 1999). It involves a slow transition to low temperatures. One of the key aspects of acclimation involves biochemical modifications, including the accumulation of soluble sugars, proline, and other osmoprotectants (Hughes & Dunn, 1990). These compounds act as cryoprotectants, lowering the freezing point of cellular

fluids, stabilizing membranes and proteins, and protecting cells from dehydration caused by ice formation.

### Winter Injury Quantification

Winter injury in crops is a crucial concern for breeders, agronomists, and farmers alike. As the global climate is becoming increasingly unpredictable, understanding the extent and causes of winter injury is essential for ensuring food security. Winter crops, like winter wheat, barley, and winter peas, are bred and cultivated to endure colder temperatures. However, they can still suffer from various winter stresses such as freeze-thaw cycles, direct cold damage, ice encasement, desiccation, and snow mold infections. For example, snow mold, a fungal disease that proliferates under snow cover, can devastate crops, leading to substantial yield losses (Ponomareva et al., 2021).

Quantifying winter injury is a multifaceted endeavor. Historically, field evaluation through visual scoring has been the mainstay method, where disease symptoms such as necrosis or reduced growth are scored on a certain scale, often 1-9 (Munkvold et al., 2001) or 1-5 (Lejeune-Hénaut et al., 2008). These types of scoring provide an immediate and tangible measure of plant health post-winter. The percent survival method is another way of assessing winter damage. Seedlings are counted on both fall and spring to get the percent survival (Auld et al., 1983). However, with the emergence of diseases as major contributors to winter injury, tools like the Area Under the Disease Progress Curve (AUDPC) have gained prominence. The AUDPC provides an aggregated measure of disease intensity across multiple assessment dates, offering insights into disease progression over time (Jeger & Viljanen-Rollinson, 2001). It's particularly useful when tracking diseases like snow mold, which may exacerbate over the winter

season (Shaner, 1977). Besides these, biochemical assays, and even molecular and genetic tools, are now being deployed to understand winter injuries better. Despite the variety of methods available, the challenge remains in ensuring accurate quantification that encompasses all factors, both biotic and abiotic, that contribute to winter injury.

### Management Practices

The research for the best agronomic practices for winter peas is limited. However, selecting cold-tolerant and disease-resistant varieties based on regional performance is a starting point for better survival and higher yield. Severe winters often damages the apical meristem, but branching indicates the survival of the plant. Standing stubble or crop residue is necessary for trapping snow and reducing plant death (Rezgui et al., 2020). Crop residues improve soil water management by boosting infiltration and reducing runoff loss. Although the study by Chen et al., (2006) did not find the influence of stubble height on the biomass or yield of winter peas, having stubble on the field helps to catch the snow and also prevent soil erosion early in the season (Gao et al., 2016). However, it has also been observed that the high stubble and reduced tillage have increased snowmelt runoff (J. Liu & Lobb, 2021). Further research is needed to evaluate the agronomic and environmental trade-offs of crop residue management.

Winter peas thrive in well-drained clay to sandy loam soils, with a pH range of 6.5-7. While peas can fix nitrogen from the atmosphere, applying a starter phosphorus fertilizer can be beneficial, especially in soils with low phosphorus content. Biological nitrogen fixation meets 60% of the total nitrogen demand while seed inoculation is found to increase the nitrogen derived from air and also an increased aboveground biomass (Enrico et al., 2020).

Planting at the right time is another crucial agronomic practice. It is advised to plant in early September for good germination and to get a better plant stand. Mid-September is considered a good time to sow winter peas in Montana. However, for the Pacific Northwest where the climate is milder, planting winter peas in late September has also given a considerable yield. The cooler climate and severe winters of Montana require earlier planting for good stand establishment and survival. Reduced yield can be seen at delayed planting dates, especially in the Northern Great Plains and Pacific Northwest because the seedlings do not get adequate time to acclimate before the winter (Chen et al., 2006). However, Urbatzka et al. (2012), did not find any impact of sowing date on the extent of winter hardiness, but the highest biomass was found for the earliest sowings. There has not been a universally accepted planting depth, and no consensus has been reached on the correct depth. Some studies have revealed no difference in yield for peas planted at different depths (S. V. Stepanovic et al., 2018), however, germination time heavily relies on the moisture availability in the soil (Machado et al., 2008).

### Physiology of Winter Hardiness

Winter hardiness is a critical trait for crops growing in cold conditions where winter temperatures are severe. Plants have two main strategies for dealing with freezing stress: they can either survive through dormancy mechanisms (freezing-avoidant) or tolerance (freezing-tolerant). The freezing-tolerant plants develop their capacity to tolerate freezing by being exposed to low temperatures called cold acclimation (Thomashow, 1999). In the absence of acclimation or in instances of abrupt temperature drops, intracellular freezing ensues, giving rise to the crystallization of ice within plant cells, ultimately resulting in cellular rupture. Instead during acclimation, plants undergo a series of changes in physiology and biochemistry that

support them to thrive in the impending winter conditions. In the acclimation stage, the plants produce specific lipids and protein concentrations to protect from freezing injury (Kawamura & Uemura, 2013). One of the first mechanisms to avoid freezing damage is the alteration of gene expression. Many genes are activated to help the plant increase its freezing tolerance. The most well-studied genes in cold acclimation are the CBF (C-repeat Binding Factor) genes which are the part of CBF regulatory pathway that induce the expression of cold-responsive (COR) genes (Stockinger et al., 1997). COR genes like *COR15a*, *COR15b*, *COR47* and *KINI* are involved in stabilizing cell membranes during freezing (Yamaguchi-Shinozaki & Shinozaki, 1994).

There are also reported relations between the abscisic acid (ABA) concentrations and freezing tolerance in plants. Mckersie & Leshem (1994) found an increased concentration in ABA in freezing tolerant *Arabidopsis thaliana*. Higher levels of ABA are also linked to expression of stress-responsive genes and proteins, regulating membrane transport and biosynthesis of osmoprotectants and antioxidants, which together help plants to tolerate cold stress (Gusta et al., 2005; Thomashow, 1999). Accumulation of osmoprotectants such as proline, sugars (e.g., sucrose, raffinose and fructans) and other solutes help plants maintain water balance and prevent dehydration (Close et al., 1993). These molecules lower the freezing point of the cell sap and protect cellular structures by preventing or reducing ice formation (Close et al., 1993). ABA levels are also found to induce maximum accumulation of proline which inhibits callus growth and increases tolerance to cold (Duncan & Widholm, 1987). In herbaceous plants, the hardening and de-hardening cycle is primarily controlled by temperature where resistance to low temperature is better during cold, and the process is reversed when the temperature rises (Levitt, 1980). Junttila (1996), studied the effect of photoperiod on frost resistance in white clover and

suggested that the hardening is enhanced by short and de-hardening by long photoperiod. The same author also explained that some genes are responsible for producing antifreeze proteins and synthesis of other compatible solutes.

Genetic engineering can be used to make freezing-susceptible crops tolerant to freezing conditions by incorporating genes and proteins from freezing-tolerant plants (Gupta & Deswal, 2014). Antifreeze proteins (AFPs) found in freezing-tolerant plants are a good freeze-tolerant source for use in freezing intolerant plants. The AFPs in the apoplast of many gymnosperms, dicots and monocots provide freezing tolerance by inhibiting the growth of ice crystals within plant tissues during sub-zero temperatures (Gupta & Deswal, 2014). These AFPs are reported to lower the freezing temperatures of freezing-sensitive plants by ~1 degree Celsius (Griffith & Yaish, 2004). The photoperiod response is also found to regulate the expression of cold-tolerant genes by affecting plant development rate (Mahfoozi et al., 2001).

Reactive oxygen species (ROS) are the byproducts of cellular metabolism. These molecules are highly reactive and are involved in different biological processes like stress responses and signal transduction. In plants, ROS plays a key role against various abiotic stresses, including cold stress. Crops growing in cold areas also suffer oxidative stress due to the uncontrollable production of ROS. An enzymatic system has developed in plants to protect from oxidative stress by scavenging the ROS (Baek & Skinner, 2012). Many anti-oxidant genes increase in their expression during low temperature and induce freezing tolerance (Baek & Skinner, 2012). Another study investigated the role of ROS in the winter hardiness of winter peas by examining the expression of ROS-scavenging enzymes. The authors found that the expression of several ROS-scavenging enzymes increased during the acclimation process, suggesting that

ROS are produced and scavenged during cold acclimation in winter peas (X. Liu & Huang, 2000). In addition to their response to stress, ROS also plays a role in the accumulation of cryoprotectants in winter pea. Cryoprotectants are compounds that protect plant tissues from freezing damage. Studies have found that the accumulation of cryoprotectants in winter pea was associated with an increased level of ROS production (Kosová et al., 2011; Taylor et al., 2005).

### Vernalization

Vernalization is a process where the plant is exposed to a certain period of cold temperature to initiate flowering. The vernalization requirement for winter peas is still unknown. It is unclear to which level of vernalization is necessary for the development of cold tolerance. Trevino & Murray (1975) found that vernalization inhibited pea dry matter production and also slightly affected the seed yield. Their result also showed accelerated flowering due to vernalization.

For winter wheat, exposure to cold temperatures needed for the vernalization response varies between 2-6 weeks and can even go up to 12 weeks (L. Yan et al., 2015). During this time, the plant undergoes a series of physiological changes, such as an increase in sugar content, that help it tolerate freezing temperatures and other stresses associated with winter conditions (X. Liu et al., 2014).

Vernalization control not yet well-understood in pea, but details have been worked out in wheat. One of the key genes involved in vernalization in wheat is *VERNALIZATION1 (VRN1)*, which is activated in low temperatures and is involved in flowering time regulation (Trevaskis, 2010). *VRN1* is expressed in the leaves and the apical meristem, where it subsequently promotes flowering. It encodes MADS-box transcription factor which promotes shifting vegetative to

reproductive stage (Trevaskis et al., 2003). The vernalization requirement is controlled by allelic variation in the *VRN1* gene (Brule-Babel & Fowler, 1988). Distinct *VRN1* homeologs are known in wheat, including *VRN-A1*, *VRN-B1*, and *VRN-D1* (Li et al., 2013). Winter wheat cultivars have vernalization requirement alleles for each vernalization homeolog (Li et al., 2013). *VRN2* acts as a repressor of *VRN1* and regulates flowering time (Distelfeld et al., 2009). In the absence of vernalization, *VRN2* prevents *VRN1* expression and keeps the plant in the vegetative phase (Distelfeld et al., 2009). *VRN3* encodes the protein that regulates flowering and is upregulated during vernalization functions downstream of *VRN1* to induce flowering (Faure et al., 2007).

### Genetics of Winter Hardiness

Winter hardiness in plants is a complex trait controlled by many genes. Quantitative trait loci (QTL) mapping and genetic analysis have identified several regions of the pea genome that are associated with winter hardiness. These regions contain genes involved in different physiological processes in plants such as growth and development, stress response, and carbohydrate metabolism.

One of the key genes involved in winter hardiness in peas is the CBF (C-repeat binding factor) transcripts. These appear to serve as core regulators of cold signal transduction and initiation of cold acclimation (J. Liu et al., 2021). CBF transcripts encode a transcription factor that regulates the expression of genes involved in cold acclimation. The CBF gene has been upregulated in response to cold temperatures and is involved in activating other genes that confer cold tolerance (S. Fowler & Thomashow, 2002). In *Arabidopsis*, 306 genes were cold-responsive with transcripts for 218 genes upregulated and 88 genes downregulated during cold (S. Fowler & Thomashow, 2002). A genome-wide association study by Beji et al., (2020) identified 62 SNPs

that were associated with cold tolerance and were distributed over six of the seven pea linkage groups. The same study also revealed the presence of CBF as a potential genetic determinant of the frost tolerance locus in linkage group VI.

### Genetic Tools in Peas

Mendel's experiments with peas laid the foundation for understanding the principles of inheritance, which are now known as Mendel's Laws. Following Mendel's work, the field of genetics continued to evolve, with researchers building upon his discoveries. In the early 20th century, William Bateson, a British biologist, began using pea plants in his research program to further Mendelian genetics. The John Innes Centre, a leading plant science research institute in the UK, has a long history of pea research, dating back to its establishment in 1910.

In recent years, advancements in molecular biology and genomics have allowed researchers to identify and characterize the genes responsible for the traits Mendel studied in pea plants. The advancement of genetic tools has played a crucial role in advancing pea breeding programs and understanding the molecular basis of important traits. The development of these tools has been driven by significant technological breakthroughs. The first fully annotated genome assembly for pea was published in 2019, providing the knowledge of genome evolution and facilitating the identification of favorable alleles underlying phenotypic variations (Kreplak et al., 2019). This complete and accurate reference genome has provided a big resource for genomics and breeding research. It has enabled researchers to better understand the genetics of important traits in peas, such as disease resistance, stress tolerance, and yield potential, ultimately contributing to the development of improved pea varieties.

A more recent study in 2022 reported an improved pea reference genome and pan-genome, highlighting genomic features and evolutionary characteristics (Yang et al., 2022). This high-quality genome and pan-genome serve as an important resource for pea genetics and breeding research, allowing scientists to delve deeper into the molecular mechanisms underlying pea domestication and adaptation. This improved understanding of the pea genome has the potential to inform breeding strategies and accelerate the release of new pea varieties with desirable traits.

The availability of comprehensive genetic maps and reliable DNA markers has greatly facilitated the development of genomic tools for pea breeding (Parihar et al., 2022). Genetic maps play a crucial role in genetics and genomics research by providing a way to locate and identify the relative positions of genes or genetic markers on chromosomes. This enables researchers to identify regions of the genome associated with specific traits. DNA markers (SNPs) and simple sequence repeats (SSRs), serve as signposts for these regions, allowing breeders to track the inheritance of desirable traits in breeding populations. Whole-genome sequencing information has been instrumental in uncovering the molecular bases of vital traits in pea and breeding efforts (Tayeh et al., 2015). Single nucleotide polymorphisms (SNPs) are valuable markers for genetic studies and breeding.

A resource of pea mutants provides a valuable tool for both basic science and crop improvement. The John Innes collection in the UK and the Institute of Plant Genetics Resources Collection in Bulgaria hold a combined 697 pea mutant stock accessions (Smýkal et al., 2015). These mutant resources include a collection of pea lines with induced mutations in specific

genes, enabling researchers to study the function of individual genes and their roles in various biological processes.

Marker-assisted selection (MAS) utilizes genetic markers to select individuals likely to carry desirable traits (Lande & Thompson, 1990). This technique significantly enhances the efficiency and precision of breeding programs by allowing breeders to identify and select desired traits earlier in the process. MAS can also reduce the amount of time and labor required for breeding by eliminating the need to phenotype individuals for the desired traits.

Over 98,000 pea accessions, including advanced breeding lines, landraces, mutant stocks, wild species, and cultivars, are available and conserved in diverse gene banks worldwide. These genetic resources represent a wealth of genetic diversity, providing a valuable resource for pea breeding programs. By tapping into this diversity, breeders can identify novel alleles and gene combinations that contribute to desirable traits, such as high yield, disease resistance, and stress tolerance. The conservation and utilization of these genetic resources are essential for the continued improvement of pea varieties and the sustainable production of this important crop. Access to these genetic resources is facilitated through national and international seed banks, research institutions, and collaborative networks, ensuring that pea breeding and research can continue to enhance the crop's productivity, sustainability, and nutritional value.

### Summary

Winter peas can emerge as a pivotal crop in sustainable agriculture, offering exceptional benefits and resilient crop options in the face of climate change. It would be beneficial in crop rotations and break the disease cycle. The cultivation, driven by strategic management practices, improves soil health, enhances nitrogen fixation, and contributes significantly to crop rotation

systems. Emphasizing the need for ongoing research into genetic improvement and tailored agronomic techniques, winter peas represent a promising avenue for enhancing agricultural productivity, sustainability, and environmental stewardship. Integrating winter peas into cropping systems can bring about various agronomic and economic benefits, making them a valuable component of sustainable agricultural practices.

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MULTI-ENVIRONMENT EVALUATION OF WINTER PEA  
GERMPLASM LINES FOR WINTER SURVIVAL AND YIELD  
STABILITY

Contribution of Authors and Co-Authors

Manuscript in Chapter 2

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## Introduction

Field pea (*Pisum sativum* L.) holds a significant place in agriculture due to its versatile applications, both as a crop and as a contributor to sustainable farming practices. Winter pea is an important crop providing additional benefits of replacing winter fallow and eliminating the need for spring planting in adverse conditions in Montana and the Northern Great Plains (NGP). There has been success in growing winter peas as a cover crop in the northwestern regions of the US, but most winter pea lines are not adapted to the harsh environment of Montana. This requires breeding winter pea varieties suitable to grow in cold regions through rigorous selection and testing.

Winter fallow remains a common practice in Montana due to low moisture and hard winter. This has caused many concerns in the soil including erosion, soil structure degradation, and compaction. A significant economic drawback of traditional winter fallow practices for farmers is the opportunity cost involved. Winter fallow has made the system problematic in the following spring due to the nutrient loss from the fallow soil and farmers have to bear the financial pressure (Adil et al., 2022). Incorporating winter pea into cropping systems brings numerous advantages. One of the most significant benefits is the enhancement of soil health; winter peas can improve soil structure, increase organic matter, and boost nitrogen levels through nitrogen fixation, thereby reducing the need for synthetic fertilizers (Kebede, 2021). This practice also helps in controlling erosion by protecting the soil surface from wind and water damage, thus conserving topsoil and its nutrients (Adetunji et al., 2020).

However, the selection process for winter peas is harder and more time-consuming than for spring peas. Selection of winter pea involves screening the lines in weather cold enough to

differentiate hardiness among the lines without killing them. Moreover, yearly temperature and snowfall fluctuations make it challenging to collect multiple years of useful data. Efficient selection of winter hardy lines is a challenge if it is limited to a single location. The selection process can be effective if the lines are tested across multiple locations over multiple years. Multi-environment trials (MET) help to establish the variance in traits due to genotype, environment, and genotype-by-environment (G x E) interaction. These trials also help to differentiate better performing genotypes from the ones already in the market. The evaluation of winter pea genotypes for winter survival and yield stability in the multi-environment trials would make improved winter pea varieties available to the producers. This study evaluated germplasm lines for three years at four locations in Montana to identify the genotypes suitable for commercial winter pea production.

## Materials and Methods

### Germplasm Lines

Germplasm lines were obtained from the Plant Germplasm Introduction and Test Research Unit, Pullman, Washington. Forty-eight, thirty-eight, and thirty-six germplasm lines were planted in 2021, 2022, and 2023, respectively, at Bozeman, Havre, Huntley, and Moccasin, Montana. Fenn, Glacier, Granger, Lynx, Melrose, Romack, Specter and Windham were the check varieties used in the experiment. The lines were common in the experiments across years. Genotypes used in the study were dropped in the second year according to the assessment of their performance under different environmental conditions.

### Field and Experimental Design

Trials of winter pea germplasm lines were carried out over three years, from 2021 to 2023, in Bozeman, Havre, Huntley and Moccasin, Montana, using a Randomized Complete Block Design (RCBD) with consistent checks. The number of replicates varied by location, ranging from two to four. In Bozeman, plots were precisely sown using a Wintersteiger XXL dynamic disc plus precision spaced plot seeder, (Wintersteiger Incorporation, Salt Lake City, Utah). This specific seeder features six double disk openers, with row spacing 22.5 cm, and the plots spanned a length of 3 meters. Equipment variations were noted in other locations, depending on what was available. Havre and Moccasin had the same plot dimension as Bozeman. The length of the plots was 6 meters in Huntley with same row spacing as other locations. Seeding rate was 120 plants per square meter and was consistent across the locations. CruiserMaxx was used to protect against insects and fungi at 2.5 ml per kg of seed. The trials were conducted without the application of any fertilizer. Seeds were inoculated before planting using *Rhizobium leguminosarum* biovar *viceae* ( $2 \times 10^8$  CFU/g) inoculant. Weeds were hand weeded as needed and standard agronomic practices were used at each location.

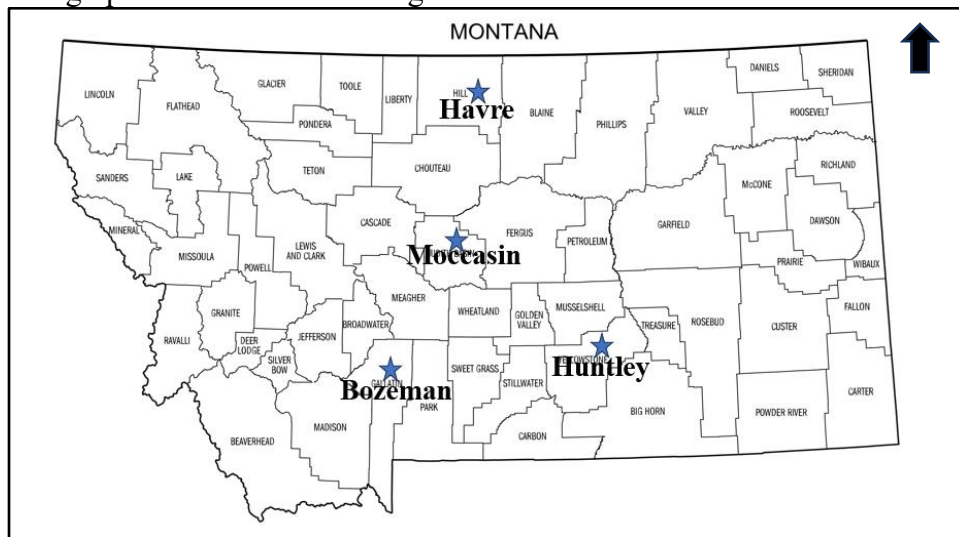
### Data Collection

Data collected included fall stand count, spring stand count, date of 10% bloom, date of 90% bloom, lodging score, canopy height at maturity (cm), maturity date, seed yield (kg/ha), 1000-seed weight (g), moisture (%), and protein content (%). Vine length (cm), number of flowering nodes, number of reproductive nodes, height of lowest pod (cm), height of lowest pod at maturity (cm), number of pods per peduncle were measured by selecting two random plants from two interior rows of each plot. Protein content was measured by near-infrared (NIR)

spectroscopy using Infratec™ 1241 Grain Analyser (Foss North America, Minneapolis, Minnesota). This device was calibrated with over 50,000 cross checked samples collected from over 20 years of harvests. A meter stick was used to record the number of plants in a meter of a plot. Fall stand count was collected after emergence of seedlings (September-October) and spring stand count after the snow was cleared off the surface in spring (April-May). Winter survival was recorded as the ratio of the number of plants survived in the spring to the number of plants emerged in the fall and expressed in percentage. Seed yield and 1000-seed weight were measured in kg/ha and g, respectively.

### Trial Environments

Figure 1.1. Geographical location of testing environments.



The trial sites were located across Montana (Figure 1.1). Bozeman and Huntley are located on the southern part of the state, whereas Havre is located to the north and Moccasin is centrally located. Every location has its own distinctive climatic characteristics posing a unique opportunity and challenge in the multi-environment trial (Table 1.1).

Table 1.1. Characteristics of the trial environments.

Year	Location	Latitude & Longitude	Altitude	Avg	Avg Max	Avg Min	Soil Type
			m	-----°C-----			
2021	Bozeman	45°40'27.5"N 111°08'48.4"W	1455	7.4	14.7	0.7	Silt loam
	Havre	48°29'33.3"N 109°48'19.1"W	820	7.1	14.4	-0.5	Telstad loam
	Huntley	45°55'15.3"N 108°14'22.1"W	920	8.1	16.3	-0.5	Clay loam
	Moccasin	47°03'21.6"N 109°57'17.4"W	1280	7.5	12.9	-0.4	Clay loam
2022	Bozeman	45°40'28.1"N 111°09'21.9"W	1448	7.6	14.7	0.6	Silt loam
	Havre	48°29'32.7"N 109°48'05.5"W	825	6.6	14	-1.2	Telstad loam
	Huntley	45°55'30.2"N 108°14'13.4"W	920	7.9	16.3	-1.4	Clay loam
	Moccasin	47°03'36.8"N 109°57'22.8"W	1280	7.8	12.3	-0.1	Clay loam
2023	Bozeman	45°40'28.8"N 111°08'59.6"W	1458	5.7	12.3	-0.9	Silt loam
	Havre	48°29'30.4"N 109°47'56.5"W	820	5.4	12.2	-1.3	Telstad loam
	Huntley	45°55'20.6"N 108°14'14.0"W	920	7	14.9	-0.8	Clay loam
	Moccasin	47°03'31.3"N 109°57'11.4"W	1280	5.3	11.4	-1	Clay loam

### Weather Data

Weather data was downloaded for the latitude and longitude of each environment from North American Land Data Assimilation System Phase 2 and Western Regional Climate Center (WRCC). The variables downloaded were precipitation, maximum air temperature, minimum air temperature, average air temperature, relative humidity, and average wind speed. Growing degree days (GDDs) were calculated as the average daily temperature above 32°F, which is the most used base temperature for winter crops (Miller et al., 2001).

Figure 1.2. Average air temperature (°C) for each research locations.

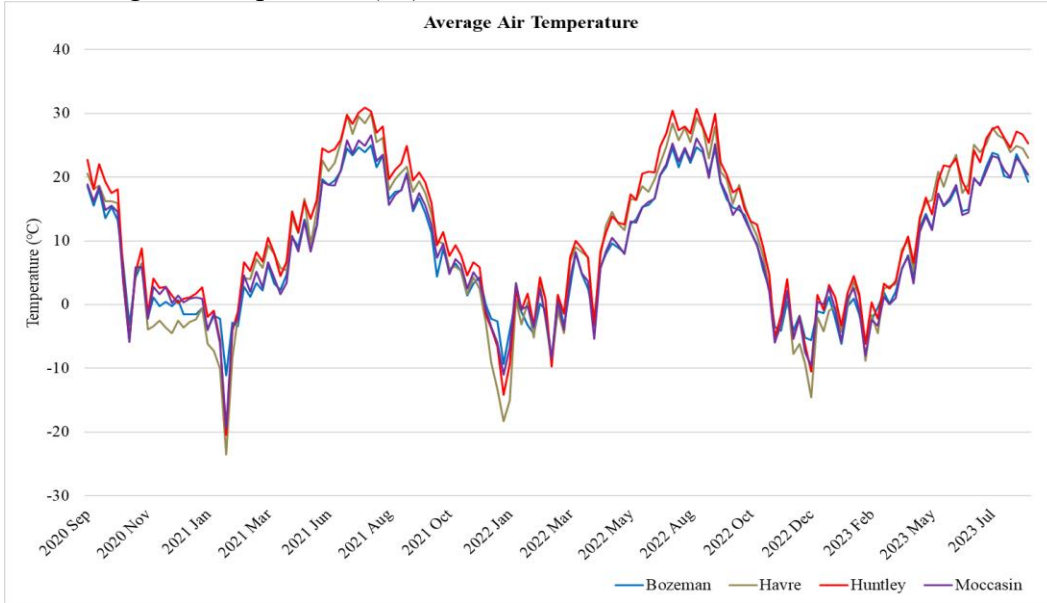


Figure 1.3. Average maximum air temperature (°C) for each research locations.

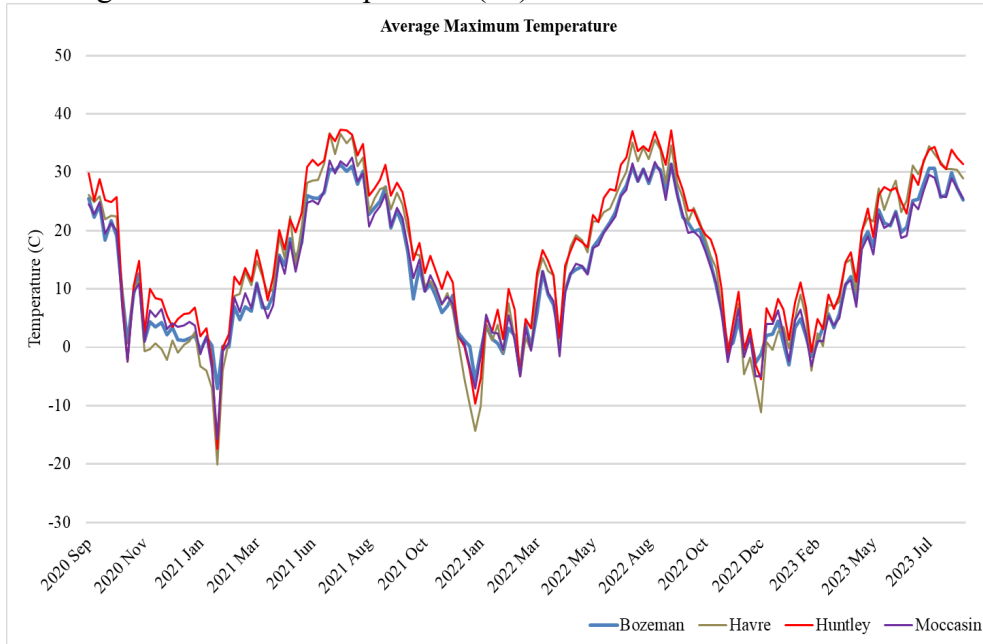


Figure 1.4. Average minimum air temperature (°C) for each research locations.

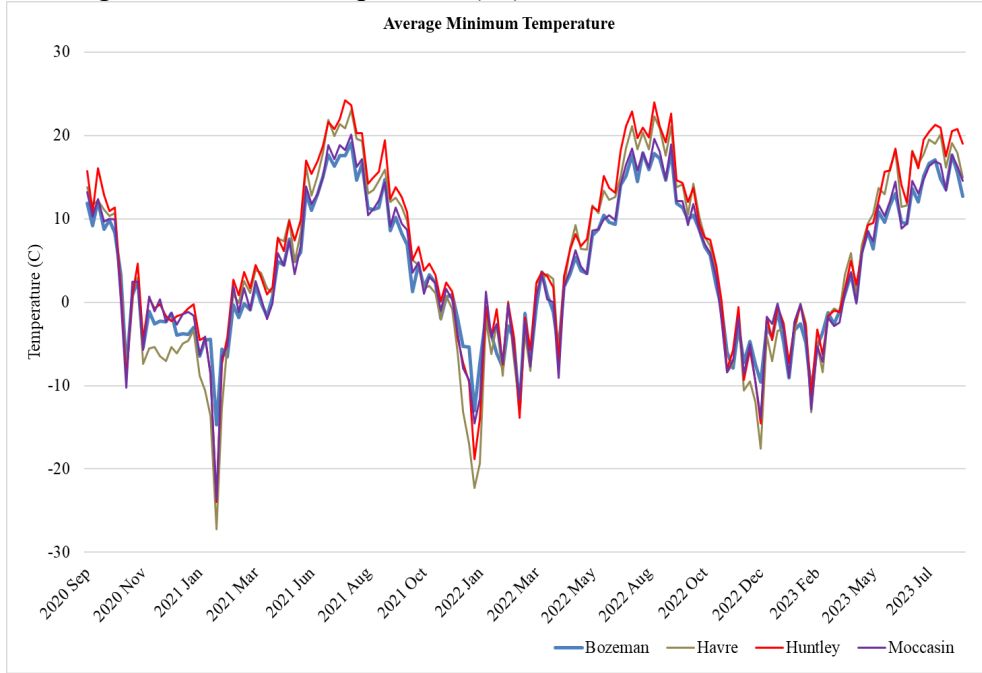
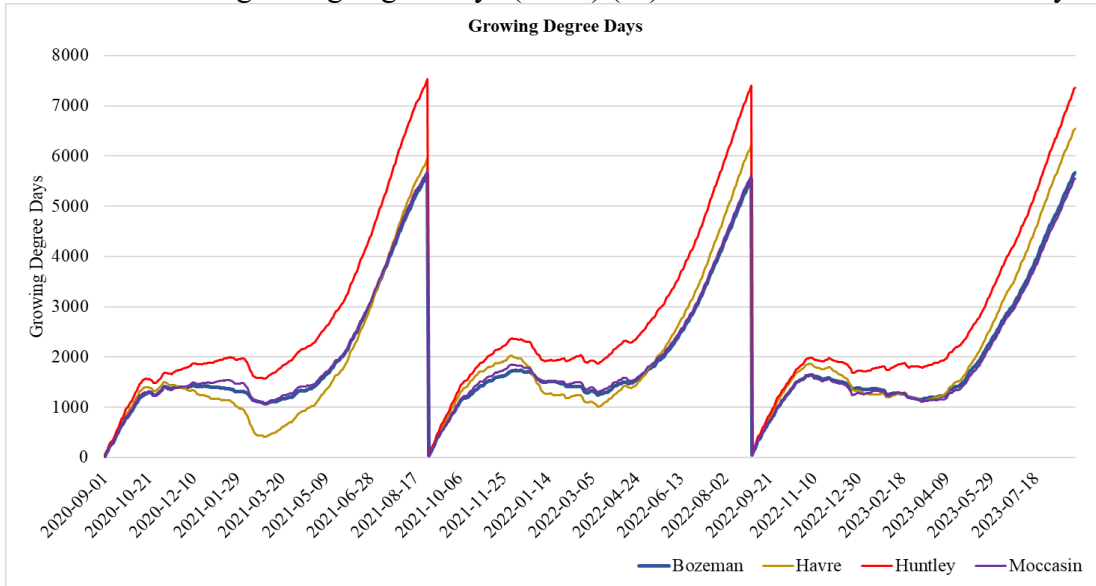


Figure 1.5. Cumulative growing degree days (GDD) (°F) of research locations for each year.



### Statistical Analysis

The dataset was filtered for missing values before analysis. To accommodate the intricacies of our MET design, we employed a generalized linear mixed model (GLMM), which is particularly suited for analyzing data that involves multiple levels of random variation. The effect of genotypes, locations, years, and interactions were considered fixed, whereas block nested over location was considered a random effect. This model allowed us to partition the variability due to specific genotypic performance, environmental effects, and their interactions, while simultaneously managing the random effects introduced by the blocks. A combined analysis of variance (ANOVA) was used to test the significance of main effects and quantify the interactions among and within the source of variation in seed yield, 1000 seed weight, protein content, and winter survival using the *lme4* package and mean separation used Tukey's HSD. The effects were considered statistically significant at  $P < 0.01$ . All analyses were done in R studio using multiple packages (RStudio 2023.06.0).

The core of our statistical analysis was the GLMM, constructed to dissect the influence of genotypes, years, locations, and their interactions on the traits yield, 1000 seed weight, protein content and winter survival. The model for each trait was structured as follows:

$$\text{trait}_{ijkl} = \mu + \text{name}_i + \text{year}_j + \text{location}_k + (\text{name} \times \text{year})_{ij} + (\text{name} \times \text{location})_{ik} + (\text{year} \times \text{location})_{jk} + (\text{name} \times \text{year} \times \text{location})_{ijk} + b_l(\text{bloc}) + \epsilon_{ijkl}$$

In this equation,  $\text{trait}_{ijkl}$  represents the observed value for the  $i^{\text{th}}$  genotype in the  $j^{\text{th}}$  year at the  $k^{\text{th}}$  location within the  $l^{\text{th}}$  block. The symbol  $\mu$  is the overall mean value, and  $\epsilon_{ijkl}$  is the residual error term, assumed to be normally distributed. By incorporating  $b_l(\text{bloc})$ , the model accounts for block-to-block variability, allowing for random fluctuations in the trait that are not explained by the fixed effects.

Pearson correlation was used to establish the relationship among the traits vine length, canopy, pod height, pod height at maturity, number of flowering nodes, number of reproductive nodes, number of pods per peduncle, fall stand count, spring stand count, winter survival, protein content, one thousand seed weight, and seed yield.

To understand which genotype performed best, genotypes were ranked based on the yield for each location and each year. They were also ranked overall for each location and year by adding up the ranks to determine the best-ranked genotypes.

However, the variance component alone was not sufficient to clarify the details of genotype by environment interaction. Additional statistical techniques involving multivariate analysis were required to understand the Genotype by Environment Interaction (GEI) more accurately. For that reason, we used the GGE biplot method to visualize the GE patterns and identify the ideal genotypes and environments for each trait. The methodology was described by (Yan et al., 2000), which is explained by the equation:

$$Y_{ij} - \mu - B_j = \lambda_1 \varepsilon_{1i} \eta_{1j} + \lambda_2 \varepsilon_{2i} \eta_{2j} + \epsilon_{ij}$$

where  $Y_{ij}$  is the genotypic value of genotypes  $i$  in  $j$  environment,  $\mu$  is the overall mean,  $B_j$  is the effect of the  $j$  environment,  $\lambda_1$  and  $\lambda_2$  are Eigenvalues associates with PC1 and PC2,  $\varepsilon_{1i}$  and  $\varepsilon_{2i}$  are the scores for the PC1 and PC2 axes for the genotype  $i$ ,  $\eta_{1j}$  and  $\eta_{2j}$  are the scores for PC1 and PC2 axes for genotype  $j$ .  $\epsilon_{ij}$  is the error associated with the model which is not explained by the first two PCs. All these analyses were performed using the metan package in R (Olivoto & Lúcio, 2020).

For genotype evaluation, genotype-focused singular value partitioning (SPV=1) was used using the ‘mean versus stability’ option whereas for location evaluation, environment-focused singular value partitioning (SVP=2) was used (Yan, 2001). The GGE biplot shows graphical images to illustrate genotype and environment interaction and genotype ranking based on mean and stability. The graphs generated are based on the ranking of the environments (discriminative versus representative), evaluation of genotypes (mean versus stability) and multi-environment evaluation (which-won-where pattern). The which-won-where pattern also identified which genotype was the winner in a given set of environments.

The broad ecological or environmental condition that influences the performance and adaptation of crop varieties are called mega-environments. Within a single mega-environment, the objective of data analysis was genotype evaluation to identify high-performing and highly stable genotypes. Test environment evaluation was to find test environments that were both discriminating (informative) and representative of other environments. Whenever there was potential G x E interaction, it was explored.

Biplots were based on the seed yield (kg/ha), thousand seed weight (g), protein content (%), and winter survival (%) data of 2021, 2022, and 2023 winter pea germplasm lines which were tested at four locations (Bozeman, Havre, Huntley, and Moccasin) in MT.

## Results

### Univariate Analysis of Germplasm Lines

The highest yield in 2021 was observed at Huntley followed by Bozeman, Moccasin, and Havre. Huntley’s yield was significantly higher from other locations (Figure 1.6). Yield in Huntley was lower in 2022, due to damage incurred by hail during the harvest period. Seed yield

at Havre was statistically similar to Moccasin but significantly higher than Huntley and lower than Bozeman. Seed yield in 2023 was high at all locations except Huntley. The highest yield was at Havre, followed by Bozeman, and Moccasin. Seed yield at Havre was significantly different from all the other locations, and Huntley had the lowest yield.

The highest yield was observed at Havre in 2023 which was significantly different from Bozeman and Moccasin. However, yields at Moccasin in 2022 and 2023 were not significantly different from each other. 2021 Huntley, 2022 Havre, and 2022 Moccasin had similar yields. Also, 2022 Bozeman and 2023 Moccasin had similar yield (Figure 1.7). 1000-seed weight was significantly higher in 2021 Bozeman, and lowest in 2022 Huntley (Figure 1.8). Similarly, higher protein content was found in all years in Havre and in 2022 Huntley (Figure 1.9).

Winter survival was one of the traits used to determine the hardiness of the lines in different environmental conditions. Figure 1.10 displays a bar graph that depicts the distribution of survival rates (%) in four distinct locations over a three-year period from 2021 to 2023. At some instances due to dormancy, survival was greater than 100% because the number of plants in the spring were more than what emerged in the fall. Moccasin 2023 had the highest winter survival (122%) whereas Bozeman 2023 had the lowest winter survival (55%) (Table 1.4). Few lines like Melrose, PI 639979 and PI 517923 had higher winter survival, whereas Junior, G10946 and L 805/5g had lower winter survival percentages.

Figure 1.6. Mean seed yield of germplasm lines grown at four locations in 2021, 2022, and 2023. Letters above the bars represent statistical groupings.

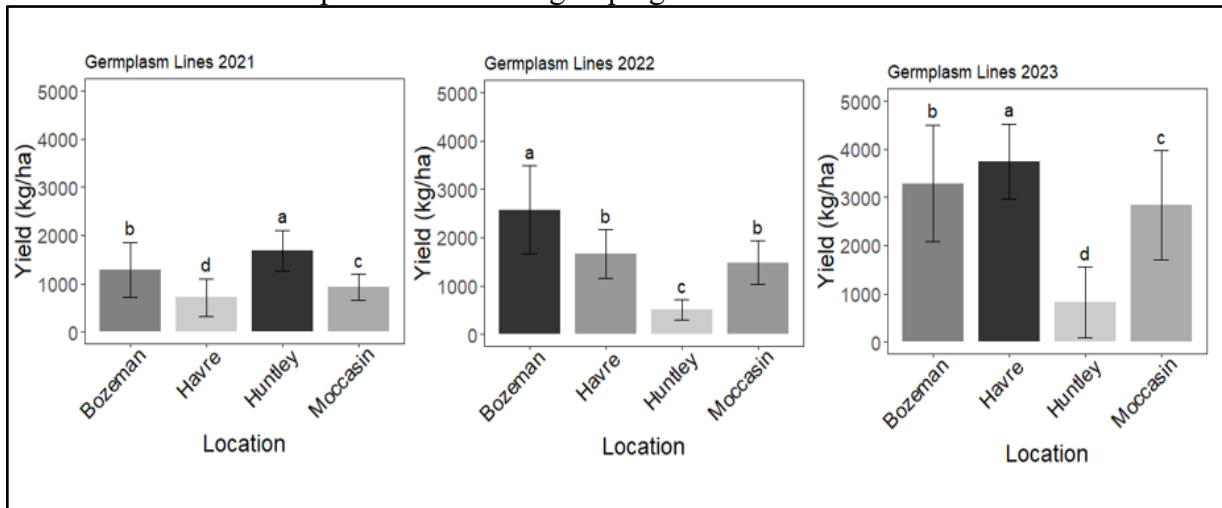


Figure 1.7. Mean seed yield of germplasm lines across environments. Letters above the bars represent statistical groupings.

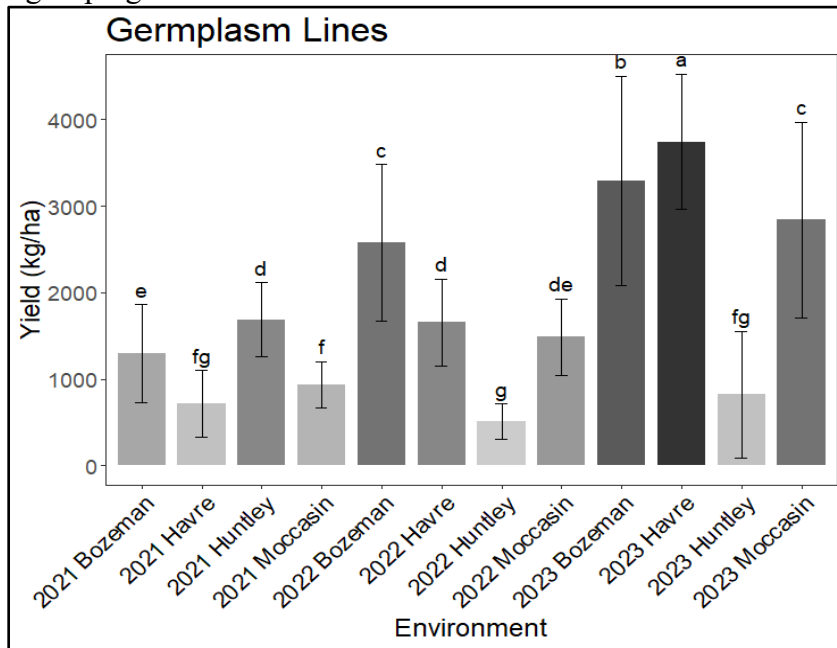


Figure 1.8. Mean one thousand seed weight of germplasm lines across environments. Letters above the bars represent statistical groupings.

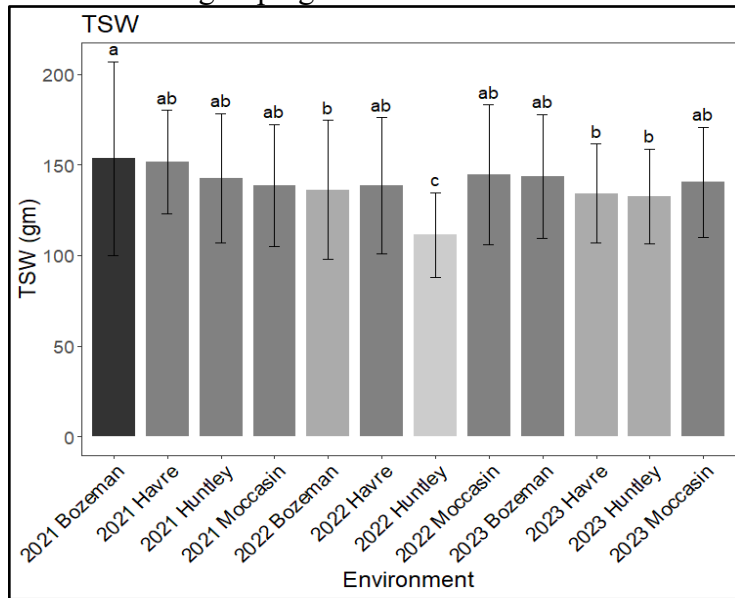


Figure 1.9. Mean protein content of germplasm lines across environments. Letters above the bars represent statistical groupings.

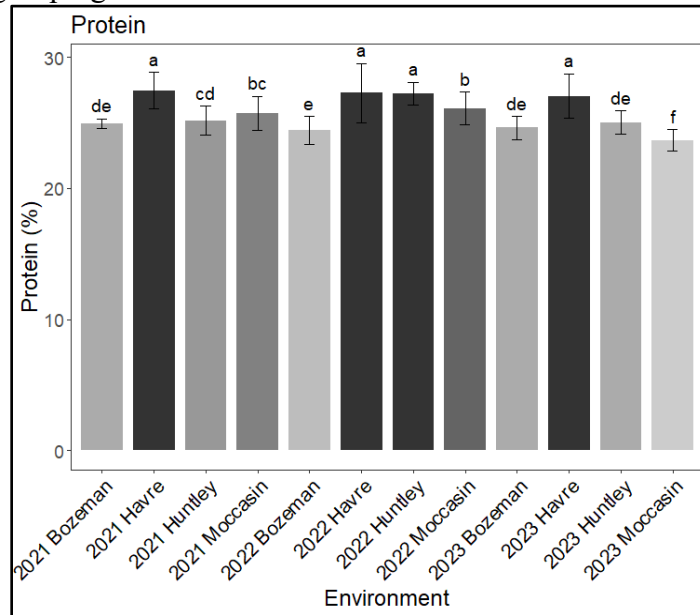


Figure 1.10. Mean winter survival of germplasm lines across environments. Letters above the bars represent statistical groupings.

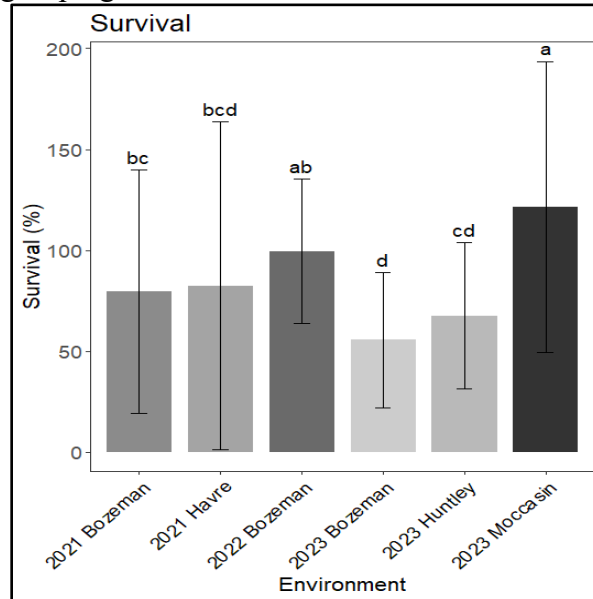


Table 1.2. Analysis of Variance (ANOVA) table showing the effects of genotype, location, and year on seed yield, 1000-seed weight, protein content, and winter survival of germplasm lines for three years across four locations.

Source	df	Yield	df	1000SW	df	Protein	df	Survival
		<i>P&gt;F</i>		<i>P&gt;F</i>		<i>P&gt;F</i>		<i>P&gt;F</i>
Genotype	37	<0.01**	37	<0.01**	37	<0.01**	37	<0.001**
Location	3	<0.01**	3	<0.01**	3	0.6	3	1
Year	2	<0.01**	2	<0.01**	2	<0.01**	2	<0.001**
Genotype:Location	109	<0.01**	109	<0.01**	110	<0.01**	95	0.01*
Genotype:Year	72	<0.01**	72	<0.01**	72	<0.01**	72	0.9
Location:Year	6	<0.01**	6	<0.01**	6	<0.01**		
Genotype:Location:Year	183	<0.01**	181	<0.01**	181	<0.01**		

\*Significant at the 0.05 probability level.

\*\*Significant at the 0.01 probability level.

Table 1.3. Percentage variation explained by different source of variation in various traits.

Source	%Variation Explained			
	Yield	1000 SW	Protein	Survival
Genotype	14.7	85.5	21.3	29.6
Location	0.4	0.3	0.3	2.7
Year	29.4	1.5	10.2	11.0
Genotype:Location	9.9	2.1	22.1	41.9
Genotype:Year	6.4	3.3	10.0	14.8
Location:Year	28.4	3.7	18.1	
Genotype:Location:Year	10.9	3.7	18.0	

Analysis of variance was performed to describe the main effect and quantify the interactions among and within the source of variations. The pooled analysis of variance is presented in Table 1.2. Genotype, location, year, and interactions showed significant differences ( $p < 0.01$ ) for seed yield and 1000-seed weight. Also, for protein, everything was significantly different except location.

The largest source of variation in yield (29.4%) and 1000-seed weight (85.5%) was attributed to the year and genotypes respectively. The interaction between the genotypes and the location (Name:Location) was also notably significant in explaining the variation in protein content (22.1%) and survival (41.9%) (Table 1.3).



The lines that connect the test environments (Bozeman, Havre, Huntley, and Moccasin) to the biplot origin are called environment vectors. An acute angle between the two environment vectors shows a positive correlation between them, whereas an obtuse angle is an indication of a negative correlation between them. A larger obtuse angle shows the strong crossover  $G \times E$  between those two environments. A right angle between two environments shows that they are not correlated to each other (Yan & Tinker, 2006).

In Figure 1.11 (a), all the environment vectors except Huntley-Havre had an acute angle between them for yield, showing a positive correlation between each of the two environments. The environment vectors of Huntley-Havre had about 90 degrees between them. This showed these two environments are unrelated to each other for yield. For 1000-seed weight, Havre did not have positive correlation with any environment, whereas all other environments had positive correlation between them (Figure 1.11 (b)). There was positive correlation between each pair of environments for protein (Figure 1.11 (c)). For survival, Havre and Moccasin were negatively correlated (Figure 1.11 (d)).

Figure 1.12. Discrimination and representative view of the GGE biplot showing the discriminating ability and representativeness of the test environments for yield (a), 1000-seed weight(b), protein (c), and survival (d).

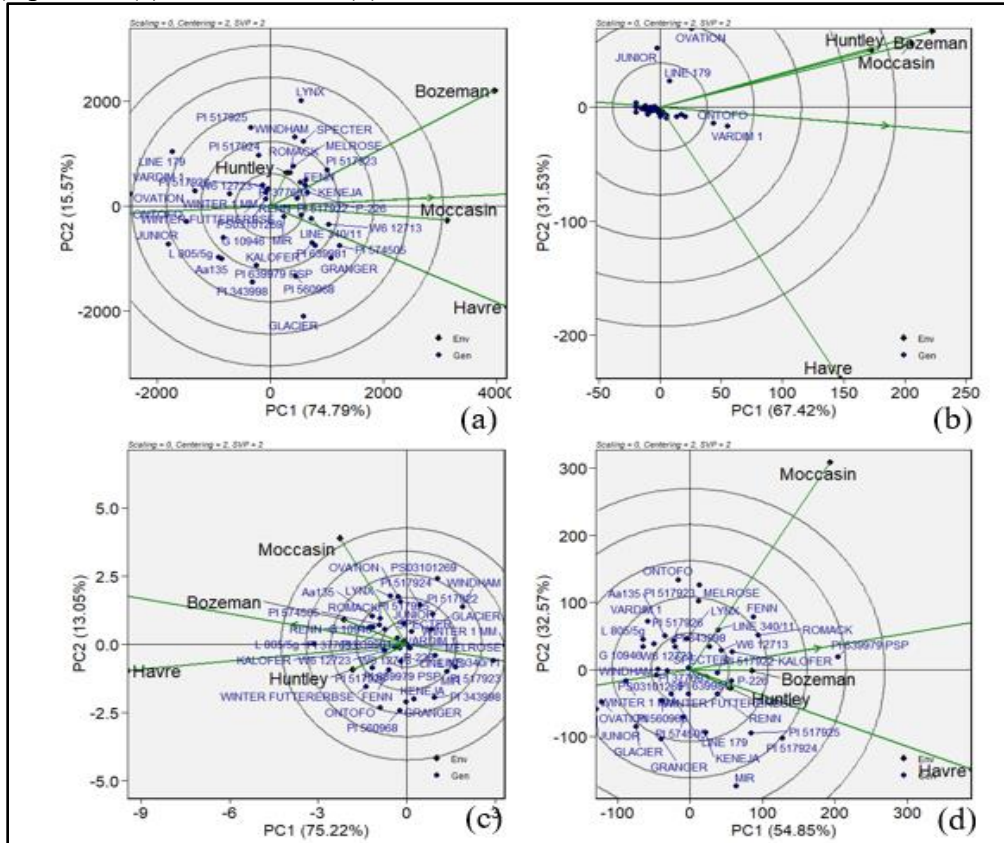


Figure 1.12 presents an Average Environment Axis (AEA), a line that passes through the average environment and the biplot origin. The average environment (represented by the point at the end of the arrow) has the average coordinates of all test environments. A test environment that has a smaller angle with the AEA is more representative.

Moccasin was the most representative test environment than other environments for yield and 1000-seed weight (Figure 1.12 (a) and 1.12 (b)). Similarly, Bozeman was the most representative environment for protein (Figure 1.12 (c)) and survival (Figure 1.12 (d)). Havre with the long vectors, was the most discriminating for 1000-seed weight and protein.

Havre was both discriminating and representative for protein, thus a good environment for selecting generally adapted genotypes. Discriminating but non-representative test environments Bozeman and Havre for yield and Havre for 1000 seed weight are useful for selecting specifically adapted genotypes if the target environment can be divided into mega-environments. These can also be used for culling unstable genotypes if the target environment is a single mega-environment. Non-discriminating test environment like Huntley with a very short vector is less useful because it provides very little discriminating information about the genotypes.

## Ranking Environments

Figure 1.13. Discrimination and representative view of the GGE biplot ranking test environments relative to an ideal test environment (center of the concentric circles) for yield (a), 1000-seed weight(b), protein (c), and survival (d).

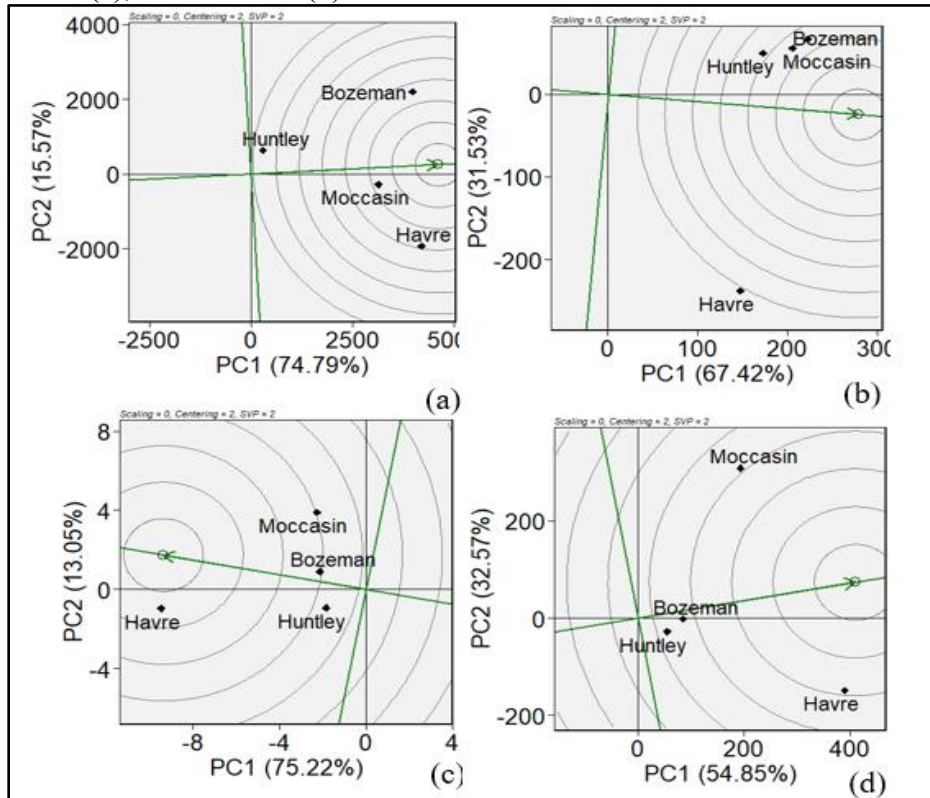


Figure 1.13 shows the ideal test environment which is the center of the concentric circles. It is a point on the AEA (most representative) which is farthest with a distance to the biplot origin equal to the longest vectors of all environments (most discriminating). Moccasin and Havre were closest to this point for yield (a) and protein (c) respectively, so are considered good environments. Huntley for yield and Havre for 1000 seed weight (b) were the poorest for selecting cultivars adapted to the region of Montana. Similarly, Havre was better than other locations for winter survival (d).

### Ranking Genotypes Relative to the Ideal Genotype

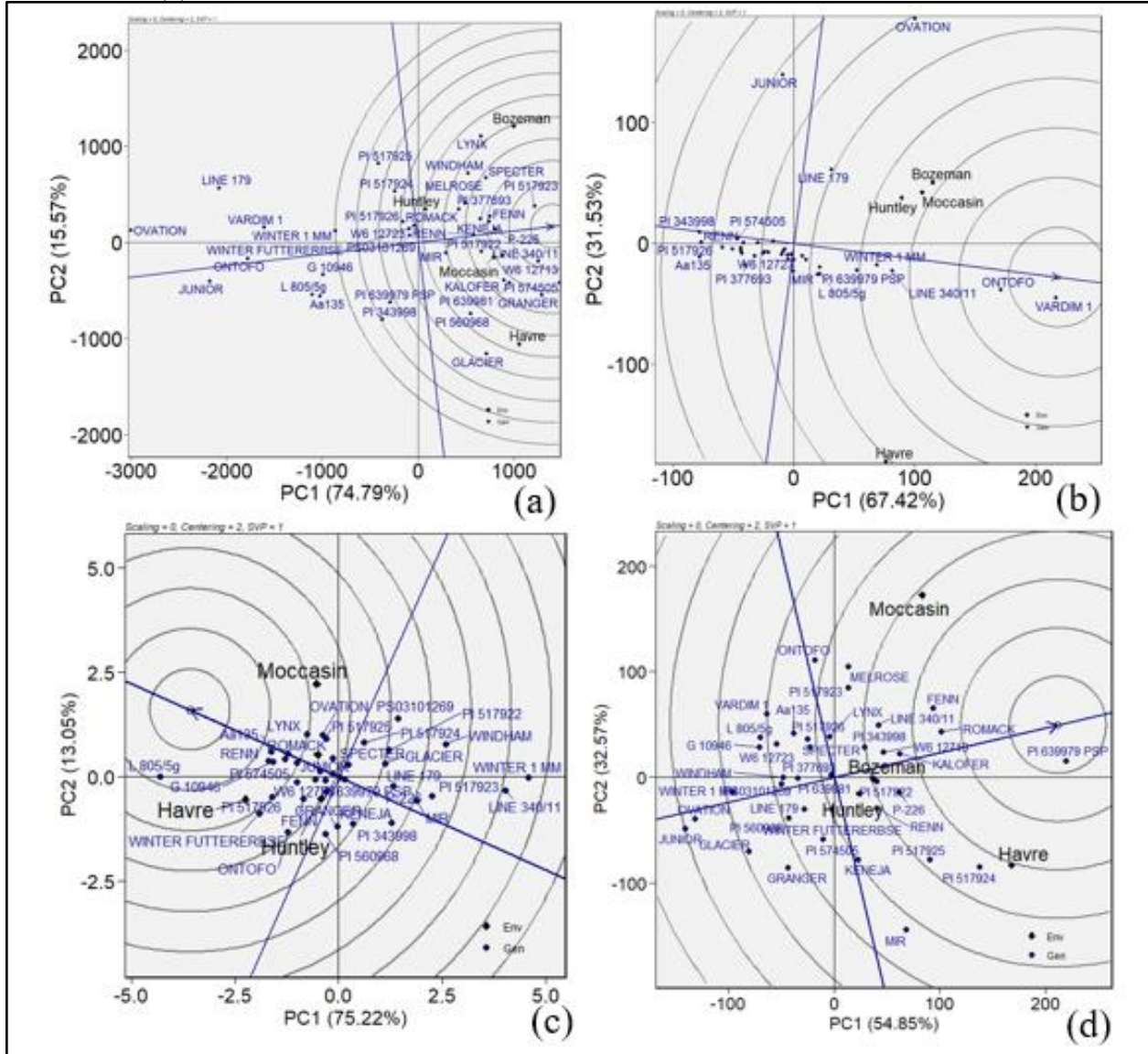
An ideal genotype should have both high mean performance and high stability across environments. Figure 1.14 also shows the ideal genotype which is the center of the concentric circles. The genotypes located closer to that point are considered good for a specific trait and are more desirable than other genotypes lying farther from the center.

PI 517923 and W6 12713 lie closer to the center of the circle (Figure 1.14 (a)), so they are considered the high yielding whereas OVATION lies far from the center, which is a considerably low-yielding genotype. Also, PI 517923, which lies closer to the center, is more desirable than PI 574505 even though the latter had a higher average yield.

L 805/5g is located closest to the center of the concentric circles (Figure 1.14 (c)), so it has good protein content. Lines like WINTER 1 MM and LINE 340/11 had the lowest protein content. VARDIM 1 and ONTOFO had the biggest seed size and were closer to the center of the concentric circles (Figure 1.14 (b)). Similarly, PI 639979 PSP had the better survival than others (Figure 1.14 (d)).

The distance between two genotypes approximates the Euclidean distance between them, which is the measure of overall dissimilarity between them. For example, from Figure 1.14 (a), PI 517923 and FC 40623 are very different whereas GRANGER and PI 574505 are quite similar in yield. The dissimilarity can be due to differences in mean yield (G) and/or in interaction with the environments (G x E).

Figure 1.14. The average-environment coordination (AEC) view ranking genotypes relative to an ideal genotype (center of the concentric circles) for yield (a), 1000-seed weight(b), protein (c), and survival (d).

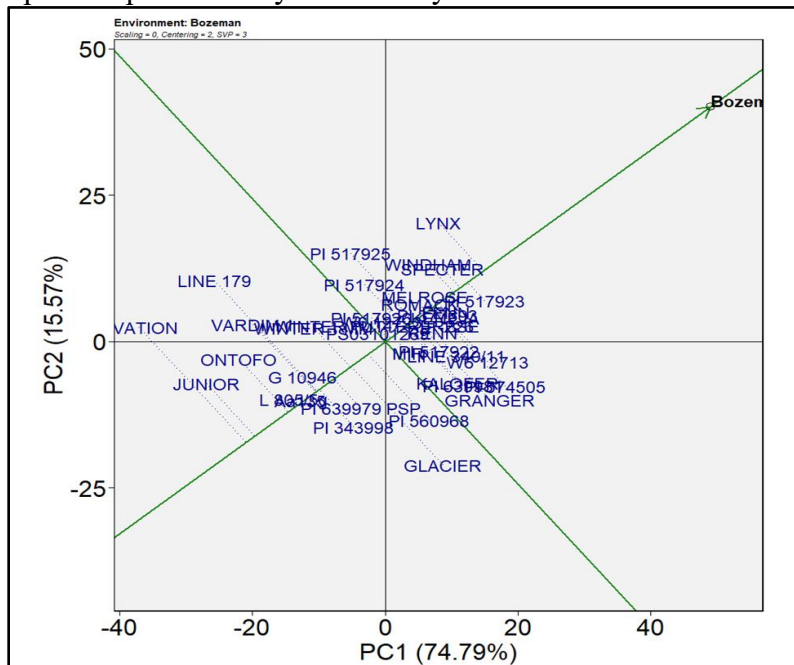


### Examine Environment Based on the Performance of Germplasm Lines

In order to examine an environment based on the performance of the genotypes, a line is drawn from the biplot origin to the environment. Genotypes are ranked along the axis for that environment.

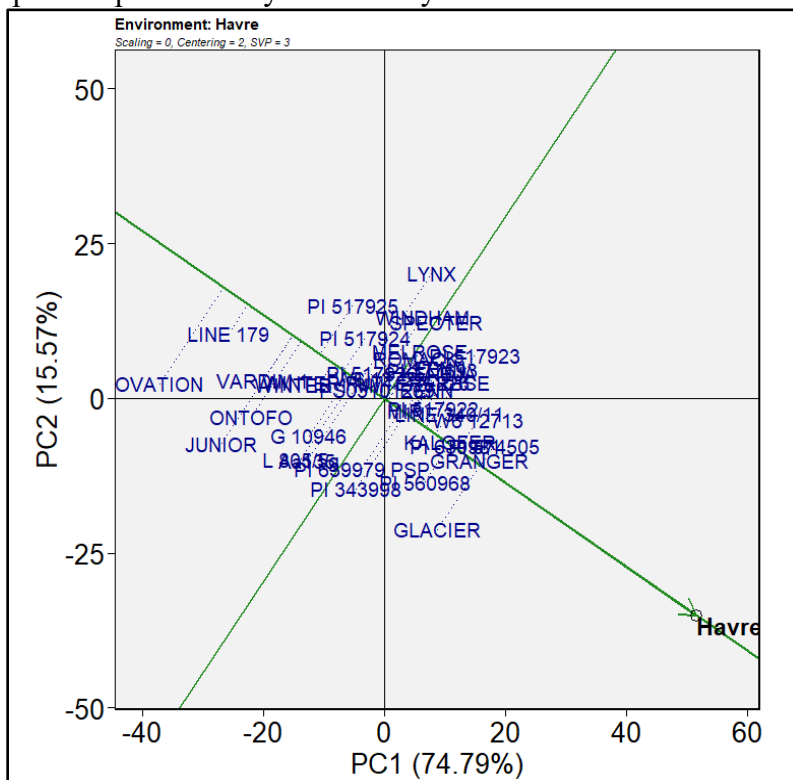
### Bozeman

Figure 1.15. Principle component analysis of seed yield at Bozeman.



## Havre

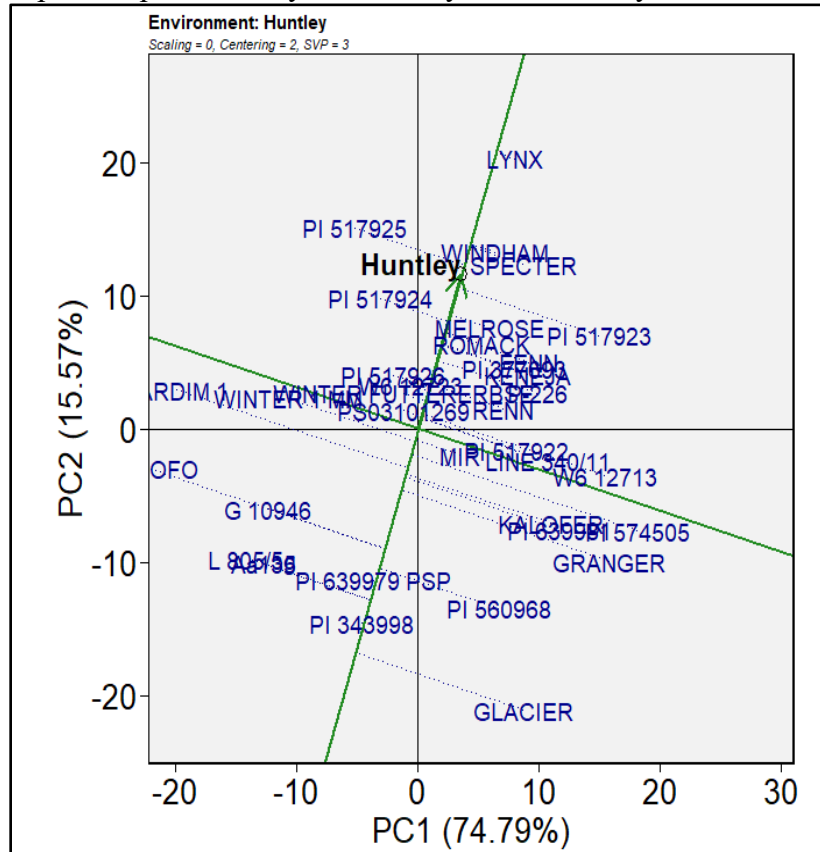
Figure 1.16 Principal component analysis of seed yield at Havre.



In Havre, Glacier, PI 574505, Granger, and W6 12713 had higher-than-average yield. PI 517924, L 805/5g, and W6 12723 had about average yield whereas Winter 1MM, PI 517925, and Ontofo had lower-than-average yield. Glacier and Line 179 were the highest and lowest yielding lines, respectively, in Havre (Figure 1.16).

Huntley

Figure 1.17. Principle component analysis of seed yield at Huntley.



In Huntley, Lynx, Specter, Windham, and PI 517925 had higher-than-average yield. PI 517922, Winter 1mm, and W6 12713 had about average yield whereas Glacier, PI 343998, and PI560968 had lower-than-average yield. Lynx and Glacier are the highest and lowest yielding lines, respectively, in Huntley (Figure 1.17).



across all environments followed by Granger, W6 12713, PI 517923, etc. PI 639979 PSP and PI 343998 had mean yield similar to the grand mean and MELROSE had the lowest mean yield. A vertical green line is the AEC ordinate. It shows greater variability (poor stability) in either direction. The dotted line drawn from the AEC to the genotype reflects the stability of the genotype. The longer the line, the less stable the genotype is, and vice versa. From Figure 1.19, Lynx and Glacier were seen as highly unstable whereas some lines lying along the AEC like Ontofo, Renn, and P-226 were highly stable for yield across years and environments.

High stability is meaningful only when associated with mean performance. According to Figure 1.19, Ontofo was highly 'stable'. This means the relative performance of Ontofo was consistent, however it had very low yield. Ontofo was even poorer than the highly variable and least stable genotype Aa135 because Aa135 performed reasonably well in at least some environments. 'Stable' genotypes are desirable only when they have high mean performance for the trait of interest.

Vardim 1 had the highest mean 1000-seed weight followed by Ontofo and Line 340/11. PI 517924, W6 12723 and PI 517923 had mean 1000-seed weight similar to the grand mean and PI 343998 had the lowest mean 1000 seed weight. Ovation, Junior and Line 179 were seen as highly unstable whereas PI 517925, PI 517923, Granger were highly stable for 1000-seed weight (Figure 1.20).

L 805/5g had the highest mean protein across environments followed by PI 577142 and Aa135. P-226, MELROSE, W6 12713 had mean protein similar to the grand mean and Winter 1mm had the lowest mean protein. PS03101269, Windham, Ontofo are seen as highly unstable while P-226, Vardim 1, PI 639981 are highly stable for protein (Figure 1.21).

PI 639979 had highest winter survival followed by PI 517924, Romack and Fenn.

Ovation, Junior and Winter 1mm had very low winter survival across environments (Figure 1.22).

Figure 1.19. Average-environment coordination (AEC) view showing mean seed yield performance and stability for genotypes.

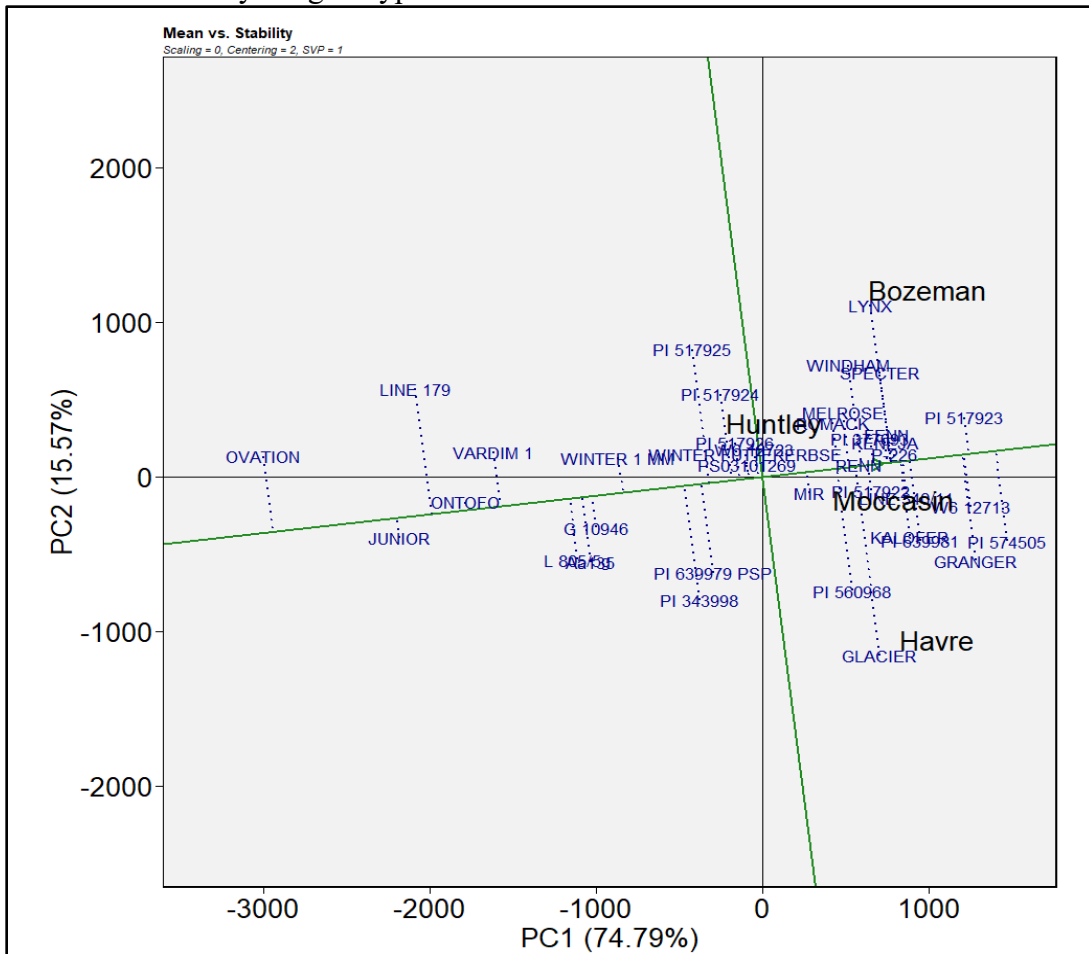
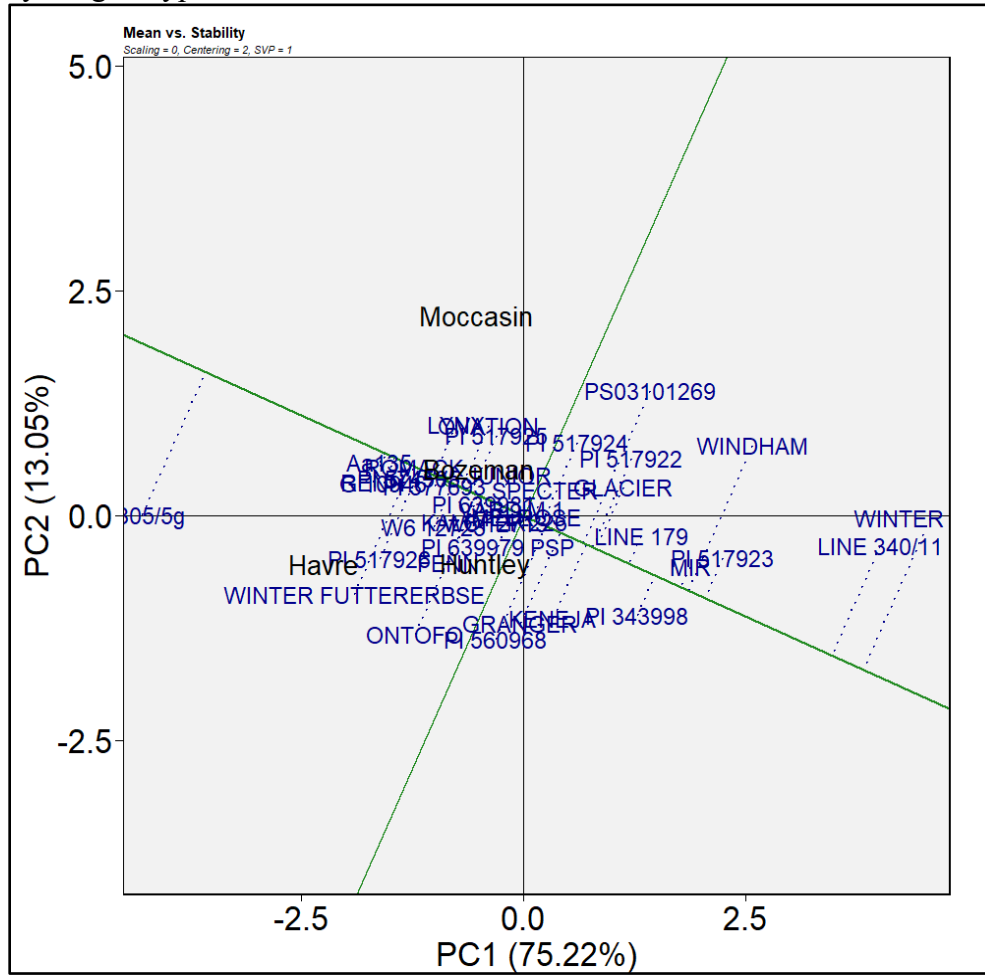




Figure 1.21. Average-environment coordination (AEC) view showing mean protein performance and stability for genotypes.

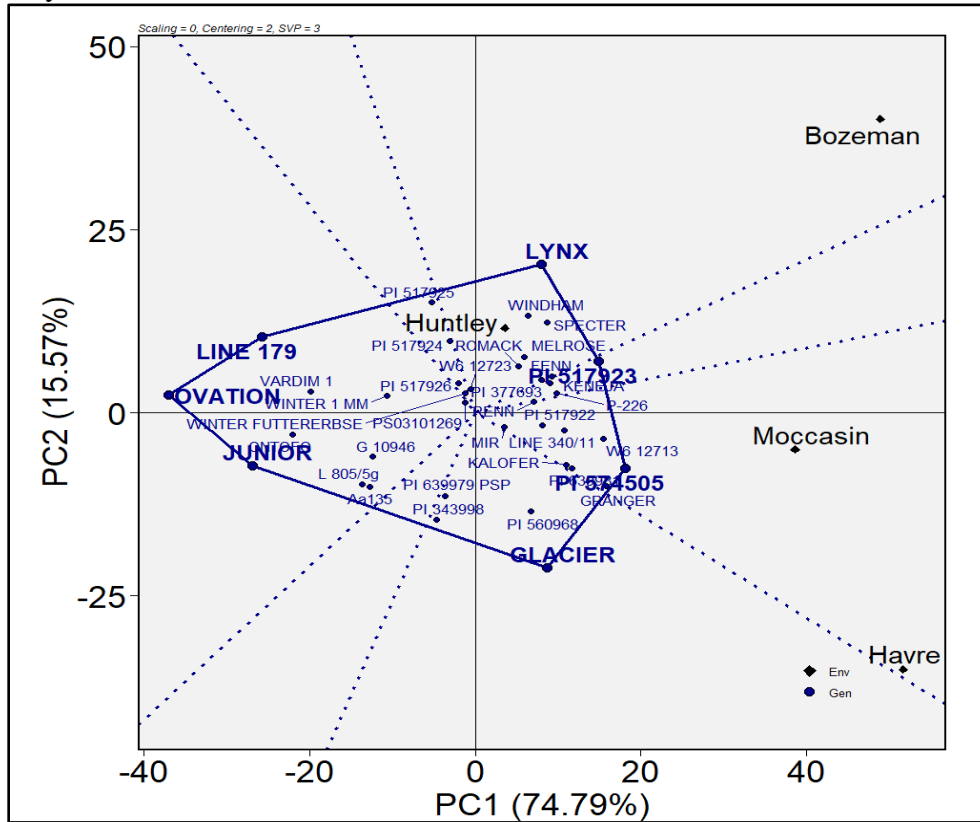




PI 574505 indicates that Lynx was better in Bozeman whereas PI 574505 was better in Moccasin. The equality line between Lynx and Line 179 indicates that Lynx was better than Line 179 in all environments. We saw PI 343998, Aa135, and L 805/5g were located on/near the line that connected Glacier and Junior. This means that the rank Glacier>PI 343998>Aa135>Junior was true in all environments.

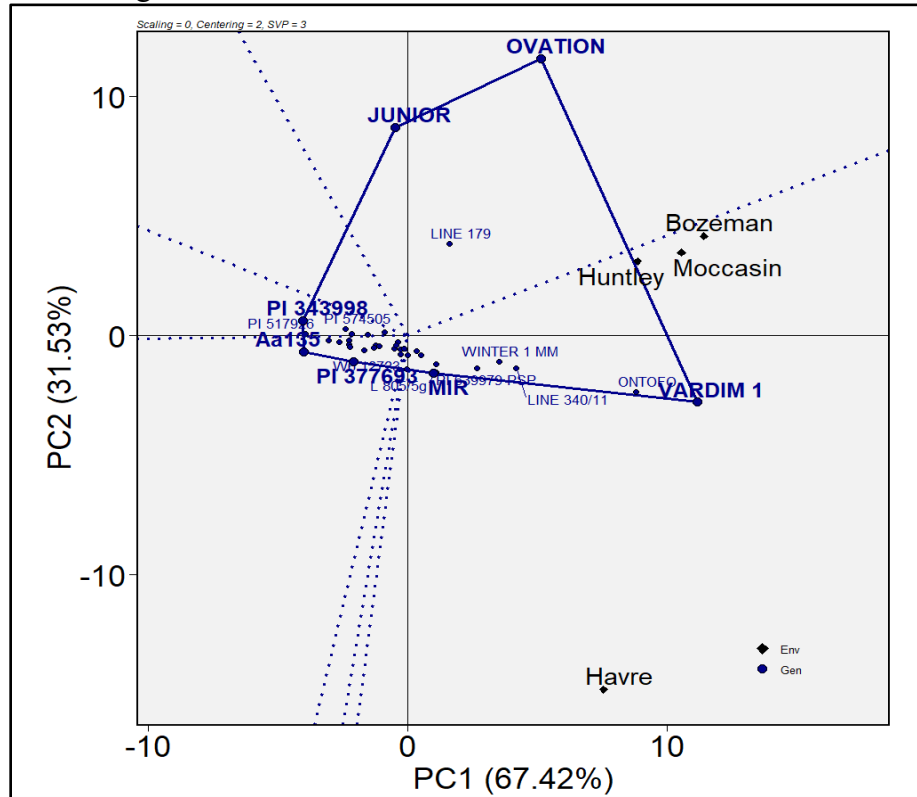
The equality lines divide the biplot into sectors, and the winning genotype for each sector is the one located on the respective vertex. From the Figure 1.23, the four environments fall into two sectors. Lynx was the winner in Bozeman and Huntley, and PI 574505 was the winner in Moccasin and Havre. This also suggests that the target environment may consist of two different mega-environments and that different varieties should be selected for each mega-environment.

Figure 1.23. The which-won-where view of the GGE biplot showing which genotype performed best for seed yield in which environments.



Vardim1, Ovation, Junior, PI 343998, Aa135, PI 377693 and Mir were located in the vertices of the polygon and performed either the best or the worst in one or all the environments for the thousand seed weight (Figure 1.24). Ovation, Vardim1 and Mir performed better in at least one of the locations whereas Aa 135, PI 343998 and PI 377693 had smaller seed size in all the locations.

Figure 1.24. The which-won-where view of the GGE biplot showing which genotype performed best for 1000-seed weight in which environments.



L 805/5g, LYNX, PS03101269, Winter 1mm, Line 340/11, PI 343998, PI 560968 and Ontofo were located on the vertices of the polygon and perform either the best or the worst in one or more environments for protein (Figure 1.25). L 805/5g performed better in locations Havre and Huntley, whereas Lynx performed better in Bozeman and Moccasin for protein. Some of the lines like Winter 1mm and Line 340/11 did not perform well in any location for protein.

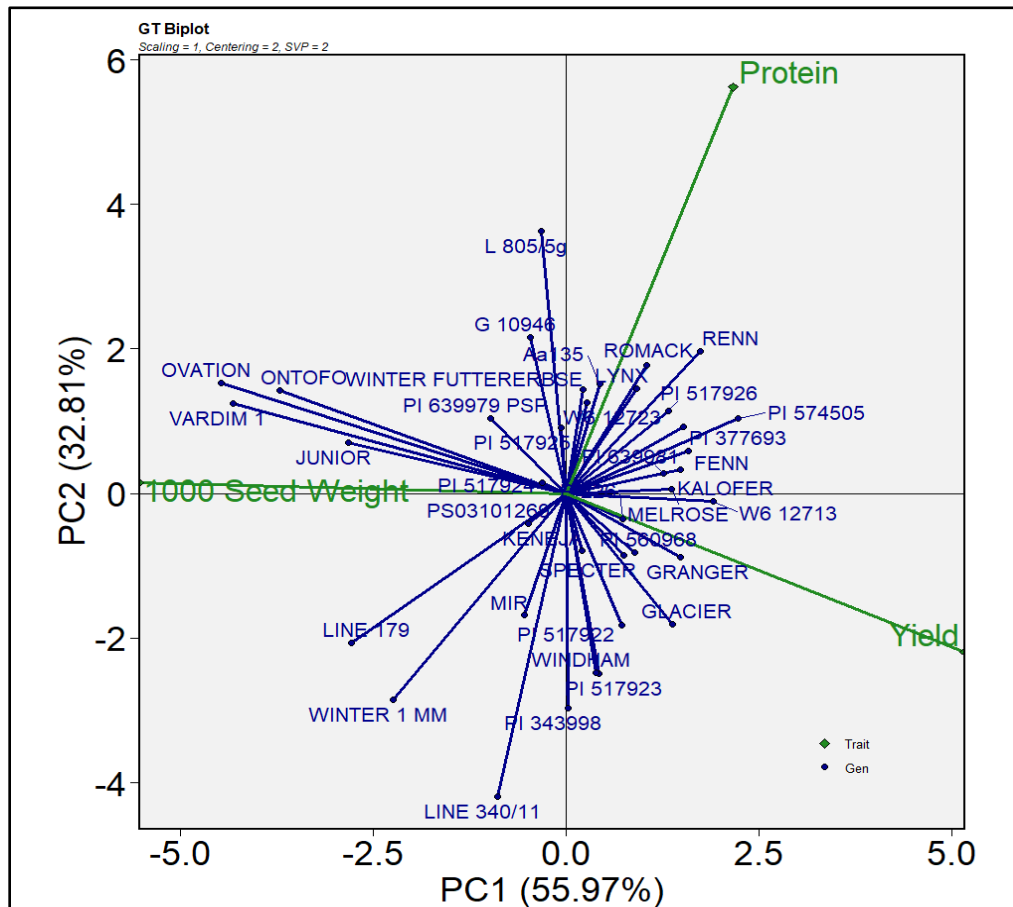




this method aids in identifying traits that could be effectively employed for indirect selection of another trait.

Further, the genotype-by-trait biplot facilitates a comprehensive visualization of the trait profiles of different genotypes. This visualization is critical as it allows for a detailed examination of the strengths and weaknesses inherent in each genotype. Understanding these aspects is crucial for making informed decisions in both parent selection and variety selection within the scope of genetic research and plant breeding (Yan & Kang, 2002).

Figure 1.27. Genotype-by-trait biplot representing genotypes measured for three traits: seed yield, 1000-seed weight, and protein. Data from 2021 to 2023 germplasm lines from four test locations.



The biplot in Figure 1.27 presents data of winter pea germplasm lines from four test environments (Bozeman, Havre, Huntley, and Moccasin) from 3 test years (2021, 2022, and 2023) for three traits (seed yield, thousand seed weight, and protein content). The axes, PC1 and PC2, represent the principal components that explain the largest portion of the variation in the dataset, with PC1 accounting for 55.9% and PC2 for 32.8% of the total variation. The proximity of a genotype to a trait vector indicates its performance for that trait. With the knowledge that higher yield, protein, and bigger seed size is desirable, the following visualization helps us to direct toward the best crosses that we should make to achieve the desired variety.

This biplot helps to visualize which genotypes stand out amongst others for a specific trait. Across the tested genotypes, seed yield and protein were found to be nearly independent of each other (near right angle), whereas seed yield was negatively associated with thousand seed weight (obtuse angle). The traits we were measuring were aligned towards three different sides and the lines are scattered in all directions. Thus, we were unable to find a line that performed the best for all of these traits. However, this biplot helped us to visualize the genotypes that can be selected for one or two traits. W6 12713 was a high-yielding line, intermediate in protein content, and lower than the average seed size. It would be ideal if W6 12713 had a bigger seed size. Vardim 1 was located opposite to W6 12713 relative to the biplot origin because its trait profile was opposite to that of W6 12713: it had the biggest seed size, intermediate protein content, and the lowest yield. It is, therefore, highly undesirable for cultivation. However, it might be a good parent for studying the genetic determination of seed size in peas. Therefore, W6 12713 may make a useful cross for this purpose.

L 805/5g had the highest protein, intermediate seed size, and lower-than-average yield. If it is desirable to improve further the protein of W6 12713, crosses of L 805/5g and W6 12713 may be useful. Similarly, Line 340/11 had an intermediate yield and seed size and the lowest protein content. The cross between Line 340/11 and L 805/5g may be useful.

Many relationships can be derived from this biplot. Genotypes like PI 517923, Windham, and PI 517922 constitute a group of genotypes with similar trait profiles; Ovation, Ontofo, Junior, and Vardim 1 form another group with similar trait profiles, etc. It would be rational to guess that the genotypes within each group share similar origins/parentages.

#### Seed Yield of Germplasm Lines for Three Years and Four Locations

Germplasm line with the greatest yield was W6 12713 (2367 kg/ha) followed by PI 574505 (2365 kg/ha) and Granger (2330 kg/ha). The average yield of all the germplasm lines was 1737 kg/ha. There was a considerable variation in yield among the lines with a coefficient of variation (CV) of 28% (Table A4). The 2023 experiments had the highest average yield (2681 kg/ha) followed by 2022 (1555 kg/ha) and 2021 (1197 kg/ha). Havre (2060 kg/ha) had the best average yield followed by Bozeman (2047 kg/ha), and Moccasin (1788 kg/ha) (Table A7).

#### Seed Size of Germplasm Lines for Three Years and Four Locations

Vardim 1 (238 g), Ontofo (216 g), and Line 340/11 (181 g) were the lines with the largest-sized seeds. However, these were also some of the lowest-yielding lines. The average 1000-seed weight for all the germplasm lines was 137 g (Table A4).

### Protein Content of Germplasm Lines for Three Years and Four Locations

Renn (26.4%), Winter Futterbse (26%) and Fenn (25.9%) had the highest protein content. The average protein content of all the germplasm lines was 22.9% with a CV of 14% (Table A4). Havre (27.3%) had the highest protein content followed by Huntley (25.8%) and Moccasin (25.2%). Bozeman (24.7%) had the lowest protein. Lines from 2022 (26.2%) had the highest protein content followed by 2021 (25.8%) and 2023 (25.1%).

### Seed Yield Ranking of Germplasm Lines by Year

PI 517923 was the highest-yielding line in the year 2023 and the highest-ranked line for yield for the overall three years. PI 574505 and W6 12713 were ranked second and third respectively for yield, consistently performing well in all the years. Ontofo, Junior, Line 179, and Ovation were consistently poor yielding in all three years (Table A5).

### Seed Yield Ranking of Germplasm Lines by Location

Granger, W6 12713 and PI 517923 were the best-ranked lines for overall locations. Line 179, Junior, and Vardim 1 did not perform well in any location and were consistently at the bottom in the yield ranking (Table A6).

### Correlation Matrix Analysis

Figure 1.28. Pearson correlation between a pair of traits.

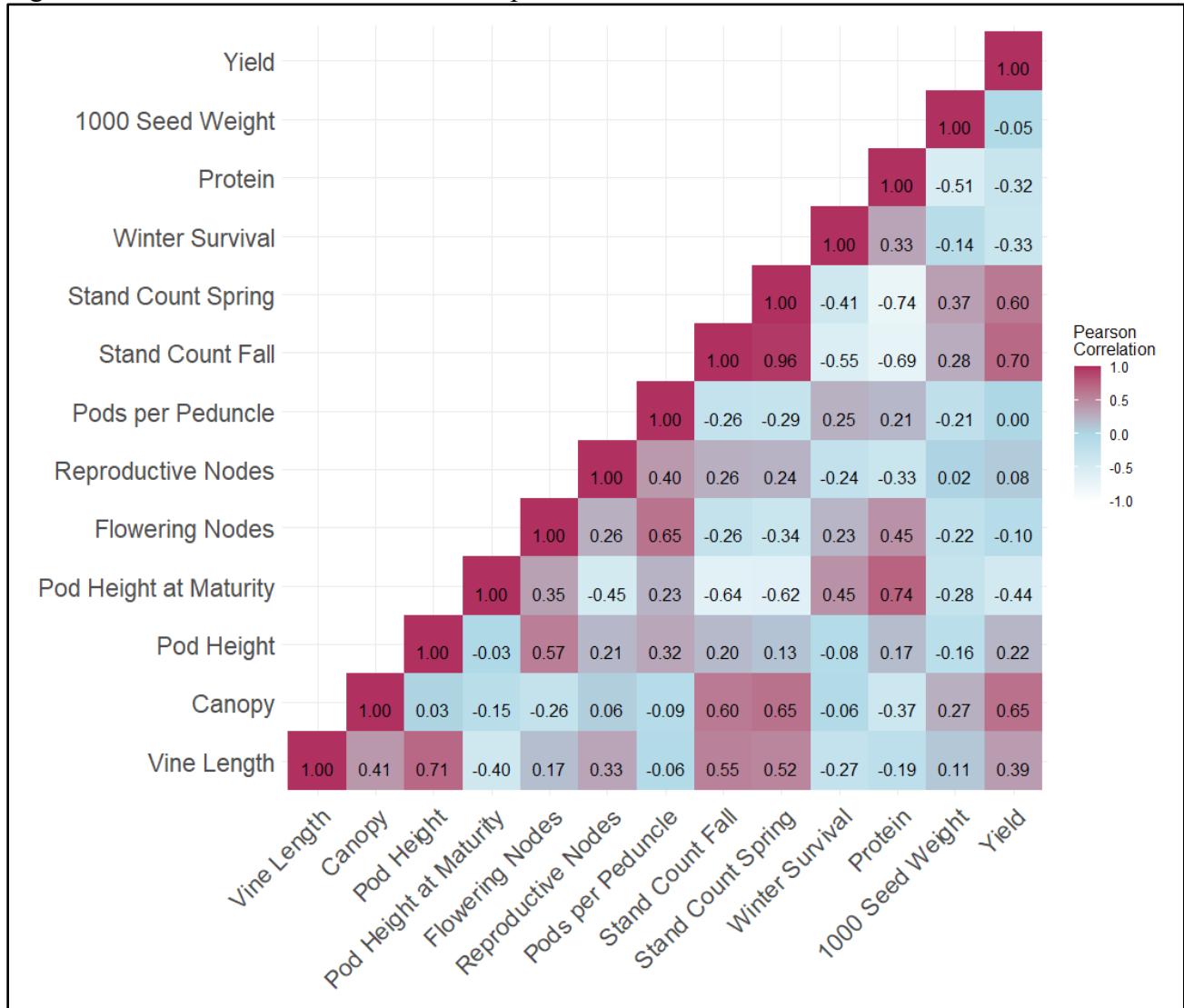


Figure 1.29 displays the Pearson correlation matrix for selected traits of pea germplasm lines of three years of data from Bozeman. The traits analyzed were seed yield, one thousand seed weight, protein content, winter survival, fall stand count, spring stand count, pods per peduncle, number of reproductive nodes, number of flowering nodes, pod height, pod height at maturity stage, canopy height, and vine length.

Seed yield was negatively correlated with 1000-seed weight and protein content, whereas it was positively correlated with the number of reproductive nodes, the height of the canopy, and the vine length. It showed taller plants tend to have higher yields than shorter plants. Winter survival showed a very weak positive correlation with spring stand count and a negative correlation with fall stand count. Winter survival was not considered the best measure to distinguish between the good winter hardy genotypes from the non-hardy ones. There was not a good germination percentage in the fall, while plants overwintered, went into dormancy, and only emerged in the spring. Thus, in some instances, there were a greater number of plants in the spring compared to what germinated in the fall. 1000-seed weight was positively correlated with fall and spring stand counts. Significant positive correlations, such as between yield and canopy height, pod height and vine length, as well as between flowering nodes and pod height, demonstrate consistent relationships across various environments. These correlations are notable for their potential in predictive modeling and trait selection strategies.

### Discussion

Germplasm lines used in the experiment displayed a broad range of agronomic performance. This multi-environment trial efficiently differentiated between superior and inferior lines and clarified the genotype by environment interaction, providing a better understanding of the underlying variations.

The seed yield of the germplasm lines saw a significant difference for each year and location. Yearly weather patterns of temperature, precipitation, and humidity would have accounted for this. Similarly, the differences in weather for each location might have caused the variation in seed yield for each location. The most variation in the yield was due to year than

other sources of variation. The agronomic practices also accounted for better seed yield at Bozeman, with the highest average seed yield. The proximity of the trial location also accounted for the extent of the standard agronomic practices that can be performed. Weed management was one of the major issues when the trial locations were located farther away. Peas compete very poorly with weeds at the early stage and account for up to 40% of yield reduction (Ullah et al., 2008). This was evident when we observed a nearly unmanageable amount of weed infestation in Huntley 2023, despite having a good plant stand. This resulted in low seed yield from that location. Similarly, some natural events like hailstorms caused a great yield and quality loss in Huntley 2022. Few environments also recorded the dormant seeding situation, where the fall-sown peas emerged in the spring and performed similarly to the spring peas. This dormancy has been attributed to many reasons, one of the most important being low soil moisture level. This was particularly evident when there was very low to no rainfall in the fall. Weather patterns may have also played a significant role in seed yield. The year 2021 was one of the driest in recent years with 60 to 80% of the average rainfall depending on the location. This might have caused the lower-than-average yield from all the locations in that particular year. The study showed higher yield variation due to year than the genotypes and location. Also, location contributed the lowest to the total variation in yield. W6 12713, PI 517923, and PI 574505 were the best lines for yield and stability across years and locations.

In this study, seed size and protein content of the germplasm lines were measured and showed significant differences. Vardim1, Ontofo, and Line 340/11 were the ones with the highest 1000-seed weight. However, these were also some of the lowest-yielding lines, meaning they had very few seeds but were large-sized. Seeds obtained from Huntley 2022, the environment, which

was hit by hail, were tiny and of low quality. Protein content was found highest in Havre, which was also the location where the 1000 seed weight was lower than average. Protein content in 2021 Havre was up to 10% higher than other environments, which might be due to the combination of heat and drought in that year. It is also important that the evaluation of protein content in various pea genotypes should consider the size of the seed. It was however found that most of the variation in protein percentage was due to within-genotype differences, and the trade-offs between the seed size and protein were not evident (Arthur et al., 1991).

This study indicated a slight negative correlation of seed yield with both protein content (-0.32) and 1000 seed weight (-0.05). Similar results of negative correlations were found by (Gaur et al., 2016), on the protein content and seed yield in chickpeas. In general, a negative correlation between seed protein and seed yield was observed, although some of the genotypes had both high seed yield and high protein.

It is worth noting that the tillage practices and previous crops varied across the environments. Precipitation was also significantly lower than average in 2021 than in 2022 and 2023. Precipitation in 2021 growing season was 20 to 30% lower than average. These differences could have influenced the moisture and nutrient availability in the soil, which in turn affected seed germination, yield, and quality. Most of our trials were planted on standing stubble, which is considered better at trapping soil moisture and making it available to the seeds for faster germination (Triplett Jr. & Dick, 2008). However, in above-average wet and cool growing conditions, tillage prior to planting might result in faster germination and better yield of field peas (S. Stepanovic, 2018).

The performance of winter pea germplasm lines was compared to the spring pea variety

trials in the same environment and showed some interesting results. Spring pea had relatively higher seed yield in 2021 and 2022 than the winter trials. However, winter pea lines had higher CV (21%) as compared to the spring varieties (16%). The top 20 winter pea germplasm lines had similar yield as the spring varieties in both the years. In 2023, winter pea in Havre had higher yield (3742 kg/ha) than spring peas (2597 kg/ha). Similarly, winter peas had higher protein content than the spring peas in 2021 (26.6 vs 26.1%), 2022 (27.3 vs 26.4), and 2023 (27 vs 26.7).

We also dissected the genotype by environment interaction by visualizing it in a biplot. Our GGE biplot captured 90%, 98%, 88% and 87% of the total variation in seed yield, 1000-seed weight, protein content, and winter survival respectively, in two principal components PC1 and PC2. The discrimination and representativeness view revealed Havre as the best location for yield and protein whereas Bozeman and Moccasin performed better in seed size.

The genotype-by-trait biplot helped to identify which genotypes performed best for specific traits. However, the lack of a genotype that performs optimally for all three traits indicated a trade-off situation. This may be due to genetic limitations or environmental interactions that prevented a simultaneous optimization of yield, protein content, and seed size. The absence of a single genotype that excelled in all three traits is typical in plant breeding. Each genotype exhibits a unique combination of trait values, representing different breeding priorities. For instance, a genotype having a higher yield may be preferred in breeding programs prioritizing yield over protein content or seed size.

Genotypes were ranked on the basis of yield and stability for three traits. After considering the multivariate statistical results from the GGE biplot analysis, the germplasm lines were categorized into three major groups:

1. Highly stable and high yield potential

The germplasm lines falling under this group had high seed yield and stable production over the years and locations. This group of genotypes was considered to be suited to a range of environments without compromising the yield. PI 517923, W6 12713, and PI 574505 fall under this category.

2. Low stability and high yield potential

The germplasm lines falling under this group had high seed yield but not a stable production over the years and locations. This group of genotypes may be appropriate for a specific environment. PI 560968, PI 517924, and PI 517925 fall under this category.

3. Low yield and high stability

The germplasm lines falling under this group had lower than average seed yield but stable production over the years and locations. This group of genotypes may be ideal in a breeding program intended to improve certain phenotypes. PI 517926, W6 12723, and G 10946 fall under this category.

Breeders must decide which traits are most important for their goals and select genotypes accordingly. This decision is often based on market demands, environmental adaptability, and end-use of the crop. The insights from this study could lead to strategies such as targeted breeding, where specific genotypes are crossed to combine desirable traits, or genomic selection, where markers associated with the traits of interest are used to accelerate the breeding process. It underscores that breeding for a suitable winter pea is a balancing act that requires careful consideration of the genetic potential of genotypes, environmental influences, and the specific objectives of the breeding program.

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MULTI-ENVIRONMENT EVALUATION OF WINTER PEA  
BREEDING LINES FOR WINTER SURVIVAL AND YIELD  
STABILITY

Contribution of Authors and Co-Authors

Manuscript in Chapter 2

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Co-Author: Kevin McPhee

Contributions: Conceptualization, Funding acquisition, Project administration, Resources,  
Supervision, Writing – review & editing

Co-Author: Jason Cook

Contributions: Writing – review & editing

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Amrit Poudel, Kevin McPhee, and Jason Cook

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## Introduction

Field pea (*Pisum sativum* L.) is an important crop of agricultural systems worldwide, recognized for its nutritional value as a rich source of plant-based protein and significant role in enhancing sustainability in farming practices. The global demand for plant-based proteins is on the rise, with the plant-based protein market is projected to reach \$9.5 billion by 2025 (Thavarajah et al., 2023), highlighting the importance of crops like field pea in meeting this demand. Field peas offer a sustainable and gluten-free source of high-quality protein, essential for dietary diversification and global nutritional security. These legumes contribute to sustainable farming by fixing atmospheric nitrogen and offering economic benefits through reduced need for synthetic fertilizers (Kebede, 2021). This aids in lowering greenhouse gas emissions and enhancing soil health, moisture conservation, and yield stability in subsequent crops.

Winter pea emerges as a compelling and equally viable crop choice and presents a promising opportunity, particularly in regions characterized by harsh winter conditions such as Montana and the Northern Great Plains. It offers distinct advantages when sown as a fall crop, strategically leveraging the extended growing season prevalent in these areas (McGee et al., 2017). One of the primary merits of opting for winter pea lies in its ability to capitalize on the ample time available for establishment during the fall months. Winter pea possesses inherent resilience and adaptability, making it well-suited to thrive in cold and challenging environmental conditions of the region. It offers the advantage of reducing or eliminating the need for fallow periods, enhancing soil health, and preventing nutrient loss, which are common in conventional fallow systems (Chen et al., 2012).

The adoption of winter pea varieties poses certain challenges, primarily due to the harsh winter conditions that can affect crop survival and yield. The selection and breeding of winter-hardy pea varieties necessitate extensive research and trials across multiple environments and years to identify genotypes that can withstand the variable and often severe weather conditions. Multi-environment trials (MET) have been crucial in this regard, helping to understand the interactions between genotype, environment, and their combined effects on pea crop performance. These trials comprehensively assess the performance and interactions of genotypes across various environmental conditions. They also help in identifying robust varieties that can contribute to sustainable agricultural practices by ensuring crop resilience, reducing the need for chemical inputs, and supporting the continuity of food supply chains even in less hospitable climates (Lee et al., 2023).

Integrating recent research findings and leveraging advancements in agricultural technologies and breeding techniques, it is possible to further enhance the viability and sustainability of winter pea cultivation in cold regions. The continued focus on developing nutritionally superior and environmentally resilient pea cultivars will benefit producers and consumers and support the broader objectives of sustainable agriculture and food security. The objective of this study was to understand the genotype-by-environment interaction (G x E) of winter peas breeding lines and find the best-suited winter pea lines in Montana.

## Materials and Methods

### Breeding Lines

The pulse crop breeding program at Montana State University (MSU) developed several breeding populations targeted at developing improved winter pea varieties. These were

developed from the germplasm lines based on the desirable traits that support winter survival. The lines had higher uniformity in seed size, flower color and vine length and were never selected for winter survival earlier in any environment. Two-hundred-three, fifty-one, and fifty breeding lines including a few checks were planted in the years 2021, 2022, and 2023, respectively, at Bozeman, Havre, Huntley, and Moccasin, Montana. The number of genotypes was dropped in the second year based on performance under different environmental conditions.

### Field and Experimental Design

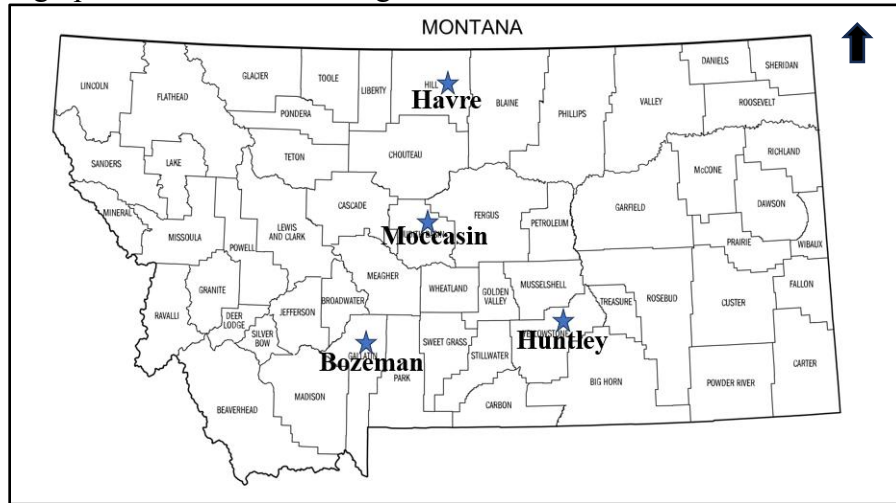
Winter pea breeding lines multi-environment trials spanning three years, from 2021 to 2023, were conducted in Bozeman, Havre, Huntley and Moccasin, Montana. The experiments were designed as a Randomized Complete Block Design (RCBD) with repeated checks and the number of replicates ranged from two to four depending upon location. In Bozeman, individual plots were established using a Wintersteiger XXL dynamic disc plus precision spaced plot seeder (Wintersteiger Inc., Salt Lake City, Utah). This seeder was equipped with six double disk openers spaced 22.5 cm apart. Each plot had a length of 3 meters. Other locations used different equipment based on the availability. Havre and Moccasin had the same plot dimension as Bozeman. The length of the plots was 6 meters in Huntley with same row spacing as other locations. Planting density was 120 plants per square meter. CruiserMaxx was used to protect against insects and fungi at 2.5 ml per kg of seed. Seeds were inoculated before planting using *Rhizobium leguminosarum* biovar *viceae* ( $2 \times 10^8$  CFU/g) inoculant. No fertilizer was used in the trials. Field trials were hand weeded as needed and standard agronomic practices were used at each location.

### Data Collection

Data collected included fall stand count, spring stand count, date of 10% bloom, date of 90% bloom, lodging score, canopy height at maturity (cm), maturity date, seed yield (kg/ha), 1000-seed weight (g), moisture (%), and protein content (%). Vine length (cm), number of flowering nodes, number of reproductive nodes, height of lowest pod (cm), height of lowest pod at maturity (cm), number of pods per peduncle were measured by selecting two random plants from two interior rows of each plot. The protein content was measured using near-infrared (NIR) spectroscopy with the Infratec™ 1241 Grain Analyzer (Foss North America, Minneapolis, Minnesota), which had been calibrated using a database of more than 50,000 cross-checked samples gathered over a span of more than two decades of harvests. A meter stick was used to record the number of plants in a meter of a plot. Fall stand count was collected after emergence of seedlings (September-October) and spring stand count after regrowth was observed in spring (April-May). Winter survival was recorded as the ratio of the number of plants survived in the spring to the number of plants emerged in the fall and expressed in percentage. Seed yield and 1000-seed weight were measured in kg/ha and g, respectively. There was complete winterkill in Havre in 2021, so no data was obtained.

## Trial environments

Figure 2.1. Geographical location of testing environments.



The trial sites were located across Montana (Figure 2.1). Bozeman and Havre are located on the southern part of the state, whereas Havre is located to the north, and Moccasin is centrally located. Every location has its own distinctive climatic characteristics posing a unique opportunity and challenge in the multi-environment trial (Table 2.1).

Table 2.1. Characteristics of the trial environments.

Year	Location	Latitude & Longitude	Altitude	Avg	Avg	Avg	Soil Type
			m		Max	Min	
				-----°C-----			
2021	Bozeman	45°40'27.5"N 111°08'48.4"W	1455	7.4	14.7	0.7	Silt loam
	Havre	48°29'33.3"N 109°48'19.1"W	820	7.1	14.4	-0.5	Telstad loam
	Huntley	45°55'15.3"N 108°14'22.1"W	920	8.1	16.3	-0.5	Clay loam
	Moccasin	47°03'21.6"N 109°57'17.4"W	1280	7.5	12.9	-0.4	Clay loam
2022	Bozeman	45°40'28.1"N 111°09'21.9"W	1448	7.6	14.7	0.6	Silt loam
	Havre	48°29'32.7"N 109°48'05.5"W	825	6.6	14	-1.2	Telstad loam
	Huntley	45°55'30.2"N 108°14'13.4"W	920	7.9	16.3	-1.4	Clay loam
	Moccasin	47°03'36.8"N 109°57'22.8"W	1280	7.8	12.3	-0.1	Clay loam
2023	Bozeman	45°40'28.8"N 111°08'59.6"W	1458	5.7	12.3	-0.9	Silt loam
	Havre	48°29'30.4"N 109°47'56.5"W	820	5.4	12.2	-1.3	Telstad loam
	Huntley	45°55'20.6"N 108°14'14.0"W	920	7	14.9	-0.8	Clay loam
	Moccasin	47°03'31.3"N 109°57'11.4"W	1280	5.3	11.4	-1	Clay loam

### Weather Data

Weather data was downloaded for the latitude and longitude of each environment using the North American Land Data Assimilation System Phase 2 and Western Regional Climate Center (WRCC). The following variables were downloaded: precipitation, maximum air temperature, minimum air temperature, average air temperature, relative humidity, and average wind speed. Growing degree days (GDDs) were calculated as the average daily temperature above 32°F, which is the most used base temperature for winter crops (Miller et al., 2001).

Figure 2.2. Average air temperature (°C) for each research locations.

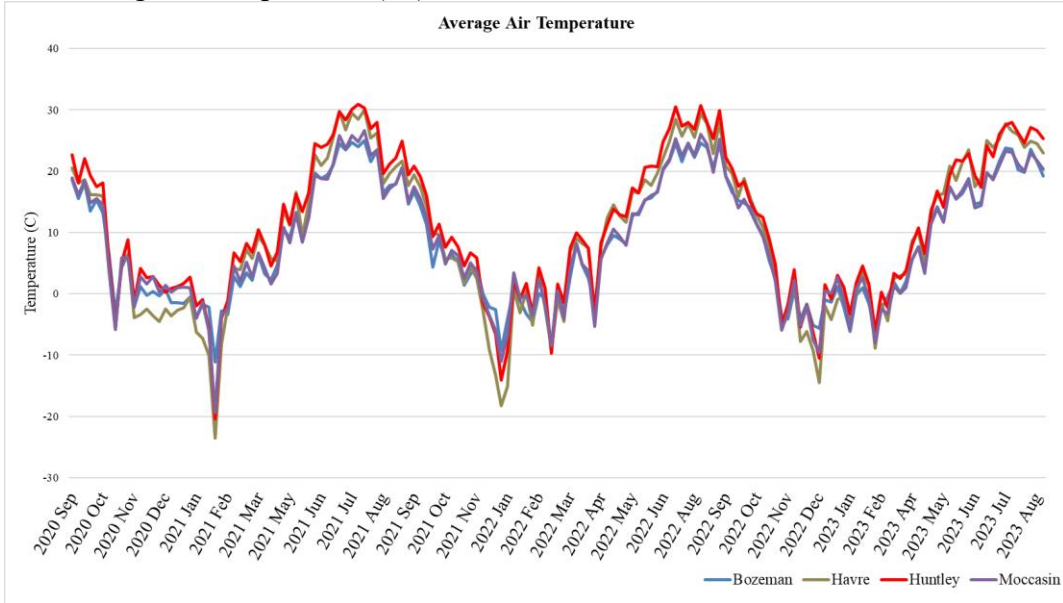


Figure 2.3. Average maximum air temperature (°C) for each research locations.

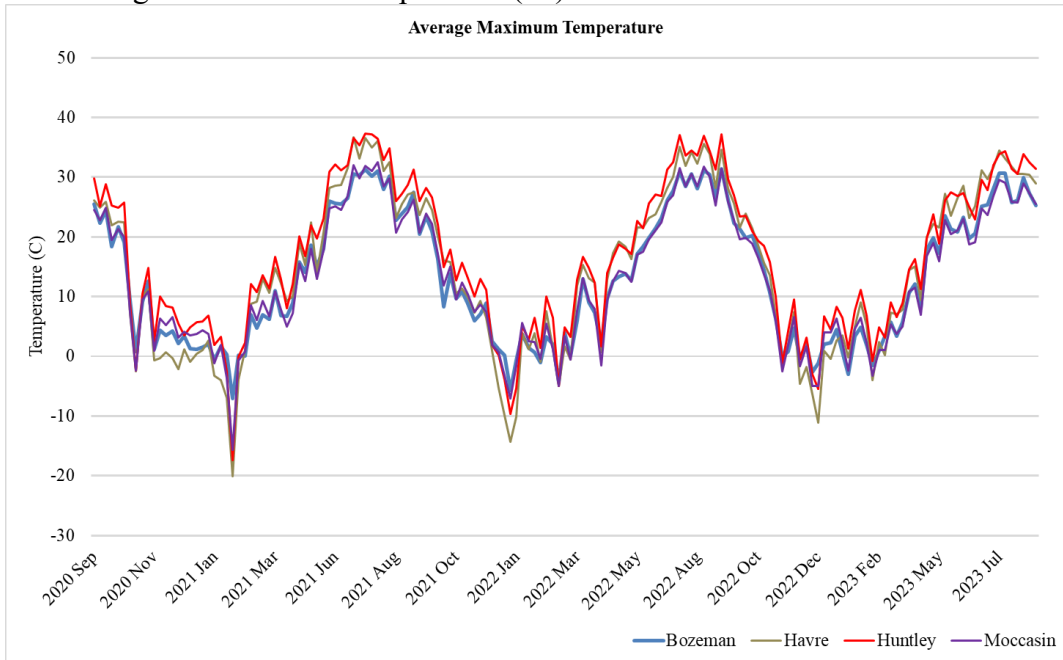


Figure 2.4. Average minimum air temperature (°C) for each research locations.

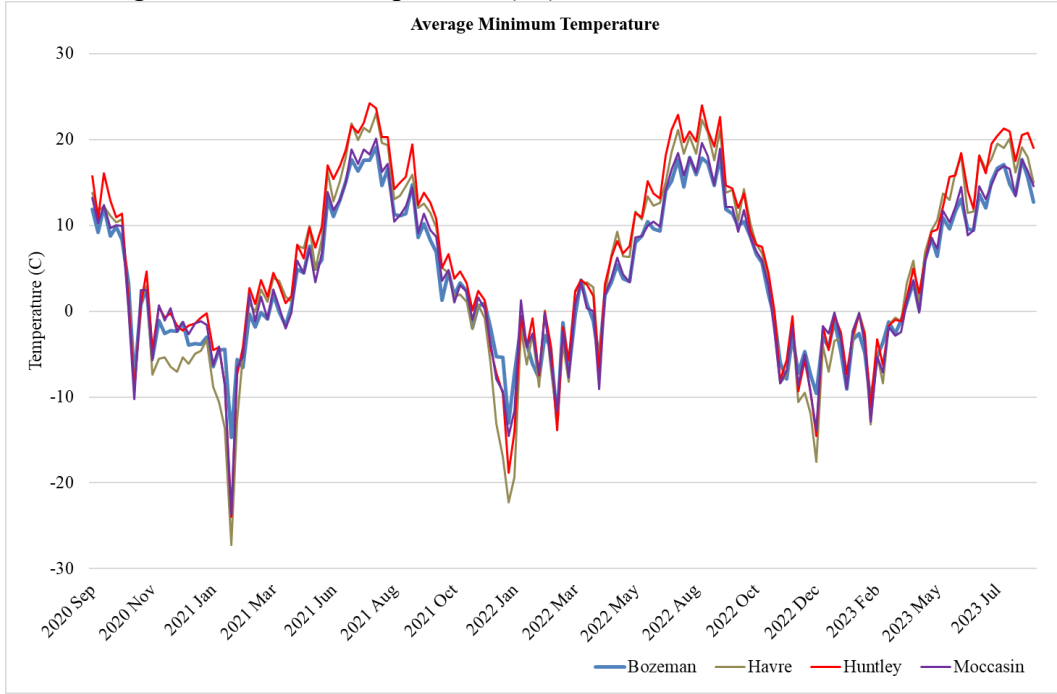
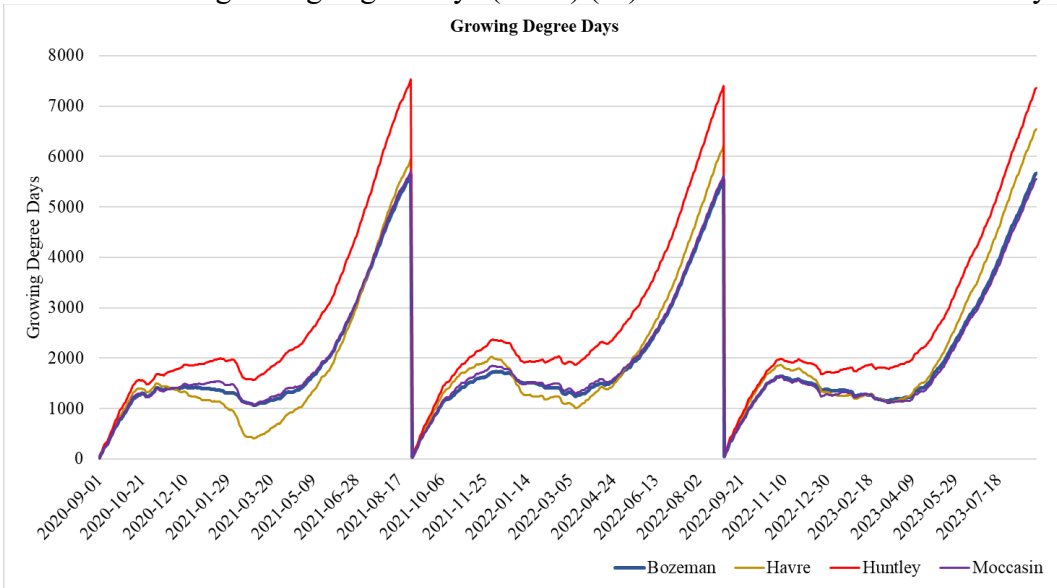


Figure 2.5. Cumulative growing degree days (GDD) (°F) of research locations for each year.



### Statistical Analysis

The dataset was filtered for missing values before analysis. To accommodate the intricacies of our MET design, we employed a generalized linear mixed model (GLMM), which is particularly suited for analyzing data that involves multiple levels of random variation. The effect of genotypes, locations, years, and interactions were considered fixed, whereas block nested over location was considered a random effect. This model allowed us to partition the variability due to specific genotypic performance, environmental effects, and their interactions, while simultaneously managing the random effects introduced by the blocks. Analysis of variance (ANOVA) was used to test the significance of main effects and quantify the interactions among and within the source of variation in seed yield, 1000 seed weight, protein content, and winter survival using the *lme4* package and mean separation used Tukey's HSD. The effects were considered statistically significant at  $P < 0.01$ . All analyses were done in R studio (RStudio 2023.06.0).

The core of our statistical analysis was the GLMM, constructed to dissect the influence of genotypes, years, locations, and their interactions on yield, 1000-seed weight, protein content and winter survival. The model for each trait was structured as follows:

$$\text{trait}_{ijkl} = \mu + \text{name}_i + \text{year}_j + \text{location}_k + (\text{name} \times \text{year})_{ij} + (\text{name} \times \text{location})_{ik} + (\text{year} \times \text{location})_{jk} + (\text{name} \times \text{year} \times \text{location})_{ijk} + b_l(\text{bloc}) + \epsilon_{ijkl}$$

In this equation,  $\text{trait}_{ijkl}$  represents the observed value for the  $i^{\text{th}}$  genotype in the  $j^{\text{th}}$  year at the  $k^{\text{th}}$  location within the  $l^{\text{th}}$  block. The symbol  $\mu$  is the overall mean value for the trait, and  $\epsilon_{ijkl}$  is the residual error term, assumed to be normally distributed. By incorporating  $b_l(\text{bloc})$ , the model accounts for block-to-block variability, allowing for random fluctuations in the trait that are not explained by the fixed effects.

Pearson correlation was used to establish the relationship among the traits vine length, canopy, pod height, pod height at maturity, number of flowering nodes, number of reproductive nodes, number of pods per peduncle, fall stand count, spring stand count, winter survival, protein content, one thousand seed weight, and seed yield.

To understand which genotype performed best, genotypes were ranked based on the yield for each location and each year. They were also ranked overall for each location and year by adding up the ranks to determine the best-ranked genotypes.

However, the variance component alone was not sufficient to clarify the details of genotype by environment interaction. Additional statistical techniques involving multivariate analysis were required to understand the GEI more accurately. For that reason, we used the GGE biplot method to visualize the GE patterns and identify the ideal genotypes and environments for each trait. The methodology was described by (W. Yan et al., 2000), which is explained by the equation:

$$Y_{ij} - \mu - B_j = \lambda_1 \varepsilon_{1i} \eta_{1j} + \lambda_2 \varepsilon_{2i} \eta_{2j} + \epsilon_{ij}$$

where  $Y_{ij}$  is the genotypic value of genotypes  $i$  in  $j$  environment,  $\mu$  is the overall mean,  $B_j$  is the effect of the  $j$  environment,  $\lambda_1$  and  $\lambda_2$  are Eigenvalues associates with PC1 and PC2,  $\varepsilon_{1i}$  and  $\varepsilon_{2i}$  are the scores for the PC1 and PC2 axes for the genotype  $i$ ,  $\eta_{1j}$  and  $\eta_{2j}$  are the scores for PC1 and PC2 axes for genotype  $j$ .  $\epsilon_{ij}$  is the error associated with the model which is not explained by the first two PCs. All these analyses were performed using the Metan package in R (Olivoto & Lúcio, 2020).

For genotype evaluation, genotype-focused singular value partitioning (SPV=1) was used using the ‘mean versus stability’ option using package *metan* in R. For location evaluation, environment-focused singular value partitioning (SVP=2) was used (W. Yan, 2001).

The GGE biplot shows graphical images to illustrate genotype and environment interaction and genotype ranking based on mean and stability. The graphs generated are based on the evaluation of multi-environment (which-won-where pattern), evaluation of genotypes (mean versus stability), and ranking of the environments (discriminative versus representative). The which-won-where pattern also identified which genotype was the winner in a given set of environments.

The broad ecological or environmental condition that influences the performance and adaptation of crop varieties are called mega-environments. Within a single mega-environment, the objective of data analysis was genotype evaluation to identify high-performing and highly stable genotypes. Test environment evaluation was to find test environments that were both discriminating (informative) and representative of other environments. Whenever there was potential G x E, it was explored.

Biplots were based on the seed yield (kg/ha), thousand seed weight (g), protein content (%), and winter survival (%) data of 2021, 2022, and 2023 winter pea breeding lines which were tested at four locations (Bozeman, Havre, Huntley, and Moccasin) in MT.

## Results

### Univariate Analyses of Breeding Lines

The highest yield in 2021 was observed at Huntley followed by Moccasin and Bozeman (Figure 2.6). All locations had significantly different yield from each other. In 2022, Bozeman

had the highest and significantly different yield than the other locations. Huntley and Moccasin had similar yields.

Seed yield in 2023 at all locations was high and none of the locations was significantly different from the other. In Bozeman, the highest seed yield was recorded in 2022, whereas all other locations had their respective highest seed yield in the year 2023.

The highest yield was observed in 2022 Bozeman which is significantly different from the rest, followed by 2023 Huntley and 2023 Havre (Figure 2.8). Huntley and Moccasin in 2022 had the lowest yield among the tested environments. Bozeman and Moccasin in 2021 were also not different from each other, making them the lowest-yielding environments.

Largest seed size was obtained from Moccasin 2023 (Figure 2.9), whereas year 2022 had higher protein content and Havre consistently had higher protein than other locations (Figure 2.10).

Winter survival was used to determine the hardiness of the lines in different environmental conditions. At some instances due to dormancy, survival was greater than 100% because the number of plants in the spring were more than what emerged in the fall. Figure 2.11 presents a boxplot illustrating the distribution of survival rates (%) for three locations over a period spanning from 2021 to 2023. Bozeman 2022 had the highest winter survival (135%) whereas Huntley 2023 had the lowest winter survival (32%). Few lines like MTP190664 (253%), MTP190272 (144%), and MTP190766 (127%) had higher winter survival, whereas MTP190918 (48%), MTP190681 (56%), and MTP190583 (57%) had lower winter survival percentages.

Figure 2.6. Mean seed yield of breeding lines grown at four locations in 2021, 2022, and 2023. Letters above the bars represent statistical groupings.

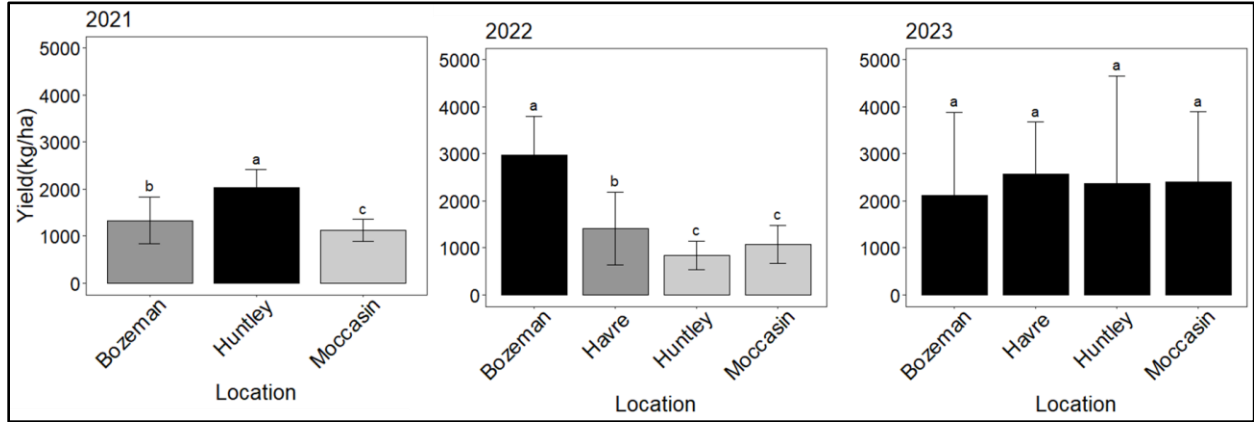


Figure 2.7. Mean seed yield of breeding lines across locations. Letters above the bars represent statistical groupings.

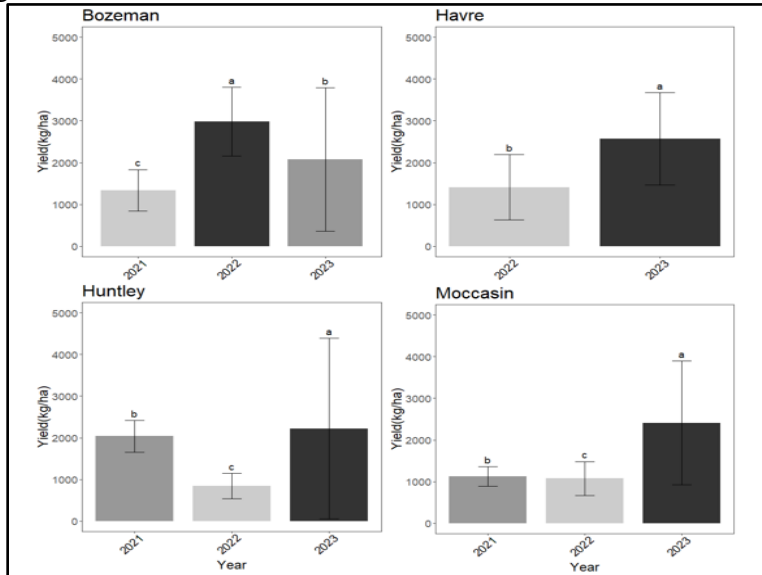


Figure 2.8. Mean seed yield of breeding lines across environments. Letters above the bars represent statistical groupings.

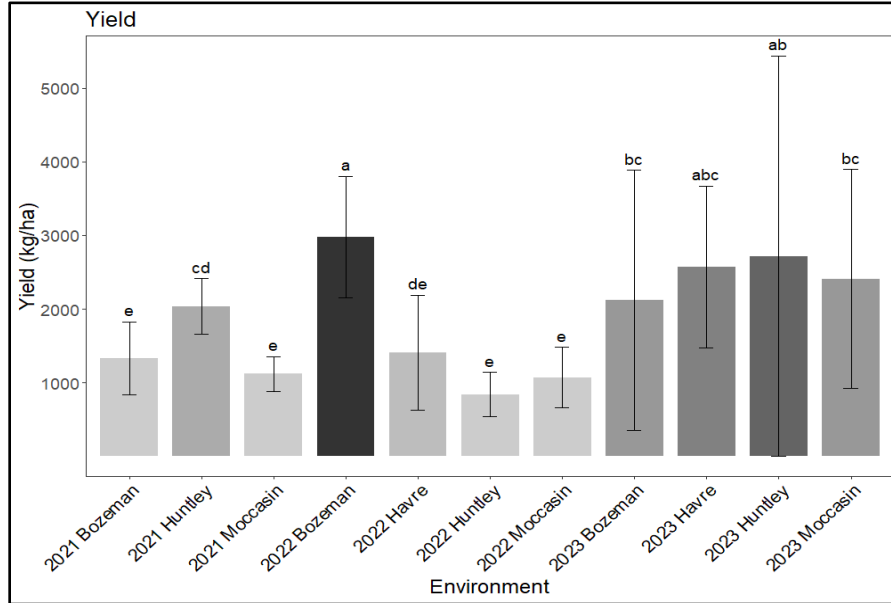


Figure 2.9. One thousand seed weight of breeding lines across environments. Letters above the bars represent statistical groupings.

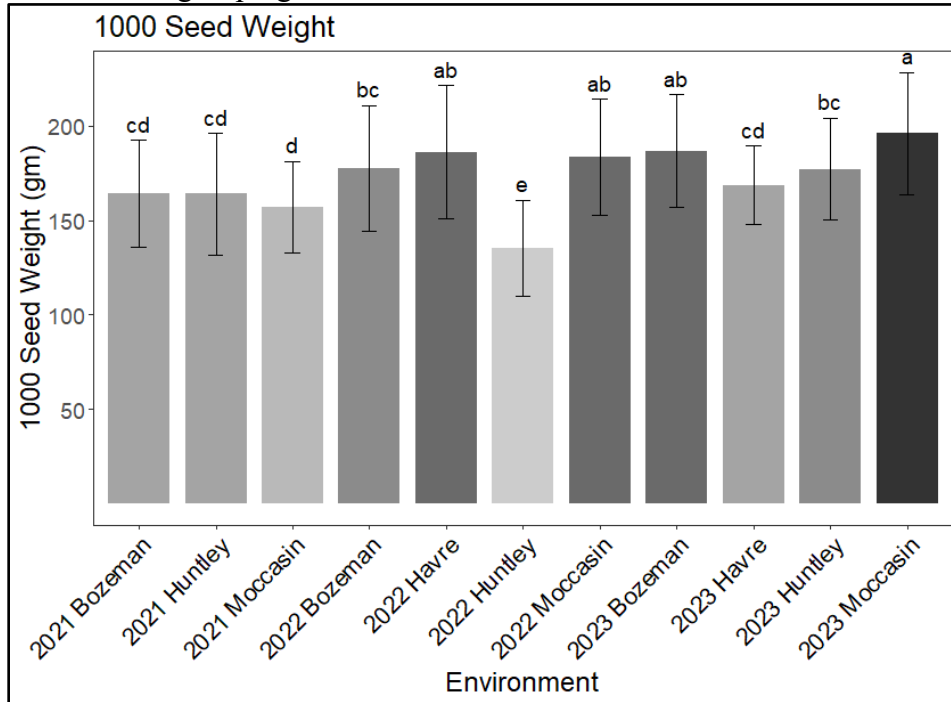


Figure 2.10. Protein content of breeding lines across environments. Letters above the bars represent statistical groupings.

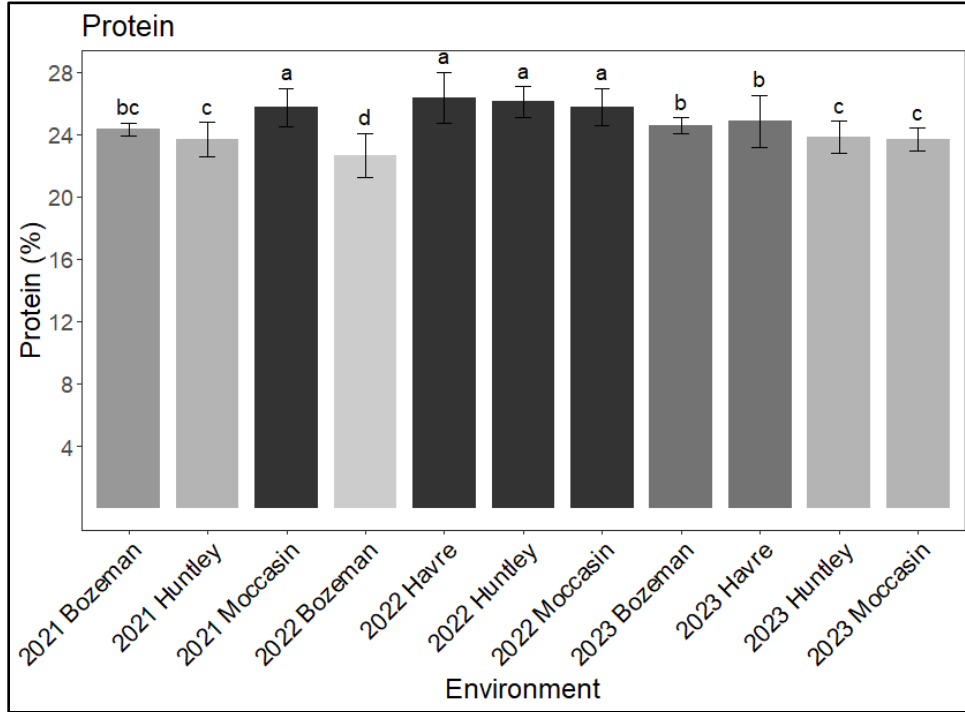
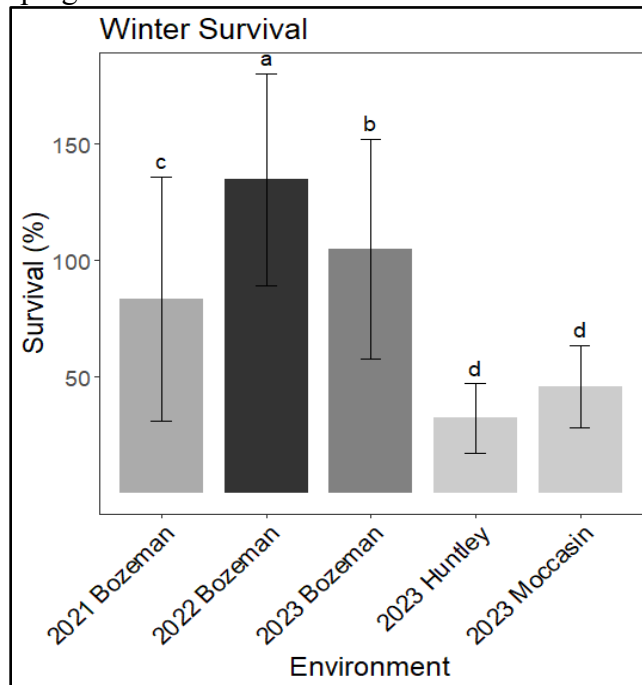


Figure 2.11. Winter survival of breeding lines across environments. Letters above the bars represent statistical groupings.



Analysis of variance was performed to describe the main effect and quantify the interactions among and within the sources of variations. The pooled analysis of variance is presented in Table 2.2. Genotype, year, genotype by location, genotype by year, and genotype by location by year showed significant differences ( $p < 0.01$ ) for seed yield, 1000 seed weight, and protein content. Location had no significant effect on all the traits examined. For winter survival, a significant difference was seen for year ( $p < 0.01$ ) and genotype by year ( $p < 0.05$ ). All other sources of variations were non-significant for survival.

Table 2.2. Analysis of Variance (ANOVA) table showing the effects of genotype, location, and year on seed yield, 1000-seed weight, protein content, and winter survival of breeding lines for three years across four locations.

Source	Yield		1000SW		Protein		Survival	
	df	<i>P&gt;F</i>	df	<i>P&gt;F</i>	df	<i>P&gt;F</i>	df	<i>P&gt;F</i>
Genotype	50	<0.01**	50	<0.01**	50	<0.01**	50	0.6
Location	3	0.3	3	0.98	3	0.9	2	1
Year	2	<0.01**	2	<0.01**	2	<0.01**	2	<0.01**
Genotype:Location	149	<0.01**	148	<0.01**	145	<0.01**	86	0.99
Genotype:Year	96	<0.01**	96	<0.01**	92	<0.01**	72	0.04*
Location:Year	5	<0.01**	5	<0.01**	4	<0.01**		NA
Genotype:Location:Year	109	<0.01**	98	<0.01**	77	<0.01**		NA

\*Significant at the 0.05 probability level.

\*\*Significant at the 0.01 probability level.

Biplot Analysis of Breeding Lines

Figure 2.12. The environment-vector view of the GGE biplot showing similarities among test environments in discriminating the breeding lines for yield (a), 1000-seed weight(b), protein (c), and survival (d).

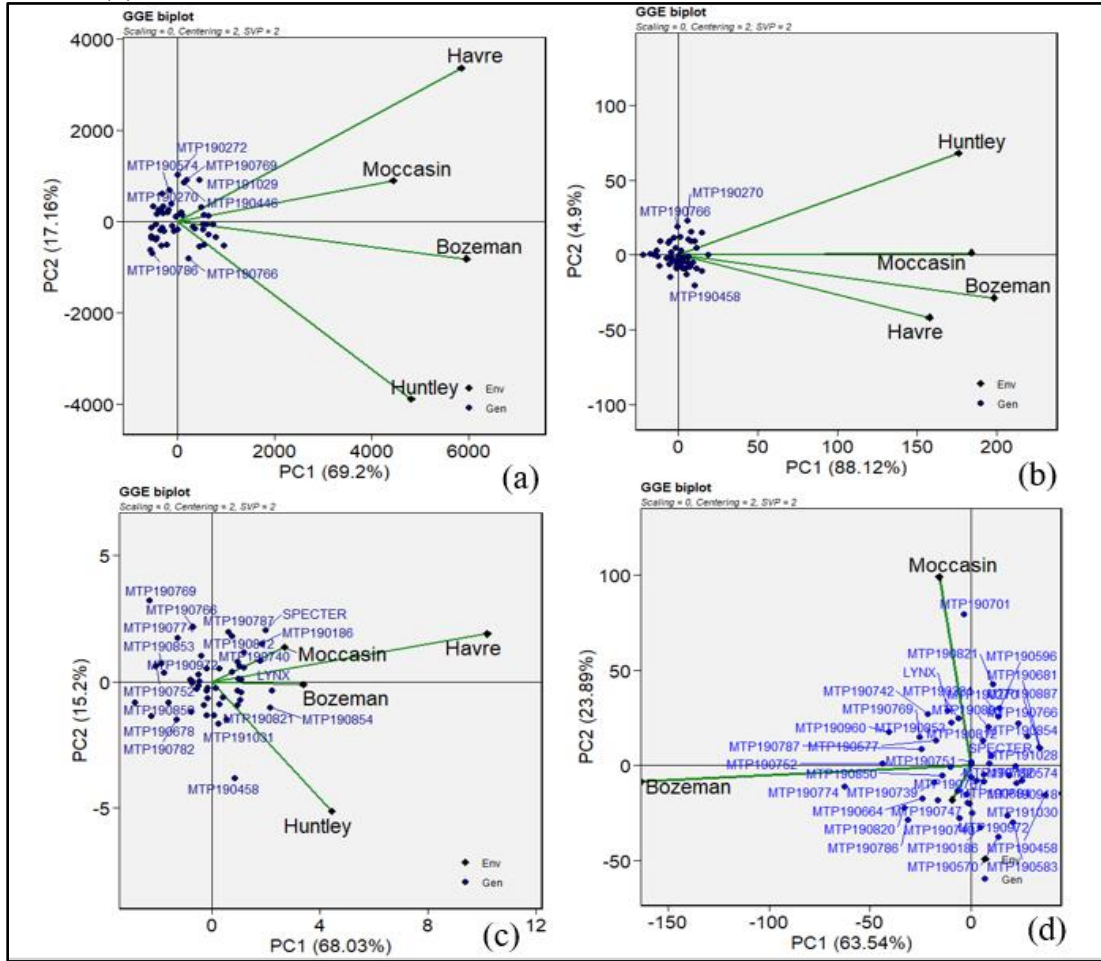


Figure 2.12 is the environment-vector view of the GGE biplot and is based on the environment-centered (centering = 2) G x E table without scaling (scaling = 0), and it is environment-metric preserving (SVP = 2). The axes are drawn to scale (default of GGE Biplot) and this biplot explains 86% of the total variation for yield, 93% for 1000 seed weight, 83% for protein, and 87% for survival.

The lines that connect the test environments (Bozeman, Havre, Huntley, and Moccasin) to the biplot origin are called environment vectors. The cosine of the angle between the vectors of two environments approximates their correlation. An acute angle between the two environment vectors shows a positive correlation between them, whereas an obtuse angle is an indication of a negative correlation between them. A larger obtuse angle shows the strong crossover G x E between those two environments, implying that G x E is moderately large. A right angle between two environments shows that they are not correlated with each other. In Figure 2.12 (a), all the environment vectors had an acute angle between them for yield, except for Havre-Huntley, which showed a positive correlation between each pair of environments. Havre and Huntley were nearly at a right angle, indicating they were not correlated to each other for yield. For 1000-seed weight and protein, there was a positive correlation between each pair of environments. However, for survival, only Bozeman and Huntley had positive correlation between them.

Figure 2.13. Discrimination and representative view of the GGE biplot showing discriminating ability and representativeness of the test environments for seed yield (a), 1000-seed weight(b), protein (c), and survival (d).

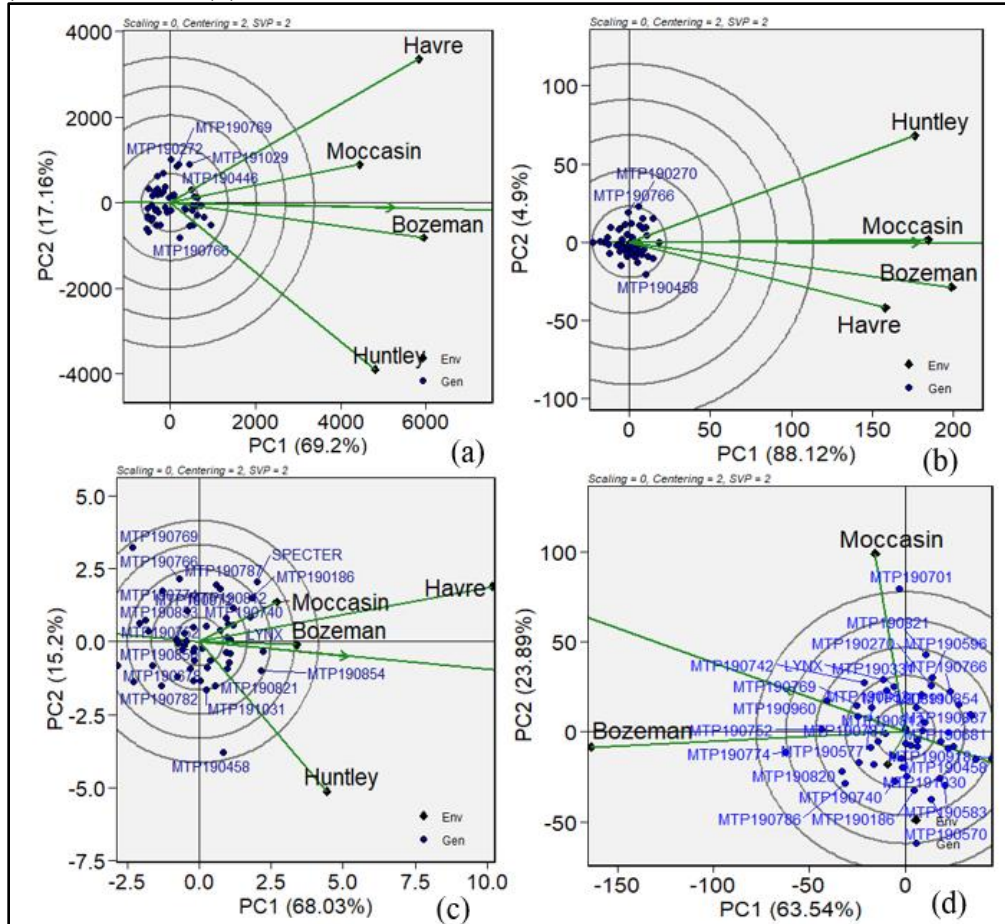


Figure 2.13 presents an Average Environment Axis (AEA), a line that passes through the average environment and the biplot origin. A test environment that has a smaller angle with the AEA is more representative of all the locations. Figures 2.13(a) and 2.13(b) showed Bozeman and Moccasin were closer to the AEA and were more representative than Havre and Huntley for yield and 1000-seed weight. Bozeman was closest to the AEA (Figure 2.13 (c)) and was the most representative of all environments for protein.

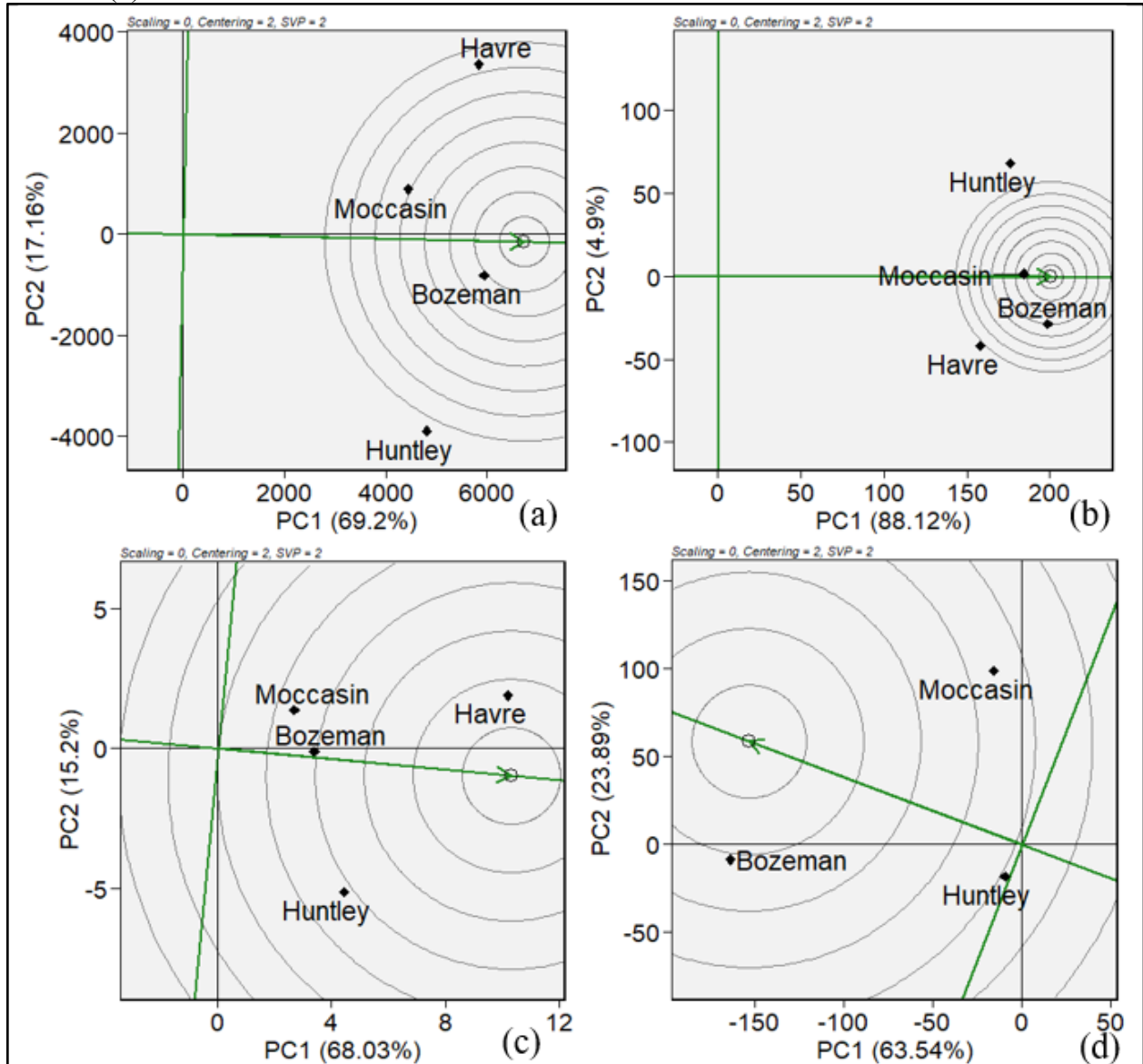
The concentric circles on the biplot help to visualize the length of the environment vectors. An environment vector that has the longest length is the most discriminative. Therefore,

from Figure 2.13, among the four environments, Havre was the most discriminating (informative) than the other locations for yield (a) and protein (c). Bozeman is both discriminating and representative for yield (a), thus is a good location for selecting generally adapted genotypes. Discriminating but non-representative test environments (e.g., Havre and Huntley in (a)) are useful for culling unstable genotypes. Non-discriminating test locations like Moccasin for protein (Figure 2.13 (c)) with a very short vector are less useful because they provide very little discriminating information about the genotypes.

### Ranking Environments

Figure 2.14 shows the ideal test environment which is the center of the concentric circles. It is a point on the AEA (most representative) which is farthest with a distance to the biplot origin equal to the longest vectors of all environments (most discriminating). Bozeman is closest to this point, and is, therefore, best, whereas Huntley is the poorest for selecting cultivars adapted for yield (a). Similarly, Moccasin and Bozeman are better than Havre and Huntley for 1000 seed weight. Similarly, Havre was better than other locations for protein content (c), whereas Bozeman performed better in winter survival (d).

Figure 2.14. Discrimination and representative view of the GGE biplot ranking test environments relative to an ideal test environment for seed yield (a), 1000-seed weight(b), protein (c), and survival (d).



### Ranking Genotypes Relative to the Ideal Genotype

An ideal genotype should have both high mean performance and high stability across environments. Figure 2.15 identifies an ‘ideal’ genotype (the center of the concentric circles) to be a point on the AEA (‘absolutely stable’) that has a vector length equal to the longest vectors of the genotypes (‘highest mean performance’). So, the genotypes located closer to the ‘ideal genotype’ are more desirable than others. Thus, MTP190753 is more desirable than MTP190766 for yield even though the latter had a higher average yield. MTP190960, MTP190674, MTP190383, etc. are the poorest genotypes because they are consistently the poorest yielding lines.

High stability is meaningful only when associated with mean performance for the trait of interest. According to Figure 2.15 (a), MTP190749 was highly ‘stable’. This does not necessarily mean MTP 190749 was any good; it only means the relative performance of MTP190749 was consistent. ‘Stable’ genotypes are desirable only when they have high mean performance for the desired trait.

MTP190739 was located closest to the center of the concentric circles in Figure 2.15 (b). So, it was the best for the 1000 seed weight. Genotypes like MTP190742 and MTP190960 were also located closer to the center, so they were good for 1000 seed weight, whereas MTP190854 and Specter located farthest from the center of the concentric circles were the ones with the smallest seed size.

MTP190854 and MTP 190331 were the best for protein content (Figure 2.15 (c)). MTP190769, MTP190853 and MTP190752 did not have good protein content. Similarly, MTP190960 and MTP190742 had higher winter survival (Figure 2.15 (d)).



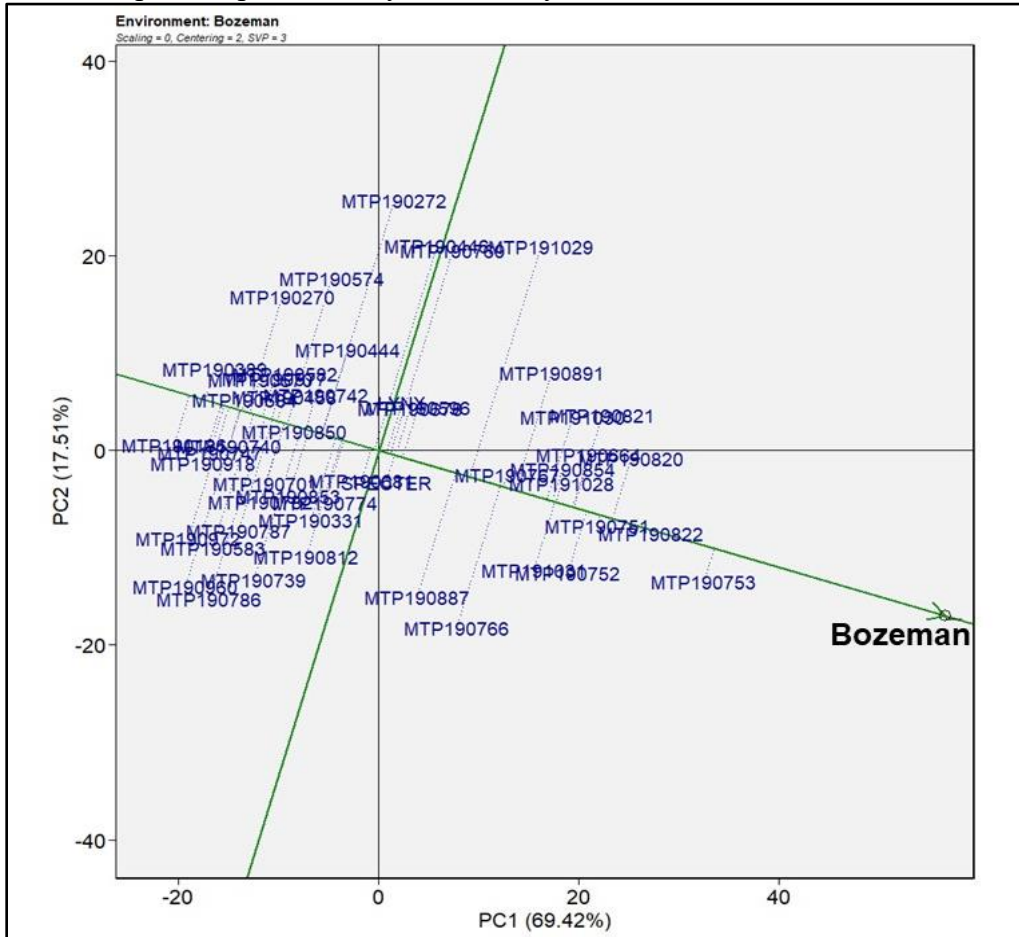
### Examine Environment Based the Seed Yield of Breeding Lines

The axis, a line drawn from the biplot origin to the environment, is used to assess the mean seed yield and stability of genotypes for that environment, by ranking the genotypes along it.

### Bozeman

In Bozeman, MTP190753, MTP190822, MTP190751, and MTP190752 had higher than average yield, MTP190850, MTP190684, MTP190570, and MTP190270 had near average yield, while MTP190389 and MTP190747 had lower than average yield. Figure 2.16 shows the highest-yielding breeding line in Bozeman across three years was MTP190753.

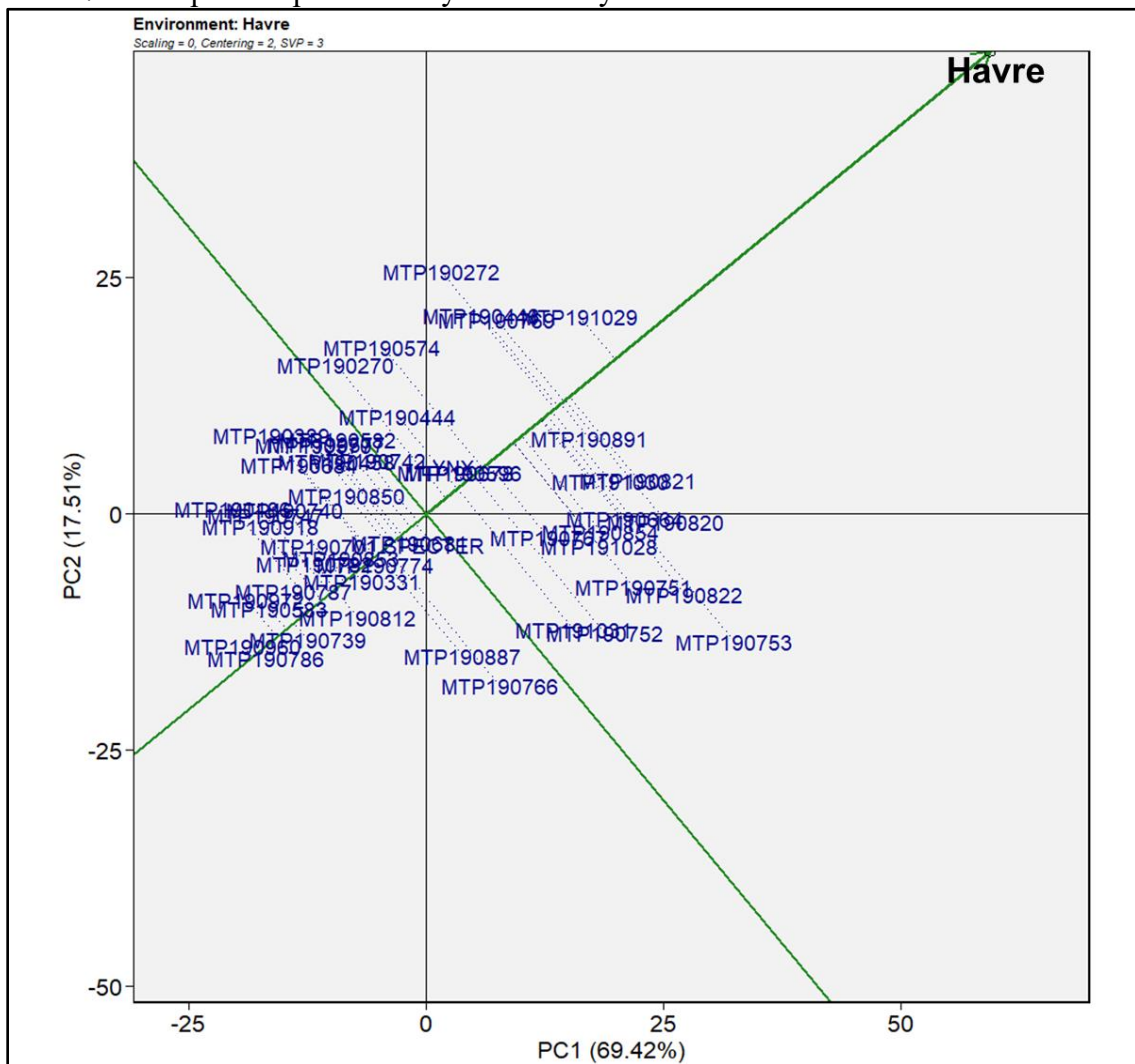
Figure 2.16. Principle component analysis of seed yield at Bozeman.



Havre

In Havre, MTP191029, MTP190272, and MTP190169 had higher-than-average yield. MTP190684, MTP190797, and MTP190766 had average yield, whereas MTP190786 and MTP190960 had lower-than-average yield. From Figure 2.17, the highest-yielding breeding line in Havre was MTP191029.

Figure 2.17. Principle component analysis of seed yield at Havre.

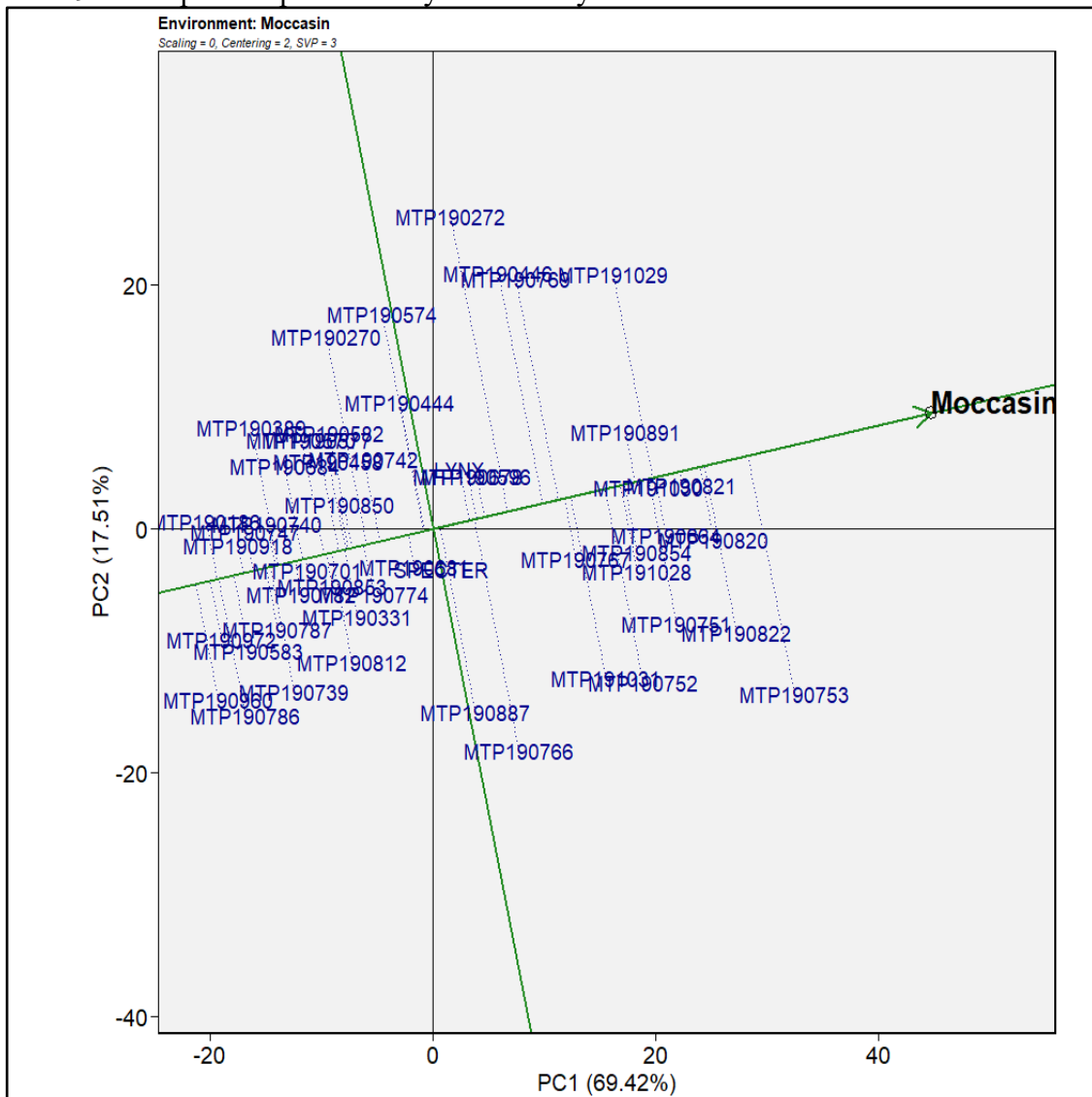




Moccasin

In Moccasin, MTP190753, MTP190822, and MTP190821 had higher than average yield, MTP190782, MTP190684, and MTP190570 had near average yield, while MTP190960 and MTP190972 had lower than average yield. The highest-yielding breeding line in Moccasin was MTP190753 (Figure 2.19).

Figure 2.19. Principle component analysis of seed yield at Moccasin.



### Mean vs Stability

Breeding lines used in the experiment were evaluated for both mean performance and stability across environments. Figure 2.20 shows the average environment coordination (AEC) view of the biplot.

The horizontal green line is the AEC abscissa (or AEA). It points to the higher mean yield across environments. From Figure 2.20, we can see that MTP190753 had the highest mean yield across all environments followed by MTP190822 and MTP190820. MTP190272 and MTP191081 had mean yields similar to the grand mean and MTP190186 had the lowest mean yield.

A vertical green line is the AEC ordinate. It shows greater variability (poor stability) in either direction. The dotted line drawn from the AEC to the genotype reflects the stability of the genotype. The longer the line, the less stable the genotype is, and vice versa. From Figure 2.20, we saw that MTP190272 and MTP191029 were highly unstable whereas some lines lying along the AEC including MTP190854 and MTP190767 were highly stable for yield across years and environments.

High stability is meaningful only when associated with mean performance. According to Figure 2.20, MTP190850 was highly stable. It does not necessarily mean MTP190850 was any good because it had a very low yield; it only means its relative performance was consistent. ‘Stable’ genotypes are desirable only when they have high mean performance.

MTP190739 had the highest mean 1000 seed weight followed by MTP190960 and MTP190742. MTP190766 and MTP190774 had a 1000 seed weight close to that of the grand mean, whereas genotypes such as MTP190854 and MTP191029 had the lowest 1000 seed

weight. MTP190270 and MTP 190766 were highly unstable genotypes whereas MTP190739 and MTP190751 were highly stable genotypes for 1000 seed weight (Figure 2.21).

MTP190854 and MTP190331 had highest protein content and were also stable (Figure 2.22). Similarly, MTP190774, MTP190960 and MTP190752 had higher winter survival (Figure 2.23). MTP190577 and MTP190753 had average but stable winter survival across years and locations.

Figure 2.20. Mean performance and stability of genotypes based on seed yield visualized with the average environment coordination (AEC) view.

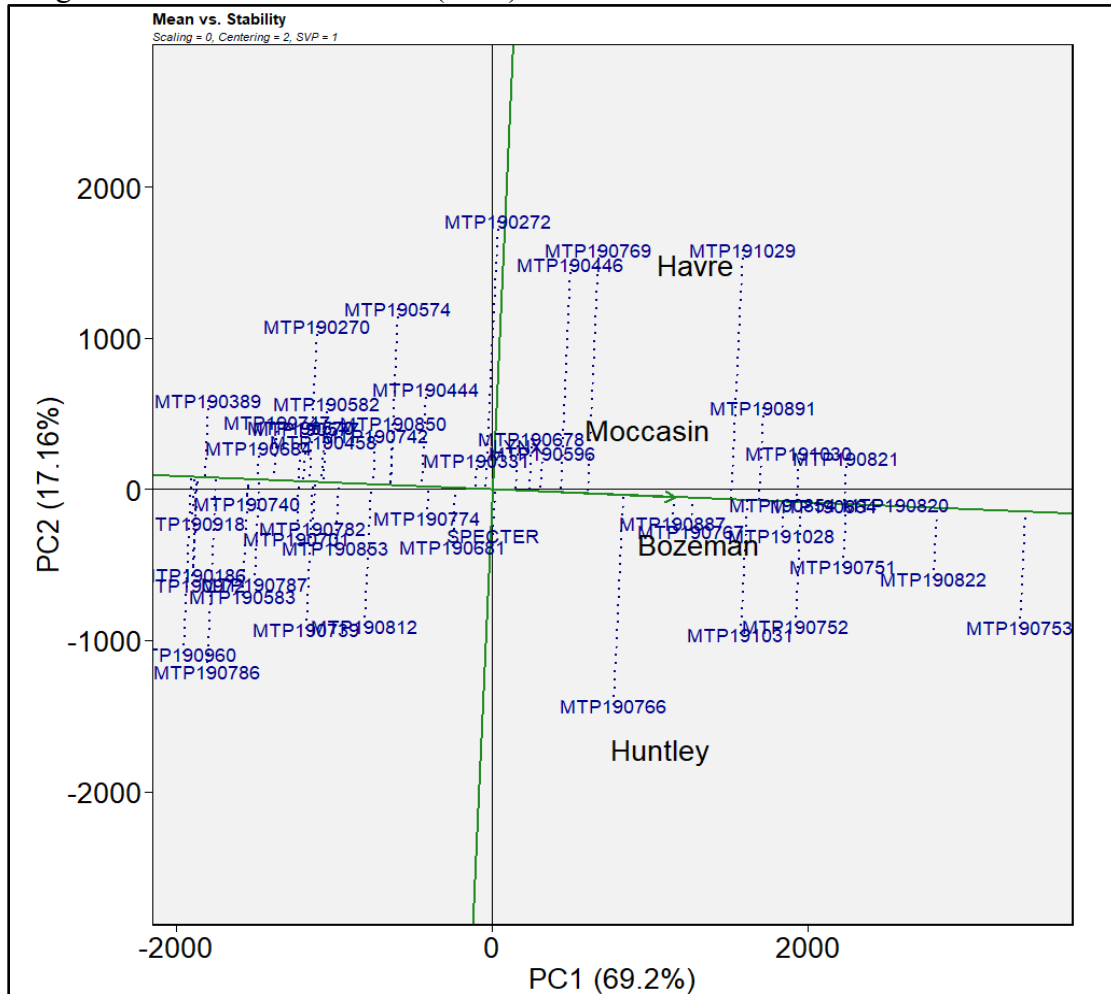
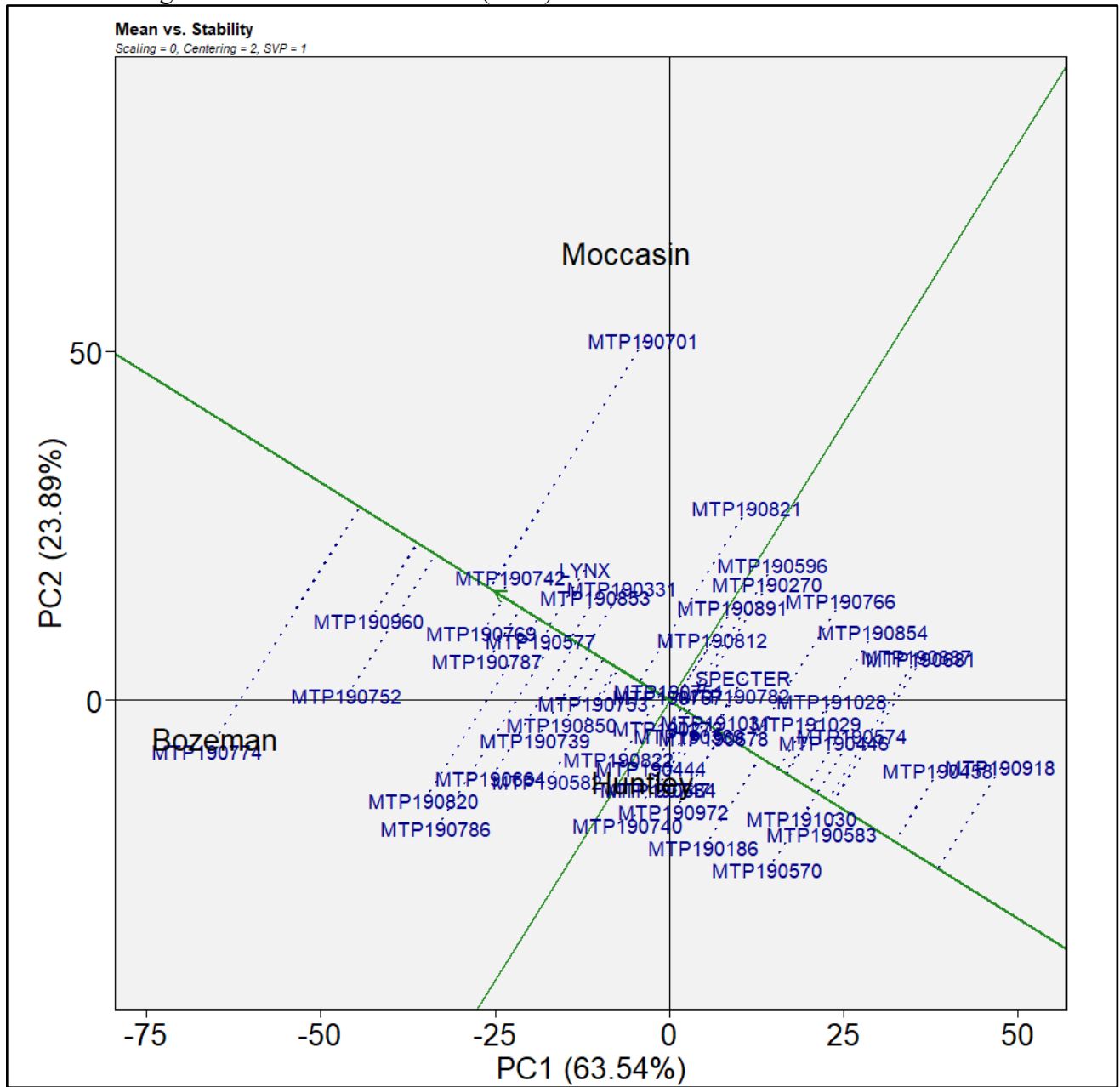






Figure 2.23. Mean performance and stability of genotypes based on winter survival visualized with the average environment coordination (AEC) view.



### Which-Won-Where Pattern

This shows the which-won-where pattern of genotype by environment dataset (Figure 2.24). It helps to address important concepts such as crossover G x E, mega-environment differentiation, specific adaptations, etc.

The polygon was drawn on genotypes that were furthest from the biplot origin, so all other genotypes were contained within the polygon. Genotypes located on the vertices of the polygon (MTP190753, MTP191029, MTP190272, MTP190270, MTP190389, MTP190186, MTP190960, MTP190960, MTP190786, and MTP190766) performed either the best or worst in one or more environments for yield. The perpendicular dotted lines were equality lines between adjacent genotypes which facilitate the visual comparison between them. For example, the equality line between MTP191029 and MTP190753 indicates that MTP191029 was better in Havre and MTP190753 was better in Bozeman. We saw that MTP190766 and MTP190786 were located on the line that connected MTP190753 and MTP190960. This meant that the rank MTP190753>MTP190766>MTP190786>MTP190960 was true in all environments for yield.

Similarly, from the Figure 2.25, we saw MTP190739, MTP19060, MTP190270, MTP190766, MTP190854, MTP190458, and MTP190742 performed either the best or the worst in one or more environments for 1000 seed weight. MTP190742 was better in Bozeman and Moccasin whereas MTP190270 performed better in Huntley for 1000 seed weight. A few lines like MTP190444 and MTP190854 did not perform well in any of the locations.

From the Figure 2.26, we saw MTP190331, Specter, MTP190769, MTP190850, MTP190678, MTP190458, and MTP190854 performed either the best or worst in one or more locations for protein. MTP190331 was better in Bozeman whereas MTP190458 was better in



Figure 2.25. The which-won-where view of the GGE biplot showing which genotype performed best in which environments for 1000-seed weight.

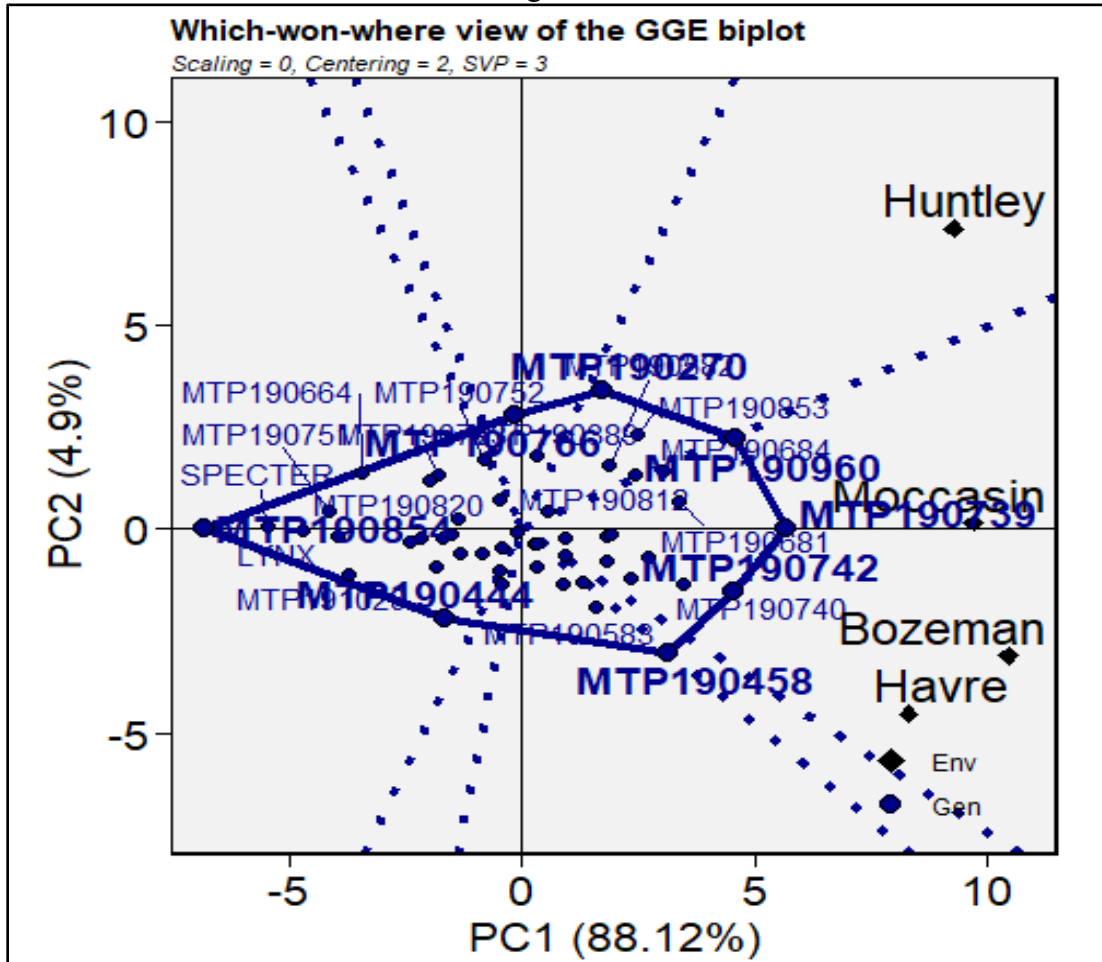
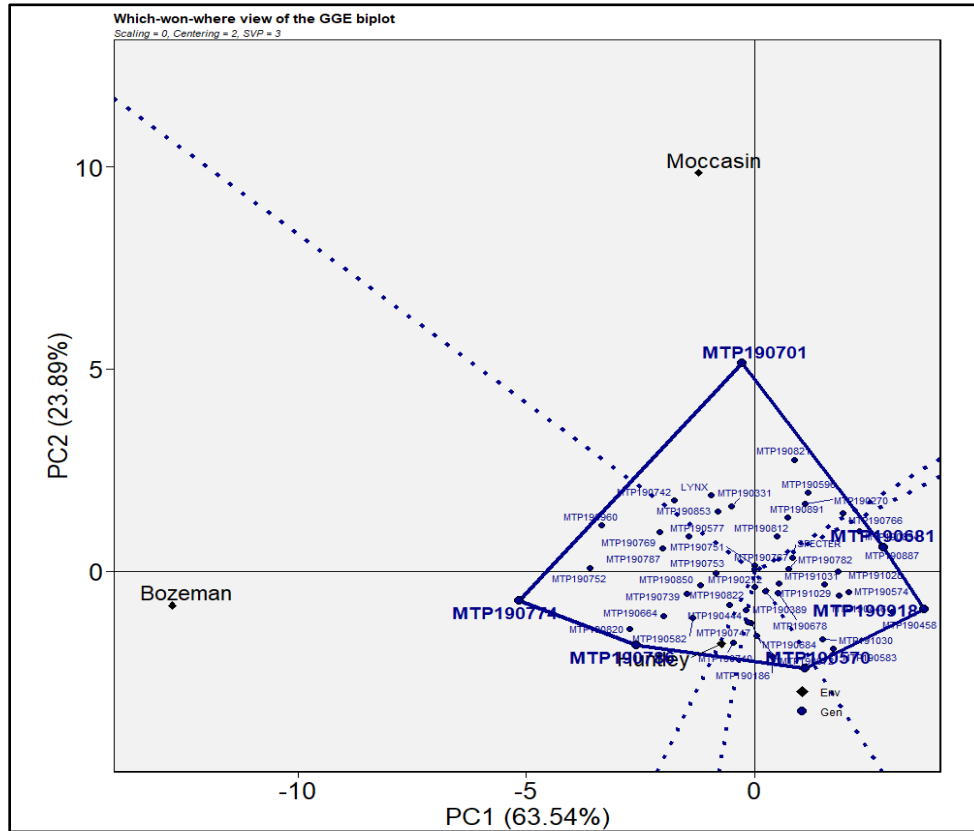




Figure 2.27. The which-won-where view of the GGE biplot showing which genotype performed best in which environments for winter survival.

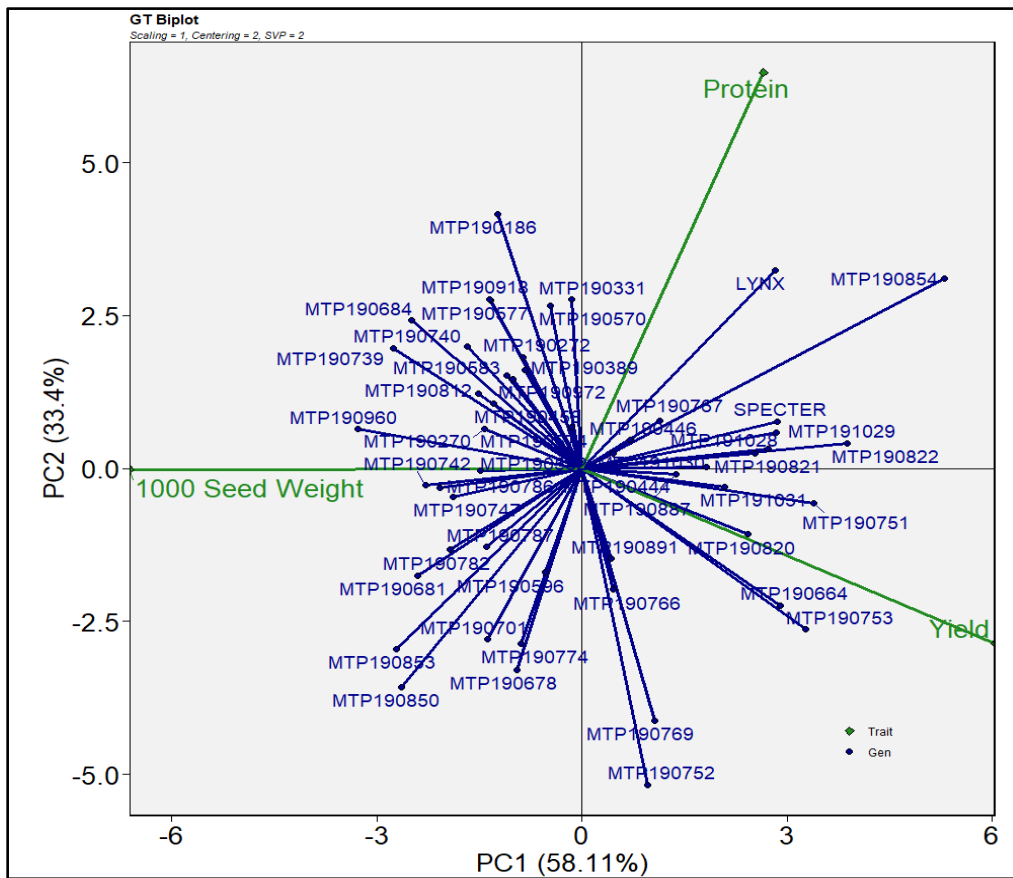


### Genotype-by-Trait Biplot

Biplots were used to scrutinize genotype-by-trait tables, with data from a single environment, an average of multiple environments, or a selective average of specific environments. The genotype-by-trait biplot serves as an important method for the biplot analysis of multivariate data. Such an approach is instrumental in elucidating the interrelationships among various traits. These relationships can be either positive or negative associations, or in some cases, traits might be identified that redundantly measure the same characteristic. Importantly, this method aids in identifying traits that could be effectively employed for indirect selection of another trait.

Further, the genotype-by-trait biplot facilitates a comprehensive visualization of the trait profiles of different genotypes. This visualization is critical as it allows for a detailed examination of the strengths and weaknesses inherent in each genotype. Understanding these aspects is crucial for making informed decisions in both parent selection and variety selection within the scope of genetic research and plant breeding (W. Yan & Kang, 2002).

Figure 2.28. A genotype-by-trait biplot representing genotypes measured for three traits: seed yield, 1000-seed weight, and protein. Data from 2021 to 2023 breeding lines from four test locations.



The biplot in Figure 2.28 presents data of winter pea breeding lines from four test environments (Bozeman, Havre, Huntley, and Moccasin) from three test years (2021, 2022, and 2023) for three traits (seed yield, 1000-seed weight, and protein). With the knowledge that higher

yield, better protein content, and larger seed size is desirable, the genotype by trait (G x T) visualization helps direct the best crosses that should achieve the desired variety.

This biplot helps to visualize which genotypes stand out among others for a specific trait. Across the tested genotypes, seed yield was found to be independent of protein content (near right angle) and negatively associated with 1000-seed weight (obtuse angle). The protein content was also found to be negatively associated with the 1000-seed weight.

The traits measured were aligned towards three different sides of the biplot and the genotypes were scattered in all directions. Thus, it was not possible to find a genotype that performed best for all the traits. This biplot helped visualize the genotypes that can be selected for one or two traits. MTP190751 was a high-yielding genotype with intermediate protein and lower-than-average seed size. MTP190751 was positioned directly across from MTP190742 in relation to the origin of the biplot, due to its contrasting trait profile compared to MTP190751. MTP190742 exhibited a greater 1000-seed weight, protein content lower than average, and an extremely low yield. As such, it was considered highly unsuitable for progression in its current state. However, it might be a good parent for studying the genetic determination of seed size in peas.

MTP190854 had high protein content, intermediate yield, and lower-than-average seed size. If it is desirable to improve further the protein of MTP190751, the cross of MTP190854 and MTP190751 may be useful. Similarly, the yield improvement of the high protein content line MTP190446 might be possible by crossing it with high-yielding MTP190753.

Many relationships can be derived from this biplot. Genotypes like MTP190820, MTP190753, and MTP190664 constitute a group of genotypes with similar trait profiles;

MTP190272, MTP190389, and MTP190577 form another group with similar trait profiles, etc. It would be rational to presume that the genotypes within each group share similar origins/parentages.

#### Seed Yield of Breeding Lines for Three Years and Four Locations

Breeding line with the greatest seed yield was MTP190753 (3662 kg/ha) followed by MTP190822 (3296 kg/ha) and MTP190664 (3285 kg/ha) (Table B4). The average yield of breeding lines was 2004 kg/ha. There was considerable variation in yield among the lines with a coefficient of variation (CV) of 35%. The 2023 experiments had the highest average yield (2576 kg/ha) followed by 2022 (1819 kg/ha) and 2021 (1548 kg/ha). Bozeman (2486 kg/ha) had the best average yield followed by Havre (2360 kg/ha), Huntley (1804 kg/ha), and Moccasin (1635 kg/ha) (Table B7).

#### Seed Size of Breeding Lines for Three Years and Four Locations

MTP190739 (226 g), MTP190960 (221 g), and MTP190681 (213 g) are the lines with the largest-sized seeds (Table B4). However, these genotypes had lower than average seed yield. The average 1000-seed weight for all the breeding lines was 178 g.

Moccasin (181 g) had the highest 1000-seed weight followed by Bozeman (179 g) and Havre (173 g). Lines from the year 2023 (182 g) had the highest 1000-seed weight followed by the year 2022 (181g) and 2021 (161 g).

#### Protein Content of Breeding Lines for Three Years and Four Locations

MTP190854 (25.8%), MTP190186 (25.4), and MTP190331 (25.2) had the highest

protein content. The average protein content of all the breeding lines was 24.4% with a CV of 2% (Table B4). Moccasin (25.0%) had the highest protein content followed by Havre (24.9%) and Huntley (24.6%). Bozeman (23.6%) had the lowest protein. Lines from 2021 (24.6%) had the highest protein content followed by 2022 (24.5%) and 2023 (24.3%) (Table B1).

#### Seed Yield Ranking of Breeding Lines by Year

MTP190769 was the highest-yielding line in 2021 and 2022 and the highest-ranked line over all three years. MTP190751, MTP191029 and MTP190753 were the other high-yielding lines that performed well in all the years. MTP190960, MTP190186 and MTP190918 were consistently poor yielding in all three years (Table B5).

#### Seed Yield Ranking of Breeding Lines by Location

MTP190753, MTP190820, and MTP190822 were the best-ranked lines for overall locations. MTP190972 and MTP190918 did not perform well in any location and were consistently at the bottom of the yield ranking (Table B6).

Correlation Matrix Analysis

Figure 2.29. Pearson correlation between a pair of traits.

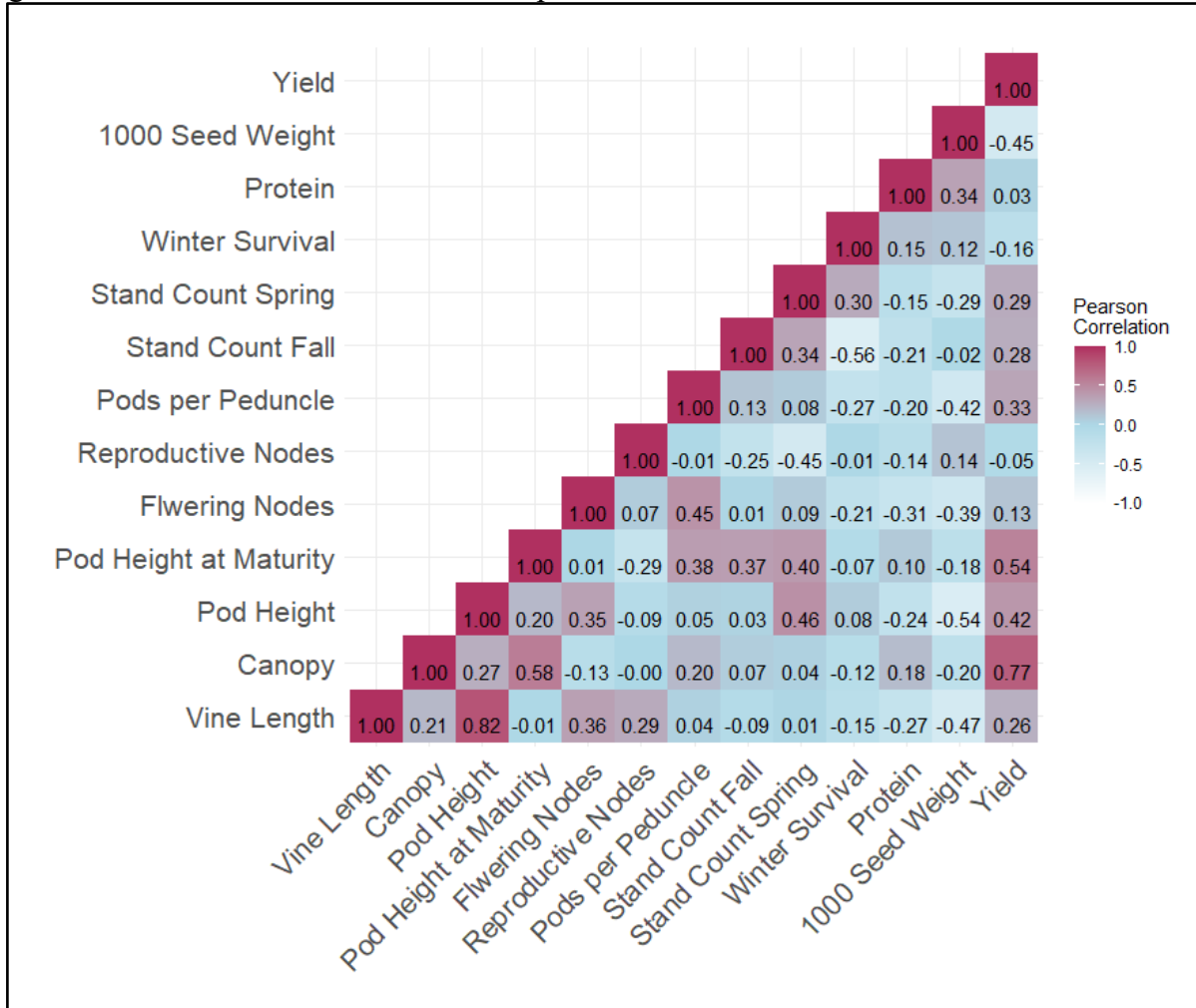


Figure 2.29 displays the Pearson correlation matrix for selected traits of pea breeding lines of three years of data from Bozeman. The traits analyzed were seed yield, one thousand seed weight, protein content, winter survival, fall stand count, spring stand count, pods per peduncle, number of reproductive nodes, number of flowering nodes, pod height, pod height at maturity stage, canopy height, and vine length.

Seed yield was negatively correlated with 1000-seed weight and had a very low positive

correlation with protein. Some notable traits positively correlated with seed yield were canopy and pod height, whereas vine length had a low positive correlation with the yield. This showed that an upright-standing plant had a better yield than a lodged plant which could have implications for photosynthetic efficiency and biomass accumulation. Conversely, any negative correlations observed, such as between a mature pod height and winter survival, might indicate a trade-off between these traits, potentially guiding breeders to prioritize shorter pod height plants for enhanced winter hardiness. The plant stand count in both fall and spring was also positively correlated with yield. This meant the number of plants that emerged in the fall and the number of plants that survived coming into the spring both had a positive impact on the yield. Winter survival showed a very weak positive correlation with spring stand count and a negative correlation with fall stand count. Winter survival was not considered the best measure to distinguish between the winter hardy genotypes from the non-hardy ones. There was not a good germination percentage in the fall, while plants overwintered, went into dormancy, and only emerged in the spring. Thus, in some instances, there were a greater number of plants stand in the spring as compared to what germinated in the fall. 1000-seed weight was positively correlated with protein content and negatively correlated with vine length and canopy height. Strong positive correlations, such as yield with matured pod height and canopy, pod height and vine length, and canopy and matured pod height imply a more robust and consistent relationship across multiple environments and are particularly noteworthy for their potential in predictive modeling and trait selection strategies.

However, it is important to remember that correlation does not equate to causation. While two traits may exhibit a strong correlation, this does not imply that one trait's expression causes

the other. There could be underlying genetic factors or environmental interactions affecting both traits. Hence, the correlations observed are interpreted as indicators of association, not drivers of trait expression. The elucidation of these correlations is instrumental for the advancement of breeding programs aimed at improving winter pea survival. For example, a significant positive correlation between the canopy and seed yield underscores the importance of selecting for robust upright standing pea lines. This relationship suggests that an upright-standing plant could serve as an early and reliable selection criterion for breeding winter-hardy pea varieties. Considering these findings, future research may focus on explaining the causal mechanisms behind these correlations, thereby enhancing our understanding of the genetic and physiological basis of winter hardiness in peas. The insights derived from this correlation matrix are expected to significantly contribute to the tailored selection of phenotypic traits, optimizing winter pea breeding strategies for resilience and productivity in the face of climatic challenges.

### Discussion

The breeding lines used in this experiment showed a wide range of agronomic performance. This multi-environment trial effectively distinguished the better lines from the rest. This helped clarify the genotype-by-environment interaction, aiding in a deeper understanding of the inherent distinctions.

The seed yield of the breeding lines saw a significant difference for each year and location. Yearly weather patterns of temperature, precipitation, and humidity would have accounted for this. Similarly, the differences in weather for each location might have caused the variation in seed yield for each location. Agronomic practices also accounted for better seed yield at Bozeman, with the highest average seed yield. The closeness of the trial site also

determined the scope of the standard agronomic practices feasible to carry out. The proximity of the trial location also accounted for the extent of the standard agronomic practices that can be performed. Weed management was one of the major issues when the trial locations were located farther away. Peas compete very poorly with weeds at the early stage and account for up to 40% of yield reduction (Ullah et al., 2008). This was evident when we observed a nearly unmanageable amount of weed infestation in Huntley 2023, despite having a good plant stand. This resulted in low seed yield from that location. Similarly, yield measured in 2022 Huntley indicated the lowest among all the tested environments. This significant decrease in yield can be largely attributed to an unforeseen hail event that occurred at the peak of seed fill and harvest. A hailstorm caused extensive damage to the seeds, critically impacting their development and resulting in a marked reduction in yield. Also, none of the breeding lines survived in Havre 2021 due to winter kill.

We observed an interesting phenomenon known as seed dormancy or dormant seeding in five separate instances across various locations over a span of three years. This condition occurs when seeds that are sown in the fall but fail to germinate immediately and remain dormant within the soil. These dormant seeds emerge the following spring, once the snow cover has completely melted. This process allows the crops that are planted in the fall to act as spring crops, adapting to seasonal changes. Dormancy has been attributed to many reasons with one of the most important being low soil moisture level. This was particularly evident when there was very low to no rainfall in the fall. Weather patterns may have also played a significant role in seed yield. The year 2021 was one of the driest in recent years. This might have caused the lower-than-average yield from all the locations in that particular year. The study showed higher yield

variation due to year than the genotypes and location. Also, location contributed the lowest to the total variation in yield.

Seed size and protein content of the breeding lines were measured and showed significant differences. MTP190753, MTP190822, and MTP190820 were the best breeding lines for yield and stability across years and locations. Also, most variation was due to year than other sources of variation. MTP190739, MTP190960, and MTP190742 were the ones with the highest 1000-seed weight. Protein content was highest in Havre, which was also the location where the 1000-seed weight was lower than average. There might be trade-offs between seed size and protein content. It is however found that most of the variation in protein percentage is due to within-genotype differences, and the trade-offs between the seed size and protein are not evident (Arthur et al., 1991).

The genotype by environment (G x E) interaction was visualized in a biplot. Our GGE biplot captured 86%, 93%, 83% and 87% of the total variation in seed yield, 1000-seed weight, protein content, and winter survival, respectively, in two principal components PC1 and PC2. The which-won-where view of the biplot was divided into sectors, meaning different locations had different winners. This suggests the target environment contains different mega-environments and different varieties should be selected for each mega-environment. This also highlighted the need of a specific breeding program targeted to develop varieties which suits that mega-environment.

Tillage practices and previous crops varied across the environments which might have an effect on the yield. Precipitation was also significantly lower than average in 2021 than in 2022 and 2023. These differences could have influenced the moisture and nutrient availability in the

soil, which in turn affected seed germination, yield, and quality. Most of our trials were planted on standing stubble, which is considered better at trapping soil moisture and making it available to the seeds for faster germination (Triplett Jr. & Dick, 2008). However, in above-average wet and cool growing conditions, tillage prior to planting might result in faster germination and better yield of field peas (S. Stepanovic, 2018).

The performance of winter pea breeding lines was compared to the spring pea variety trials in the same environment. The winter pea lines in 2023 had higher average yield than spring peas. However, in the years 2021 and 2022, spring pea had better yield performance. Protein content of winter lines was higher than spring lines in all the years.

The genotype-by-trait biplot helped to identify which genotypes performed best for specific traits. However, the absence of a single genotype that excels in all three traits—yield, protein, and thousand seed weight—highlights the inherent trade-offs present in plant breeding. This may be due to genetic limitations or environmental interactions that prevented a simultaneous optimization of yield, protein content, and seed size. The absence of a single genotype that excelled in all three traits is typical in plant breeding. Each genotype exhibits a unique combination of trait values, representing different breeding priorities. These trade-offs necessitate careful consideration by breeders when making selection decisions. For instance, a genotype having a higher yield may be preferred in breeding programs prioritizing yield over protein content or seed size. For example, in breeding programs that prioritize yield over protein content or seed size, a genotype with a higher yield may be more favorable.

Breeding lines were ranked based on yield and stability for three traits. After considering the multivariate statistical results from the GGE biplot analysis, these were categorized into three

major groups:

1. Highly stable and high yield potential

The breeding lines falling under this group had high seed yield and stable production over the years and locations. This group of genotypes was suited to a range of environments without compromising the yield. MTP190820, MTP190822, and MTP190664 fall under this category.

2. Low stability and high yield potential

The breeding lines falling under this group had high seed yield but not stable production over the years and locations. This group of genotypes may be appropriate for a specific environment. MTP190753, MTP191029, and MTP190752 fall under this category.

3. Low yield and high stability

The breeding lines falling under this group had lower than average seed yield but stable production over the years and locations. This group of genotypes may be ideal in a breeding program intended to improve certain phenotypes. MTP190854, MTP190767, and MTP190850 fall under this category.

Breeders must decide which traits are most important for their goals and select genotypes accordingly. This decision is often based on the needs of the market, environmental adaptability, and its intended use. The knowledge gained from this research can help in targeted breeding or adopting a genomic selection approach. It highlights that careful consideration is required keeping in mind the genetic potential of genotypes, the impact of environmental factors, and the specific objectives of the breeding program.

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## SUMMARY

Winter peas can be grown in the wheat growing regions of Montana and Northern Great Plains (NGP) because of their agronomic values in offering benefits like nitrogen fixation and providing a break in disease and pest cycles when compared to continuous cropping. The development of this cropping practice has not seen large adoption due to the unavailability of suitable winter pea lines which can sustain harsh winter of these regions. Development of a true winter hardy pea line requires a rigorous selection process accompanied by consistent winter conditions. This can be possible by testing pea lines across multiple environments which differ in climatic conditions.

This study delves into the subject of multi-environment evaluation of winter peas through analysis of the agronomic and breeding potential through rigorous multi-environment trials (METs). METs are a cornerstone of plant breeding research, providing a robust framework for evaluating the performance of plant genotypes across a range of environmental conditions. This methodology is particularly pertinent for the Northern Great Plains, a region characterized by its challenging climate and unique soil profiles. By implementing METs, the thesis presents a nuanced understanding of how winter peas can thrive in such conditions, focusing on key traits like yield, seed size, protein content and winter survival.

The trials conducted have revealed significant variability in the winter hardiness, which is a determinant factor for successful crop survival and productivity in cold climates. High yielding and stable breeding lines such as MTP190820 and MTP190822 provide a great opportunity to be developed as winter pea varieties. The yield of winter pea lines was highly dependent upon the moisture availability. High yield was obtained in 2023 as a result of good rainfall. A few lines

consistently ranked higher in yield and stability, regardless of the weather conditions. These were the promising lines for future crop improvement. A GGE biplot visualization successfully differentiated the best and worst genotypes. Complex genotype by environment (G x E) interactions were detected which play a decisive role in the manifestation of phenotypic characteristics. Understanding these interactions is essential for optimizing plant breeding strategies and ensuring that the most resilient and productive genotypes are advanced.

Further, the study extends beyond mere yield, examining other critical attributes in which producers and industries are more interested, such as protein content and seed size. In general, both the protein content and seed size were negatively associated with seed yield. This indicates that it is hard to find a genotype which performed well in all the traits. This might be due to genetic limitations and environmental interactions that prevented a simultaneous optimization for yield, seed size and protein content. This indicates a trade-off situation and genotypes which exhibit a unique combination of trait values should represent a different breeding priority. Moreover, mega-environment differentiation helped to select specific genotypes based on the trait of interest for a particular environment. Some environments posed unique characteristics (e.g., Havre for protein) that made them suitable to target genotypes for a specific trait. This signifies the selection of different varieties for each mega-environment. The identification and classification of larger regions with analogous climate and soil conditions, breeders can streamline their efforts to develop genotypes that are finely tuned to specific trait and environment.

This study enables a targeted approach to breeding winter peas for Montana. By prioritizing traits that are most valued by growers and the marketplace, breeding programs can be

more efficient and effective. It holds the promise of transforming the agricultural practice of the wheat-growing regions under study, fostering sustainability, and enhancing the economic prospects of farmers in these areas. This calls to action for ongoing research and collaboration among plant breeders, agronomists, and producers. Emphasizing the need for continued evaluation and refinement of winter pea genotypes, this study advocates for a dynamic and responsive breeding program that can adapt to the evolving challenges and opportunities presented by the changing agricultural environment.

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APPENDICES

APPENDIX A

TABLES OF GERMPLASM LINES INCLUDED IN THE  
EXPERIMENT

Table A1. List of germplasm lines included in the experiment in 2021.

SN	Name	Yield (kg/ha)	TSW (g)	Protein (%)
1	Aa135	895	102	18.8
2	COL. NO. 317	439	99	8.0
3	DRKA	335	239	
4	FC 40823	176	171	
5	FENN	1621	112	25.7
6	G 10946	513	154	19.2
7	G 16701	391	339	
8	GLACIER	910	130	
9	GRANGER	1757	134	
10	JUNIOR	789	79	11.4
11	KALOFER	1808	123	16.4
12	KENEJA	1167	147	25.6
13	L 805/5g	596	161	19.2
14	LEVAKHANE	508	253	16.5
15	LINE 179	804	182	
16	LINE 340/11	1128	185	
17	LYNX	1158	136	22.1
18	MARKOVO 1	182	199	
19	MELROSE	1426	117	25.8
20	MIR	1242	153	25.8
21	NOKALSKESNI	557	264	8.1
22	ONTOFO	753	225	18.3
23	OVATION	767	113	11.4
24	P-226	1483	138	25.3
25	PI 343991	516	144	16.5
26	PI 343993	380	124	16.6
27	PI 343998	605	99	
28	PI 377693	1534	120	22.1
29	PI 517922	1291	118	24.8
30	PI 517923	1508	141	24.3
31	PI 517924	1125	144	25.5
32	PI 517925	1282	141	25.9
33	PI 517926	1137	99	22.0
34	PI 560968	782	129	
35	PI 574505	1818	114	16.6
36	PI 577142	295	97	8.3
37	PI 639979 PSP	1128	163	25.7
38	PI 639981	1665	141	16.3

39	PS03101269	1202	154	21.8
40	RENN	1520	110	25.9
41	ROMACK	1298	126	22.4
42	SPECTER	1359	130	24.2
43	VARDIM 1	1099	181	18.5
44	W6 12713	1806	117	16.7
45	W6 12723	1282	156	22.0
46	WINDHAM	1454	135	14.9
47	WINTER 1 MM	1054	135	17.9
48	WINTER	1092	142	26.3

Table A2. List of germplasm lines included in the experiment in 2022.

SN	Name	Yield (kg/ha)	TSW (g)	Protein (%)
1	Aa135	1202	95	25.6
2	FENN	1823	108	26.4
3	G 10946	1223	117	17.5
4	GLACIER	1664	122	15.9
5	GRANGER	1834	126	25.8
6	JUNIOR	550	122	19.8
7	KALOFER	1543	111	26.9
8	KENEJA	2317	110	21.2
9	L 805/5g	1103	147	23.5
10	LINE 179	510	123	7.9
11	LINE 340/11	1756	180	24.1
12	LYNX	1684	122	27.3
13	MELROSE	1725	111	26.4
14	MIR	1841	147	24.1
15	ONTOFO	903	218	26.2
16	OVATION	284	171	25.9
17	P-226	1841	127	26.2
18	PI 343998	1571	94	24.5
19	PI 377693	1658	106	26.8
20	PI 517922	1801	111	25.8
21	PI 517923	2115	117	21.4
22	PI 517924	1147	129	26.0
23	PI 517925	1387	133	26.3
24	PI 517926	1480	96	26.0
25	PI 560968	1661	114	26.3
26	PI 574505	1808	107	27.2
27	PI 639979 PSP	1688	160	25.8

28	PI 639981	1628	113	26.6
29	PS03101269	1504	137	26.4
30	RENN	1583	103	27.8
31	ROMACK	1699	106	22.7
32	SPECTER	1742	114	25.9
33	VARDIM 1	1180	225	21.5
34	W6 12713	1789	111	26.4
35	W6 12723	1395	120	27.3
36	WINDHAM	1727	133	25.0
37	WINTER 1 MM	1359	173	24.6
38	WINTER FUTTERERBSE	1586	126	26.6

Table A3. List of germplasm lines included in the experiment in 2023.

SN	Name	Yield (kg/ha)	TSW (g)	Protein (%)
1	Aa135	1955	104	26.3
2	FENN	3056	117	25.8
3	G 10946	2121	137	26.0
4	GLACIER	2403	129	24.8
5	GRANGER	3112	131	25.3
6	KALOFR	2903	120	25.2
7	KENEJA	2981	129	24.6
8	L 805/5g	2271	135	25.9
9	LINE 179	1045	169	24.1
10	LINE 340/11	2820	181	24.2
11	LYNX	3110	141	25.1
12	MELROSE	3396	119	25.0
13	MIR	2616	145	24.6
14	ONTOFO	1198	201	25.5
15	P-226	3209	140	25.2
16	PI 343998	1690	100	24.1
17	PI 377693	3200	127	26.0
18	PI 517922	3193	119	24.3
19	PI 517923	3529	150	24.7
20	PI 517924	2772	136	24.9
21	PI 517925	2089	127	25.0
22	PI 517926	2767	103	26.6
23	PI 560968	2611	127	24.6
24	PI 574505	3194	121	25.7
25	PI 639979 PSP	1842	175	25.8

26	PI 639981	2791	126	25.4
27	PS03101269	2463	154	24.4
28	RENN	3031	110	25.8
29	ROMACK	2868	127	26.0
30	SPECTER	3089	134	24.3
31	VARDIM 1	750	263	25.4
32	W6 12713	3224	123	25.4
33	W6 12723	2570	134	25.6
34	WINDHAM	3452	147	24.2
35	WINTER 1 MM	1712	176	23.3
36	WINTER FUTTERERBSE	2549	131	25.2

Table A4. Overall ranking of germplasm lines.

SN	Name	Yield (kg/ha)	TSW (g)	Protein (%)
1	W6 12713	2367	117	24.1
2	PI 574505	2365	114	24.5
3	GRANGER	2330	129	20.3
4	PI 517923	2317	136	23.5
5	KALOFER	2140	117	24.1
6	FENN	2125	112	25.9
7	MELROSE	2124	116	25.7
8	P-226	2124	135	25.6
9	WINDHAM	2101	138	20.7
10	PI 639981	2101	124	24.1
11	PI 377693	2085	118	24.7
12	SPECTER	2084	126	24.8
13	KENEJA	2079	130	24.0
14	LINE 340/11	2056	181	19.2
15	PI 517922	2033	116	24.9
16	RENN	2004	108	26.4
17	LYNX	1916	132	24.9
18	ROMACK	1913	125	24.9
19	PI 560968	1865	122	20.2
20	MIR	1849	148	24.9
21	GLACIER	1809	126	14.4
22	PI 517926	1744	99	24.6
23	W6 12723	1713	138	24.7
24	WINTER FUTTERERBSE	1692	133	26.0

25	PS03101269	1683	149	24.0
26	PI 517924	1638	137	24.1
27	PI 517925	1625	134	25.8
28	PI 639979 PSP	1520	165	25.7
29	PI 343998	1425	97	18.7
30	WINTER 1 MM	1336	158	21.4
31	L 805/5g	1267	149	22.6
32	Aa135	1262	100	21.1
33	G 10946	1226	143	21.9
34	VARDIM 1	1006	238	24.1
35	ONTOFO	926	216	22.8
36	LINE 179	881	171	11.0
37	JUNIOR	686	128	17.9
38	OVATION	580	179	20.5
	<b>Mean</b>	<b>1737</b>	<b>137</b>	<b>22.9</b>
	<b>SD</b>	<b>478.7</b>	<b>29.6</b>	<b>3.3</b>
	<b>SE</b>	<b>77.6</b>	<b>4.8</b>	<b>0.5</b>
	<b>CV (%)</b>	<b>28</b>	<b>22</b>	<b>14</b>
	<b>Min</b>	<b>580</b>	<b>97</b>	<b>11.0</b>
	<b>Max</b>	<b>2367</b>	<b>238</b>	<b>26.4</b>

Table A5. Ranking germplasm lines by Year.

SN	Name	2021	2022	2023	Sum of ranks	Overall rank
1	PI 517923	9	2	<i>1</i>	12	<b>1</b>
2	PI 574505	<i>1</i>	7	7	15	<b>2</b>
3	W6 12713	3	9	4	16	<b>3</b>
4	GRANGER	4	5	9	18	<b>4</b>
5	P-226	10	4	5	19	<b>5</b>
6	FENN	6	6	12	24	<b>6</b>
7	WINDHAM	11	12	2	25	<b>7</b>
8	MELROSE	12	13	3	28	<b>8</b>
9	PI 517922	15	8	8	31	<b>9</b>
10	PI 377693	7	19	6	32	<b>10</b>
11	SPECTER	13	11	11	35	<b>11</b>
12	KENEJA	20	<i>1</i>	14	35	<b>12</b>
13	KALOFER	2	24	15	41	<b>13</b>
14	MIR	18	3	21	42	<b>14</b>
15	PI 639981	5	20	18	43	<b>15</b>
16	RENN	8	22	13	43	<b>16</b>

17	ROMACK	14	14	16	44	<b>17</b>
18	LYNX	21	16	10	47	<b>18</b>
19	LINE 340/11	23	10	17	50	<b>19</b>
20	W6 12723	17	27	23	67	<b>20</b>
21	PI 517926	22	26	20	68	<b>21</b>
22	PS03101269	19	25	25	69	<b>22</b>
23	PI 639979 PSP	24	15	30	69	<b>23</b>
24	WINTER FUTTERERBSE	27	21	24	72	<b>24</b>
25	GLACIER	29	17	26	72	<b>25</b>
26	PI 517925	16	28	29	73	<b>26</b>
27	PI 560968	33	18	22	73	<b>27</b>
28	PI 517924	25	33	19	77	<b>28</b>
29	WINTER 1 MM	28	29	31	88	<b>29</b>
30	PI 343998	36	23	32	91	<b>30</b>
31	VARDIM 1	26	32	36	94	<b>31</b>
32	Aa135	30	31	33	94	<b>32</b>
33	G 10946	38	30	28	96	<b>33</b>
34	L 805/5g	37	34	27	98	<b>34</b>
35	LINE 179	31	37	35	103	<b>35</b>
36	ONTOFO	35	35	34	104	<b>36</b>
37	JUNIOR	32	36	37	105	<b>37</b>
38	OVATION	34	38	38	110	<b>38</b>

Table A6. Ranking of germplasm lines by location.

SN	Name	Bozeman	Havre	Huntley	Moccasin	Sum of ranks	Overall rank
1	GRANGER	18	2	5	<i>1</i>	26	<b>1</b>
2	W6 12713	6	3	7	11	27	<b>2</b>
3	PI 517923	2	8	12	6	28	<b>3</b>
4	SPECTER	3	20	2	14	39	<b>4</b>
5	FENN	7	16	10	7	40	<b>5</b>
6	P-226	8	11	11	13	43	<b>6</b>
7	PI 574505	9	<i>1</i>	31	2	43	<b>7</b>
8	KALOFER	20	5	13	8	46	<b>8</b>
9	PI 377693	11	18	9	9	47	<b>9</b>
10	MELROSE	12	19	<i>1</i>	17	49	<b>10</b>
11	KENEJA	5	9	15	21	50	<b>11</b>
12	WINDHAM	4	25	6	16	51	<b>12</b>
13	PI 517922	16	10	16	10	52	<b>13</b>
14	RENN	14	12	14	20	60	<b>14</b>

15	PI 639981	17	7	35	5	64	<b>15</b>
16	LINE 340/11	13	13	36	3	65	<b>16</b>
17	ROMACK	10	22	21	15	68	<b>17</b>
18	LYNX	<i>1</i>	27	18	23	69	<b>18</b>
19	GLACIER	28	4	37	4	73	<b>19</b>
20	PI 560968	26	6	32	12	76	<b>20</b>
21	MIR	23	17	20	18	78	<b>21</b>
22	PI 517926	24	26	3	26	79	<b>22</b>
23	WINTER FUTTERERBSE	21	21	17	28	87	<b>23</b>
24	W6 12723	22	28	26	19	95	<b>24</b>
25	PS03101269	25	24	23	24	96	<b>25</b>
26	L 805/5g	35	29	4	30	98	<b>26</b>
27	PI 517924	19	32	27	22	100	<b>27</b>
28	PI 639979 PSP	29	14	29	29	101	<b>28</b>
29	PI 343998	30	15	34	25	104	<b>29</b>
30	PI 517925	15	33	30	27	105	<b>30</b>
31	Aa135	34	23	25	36	118	<b>31</b>
32	WINTER 1 MM	27	31	28	34	120	<b>32</b>
33	OVATION	38	37	8	38	121	<b>33</b>
34	ONTOFO	36	34	19	35	124	<b>34</b>
35	G 10946	32	30	33	31	126	<b>35</b>
36	VARDIM 1	31	35	24	37	127	<b>36</b>
37	JUNIOR	37	38	22	33	130	<b>37</b>
38	LINE 179	33	36	38	32	139	<b>38</b>

Table A7. Yield (kg/ha) of germplasm lines across locations averaged for three years.

Rank	Name	Bozeman	Havre	Huntley	Moccasin	Average
1	GRANGER	2281	3172	1236	2679	2342
2	PI 574505	2618	3197	730	2659	2301
3	W6 12713	2686	3148	1184	2129	2287
4	PI 517923	3075	2628	1046	2265	2254
5	KALOFER	2273	2907	1014	2234	2107
6	SPECTER	2908	2096	1327	2095	2107
7	MELROSE	2512	2185	1689	1982	2092
8	P-226	2621	2511	1079	2120	2083
9	FENN	2635	2322	1120	2252	2082
10	PI 377693	2541	2271	1156	2219	2047
11	PI 639981	2324	2842	589	2425	2045

12	KENEJA	2742	2553	1011	1788	2024
13	PI 517922	2340	2521	990	2180	2008
14	WINDHAM	2852	1932	1218	2004	2002
15	LYNX	3358	1854	944	1770	1982
16	RENN	2496	2485	1012	1859	1963
17	LINE 340/11	2500	2471	248	2562	1945
18	ROMACK	2550	2055	886	2054	1886
19	PI 560968	1856	2865	623	2126	1868
20	GLACIER	1650	3084	177	2482	1848
21	MIR	2121	2298	907	1980	1827
22	PI 517926	2067	1909	1301	1592	1717
23	W6 12723	2127	1853	857	1878	1679
24	PS03101269	2052	1954	865	1737	1652
	WINTER					
25	FUTTERERBSE	2177	2061	953	1416	1652
26	PI 517924	2277	1468	812	1784	1585
27	PI 517925	2467	1261	759	1754	1560
28	PI 639979 PSP	1498	2427	775	1387	1522
29	PI 343998	1274	2365	615	1598	1463
30	L 805/5g	895	1782	1252	1265	1299
31	WINTER 1 MM	1737	1592	781	1007	1279
32	Aa135	1143	2021	859	1002	1256
33	G 10946	1256	1660	616	1261	1198
34	VARDIM 1	1273	1073	862	715	981
35	ONTOFO	851	1104	937	839	933
36	LINE 179	1210	127	126	1250	678
37	JUNIOR	341	112	874	1017	586
38	OVATION	191	115	1166	578	513
	<b>Mean</b>	<b>2047</b>	<b>2060</b>	<b>910</b>	<b>1788</b>	
	<b>CV (%)</b>	<b>36.6</b>	<b>38.5</b>	<b>34.5</b>	<b>30.8</b>	
	<b>SD</b>	<b>749</b>	<b>793</b>	<b>314</b>	<b>551</b>	

Table A8. Data summary of germplasm lines Bozeman 2023.

S	Name	vinelen	cano	pod	podht	flwrn	reprn	podp	standc	standcn	survi	thseed	prote	yld
N		gth	py	ht	mat	ode	ode	ed	ntfl	tsp	val	wt	in	
1	LYNX	78	66	14	11	11	12	2	11	7	61	148	24.7	4785
2	PI 517923	78	42	37	8	12	14	2	16	10	63	151	24.7	4761
3	P-226	144	54	60	14	9	12	2	9	6	59	150	25.0	4306
4	LINE	76	39	29	6	10	11	2	9	4	39	196	24.3	4210
5	W6 12713	120	70	26	17	13	11	2	10	7	73	130	25.3	4210
6	WINDHAM	73	48	27	3	10	11	2	10	4	39	149	23.9	4139
7	ROMACK	147	34	47	3	14	10	2	14	9	63	134	25.6	4139
8	KENEJA	170	55	33	14	9	15	2	8	5	66	137	23.8	4115
9	MELROSE	143	88	49	18	10	10	2	15	8	50	130	24.8	4115
10	PI 377693	173	45	54	11	12	14	2	17	8	48	124	25.7	4019
11	PI 574505	133	46	48	9	12	13	2	11	7	62	128	25.3	3995
12	WINTER	137	69	22	9	13	12	2	9	6	66	138	23.8	3923
13	FENN	146	50	43	11	8	14	2	12	8	69	121	25.2	3875
14	RENN	118	43	26	6	12	8	2	13	8	63	120	25.2	3875
15	PI 517925	88	29	32	3	9	10	2	20	6	29	135	24.9	3732
16	PI 517924	57	52	12	8	8	11	2	11	9	82	136	25.0	3708
17	PI 517922	62	51	16	5	11	13	2	11	3	30	115	23.6	3684
18	PI 639981	165	56	53	6	9	13	2	10	6	58	131	25.2	3636
19	MIR	124	37	49	5	10	11	2	9	4	46	146	24.7	3421
20	SPECTER	126	64	48	11	13	10	2	16	11	66	132	23.7	3421
21	PI 517926	123	40	47	8	16	10	2	11	4	35	100	25.8	3301
22	KALOFER	145	67	26	12	15	11	2	7	5	81	133	25.0	3206
23	PI 560968	123	53	40	15	10	11	2	11	6	51	133	25.0	3206
24	G 10946	157	23	64	8	14	10	2	11	3	24	140	25.4	3110

25	GRANGER	127	62	46	10	13	9	2	14	7	50	133	24.1	2751
26	W6 12723	112	36	16	7	14	10	2	11	3	24	144	24.8	2751
27	PS0310126	158	42	45	1	12	10	2	14	7	47	153	24.4	2177
28	GLACIER	47	30	12	9	11	7	2	8	3	43	131	24.4	1962
29	PI 639979	131	29	27	7	10	13	2	7	2	32	172	25.2	1842
30	Aa135	91	65	18	4	7	7	2	6	2	25	102	25.3	1818
31	LINE 179	93	30	23	2	6	13	2	7	4	55	184	24.1	1699
32	L 805/5g	127	49	33	10	9	14	2	11	4	33	142	24.9	1675
33	PI 343998	90	42	37	4	11	9	1	11	2	14	86	22.4	1603
34	WINTER 1	145	28	22	5	11	12	1	5	2	35	189	24.2	1388
35	ONTOFO	90	39	32	4	11	11	1	16	2	13	214	24.7	1053
36	VARDIM 1	52	19	6	1	7	7	1	12	8	65	289	25.1	837
<b>mean</b>		<b>115.6</b>	<b>46.7</b>	<b>33.5</b>	<b>7.7</b>	<b>10.6</b>	<b>10.9</b>	<b>1.7</b>	<b>11.1</b>	<b>5.4</b>	<b>49.0</b>	<b>144.2</b>	<b>24.7</b>	<b>3179.0</b>
<b>std</b>		<b>35.2</b>	<b>15.3</b>	<b>14.8</b>	<b>4.4</b>	<b>2.3</b>	<b>2.0</b>	<b>0.3</b>	<b>3.3</b>	<b>2.5</b>	<b>21.1</b>	<b>35.7</b>	<b>0.7</b>	<b>1105.8</b>
<b>min</b>		<b>47.0</b>	<b>19.0</b>	<b>6.0</b>	<b>1.0</b>	<b>6.0</b>	<b>6.5</b>	<b>1.0</b>	<b>5.0</b>	<b>1.5</b>	<b>13</b>	<b>86.3</b>	<b>22.4</b>	<b>837.3</b>
<b>max</b>		<b>173</b>	<b>87.5</b>	<b>64</b>	<b>18</b>	<b>16</b>	<b>15</b>	<b>2</b>	<b>20</b>	<b>11</b>	<b>82</b>	<b>289</b>	<b>25.8</b>	<b>4785</b>
<b>cv (%)</b>		<b>30</b>	<b>33</b>	<b>44</b>	<b>58</b>	<b>22</b>	<b>18</b>	<b>20</b>	<b>30</b>	<b>47</b>	<b>39</b>	<b>25</b>	<b>3</b>	<b>35</b>
<b>error_varia</b>		<b>1240.4</b>	<b>234.</b>	<b>220.</b>	<b>19.4</b>	<b>5.5</b>	<b>3.9</b>	<b>0.1</b>	<b>11.2</b>	<b>6.5</b>	<b>443.8</b>	<b>1272.3</b>	<b>0.5</b>	<b>122282</b>
<b>n</b>			<b>7</b>	<b>2</b>										<b>9.3</b>

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Table A9. Data summary of germplasm lines Havre 2023.

SN	name	vinelength	canopy	standcntsp	thseedwt	protein	yld
1	PI 517923	48	38	22	142	24.8	4799
2	GRANGER	81	76	39	119	27.7	4620
3	W6 12713	82	64	27	115	26.7	4547
4	PI 517922	38	32	18	122	26.3	4334
5	MELROSE	77	61	26	119	25.9	4301
6	KENEJA	77	65	29	124	26.9	4285
7	PI 574505	73	56	37	110	27.4	4276

8	RENN	77	57	32	108	27.9	4270
9	P-226	85	67	38	133	26.5	4243
10	PI 639979 PSP	69	53	30	165	29.3	4147
11	Aa135	61	55	28	110	28.6	4128
12	PI 560968	70	56	27	131	27.4	4083
13	SPECTER	74	65	25	119	26.7	4070
14	PI 377693	76	49	30	154	27.7	4061
15	FENN	70	63	37	112	28.2	3910
16	PS03101269	84	66	32	148	25.2	3878
17	WINDHAM	42	36	18	140	24.8	3871
18	KALOFR	65	44	27	120	26.4	3862
19	WINTER 1 MM	79	43	24	158	23.0	3826
20	MIR	87	66	34	159	26.2	3818
21	GLACIER	45	30	22	126	26.1	3800
22	PI 639981	71	53	26	125	27.1	3786
23	PI 517926	71	47	25	101	30.2	3720
24	L 805/5g	66	44	28	155	30.1	3561
25	W6 12723	81	59	27	137	27.1	3553
26	WINTER	84	57	27	128	29.3	3491
27	G 10946	60	44	20	143	30.3	3437
28	ROMACK	72	54	24	124	28.7	3431
29	PI 343998	58	48	31	91	26.6	3379
30	LYNX	39	31	11	138	26.3	3364
31	LINE 340/11	56	47	13	179	24.5	3261
32	PI 517924	38	28	10	133	25.9	2914
33	ONTOFO	70	50	12	194	28.8	2419
34	PI 517925	47	25	5	115	26.4	1936
35	VARDIM 1	43	42	15	227	27.5	1678
36	LINE 179						
	<b>mean</b>	<b>66.2</b>	<b>50.5</b>	<b>24.9</b>	<b>135.0</b>	<b>27.1</b>	<b>3744.6</b>

<b>std</b>	<b>15.2</b>	<b>12.7</b>	<b>8.3</b>	<b>27.2</b>	<b>1.7</b>	<b>682.3</b>
<b>min</b>	<b>38</b>	<b>25</b>	<b>5</b>	<b>91</b>	<b>23.0</b>	<b>1678</b>
<b>max</b>	<b>87</b>	<b>76</b>	<b>39</b>	<b>227</b>	<b>30.3</b>	<b>4799.259</b>
<b>cv (%)</b>	<b>23</b>	<b>25</b>	<b>33</b>	<b>20</b>	<b>6</b>	<b>18</b>
<b>error variance</b>	<b>230.9</b>	<b>161.7</b>	<b>69.2</b>	<b>738.5</b>	<b>2.8</b>	<b>465547.4</b>

Table A10. Data summary of germplasm lines Huntley 2023.

<b>SN</b>	<b>name</b>	<b>vinelen</b>	<b>cano</b>	<b>pod</b>	<b>podht</b>	<b>flwrn</b>	<b>reprn</b>	<b>podp</b>	<b>standc</b>	<b>standc</b>	<b>survi</b>	<b>thsee</b>	<b>prot</b>	<b>yld</b>
		<b>gth</b>	<b>py</b>	<b>ht</b>	<b>mat</b>	<b>ode</b>	<b>ode</b>	<b>ed</b>	<b>ntfl</b>	<b>ntsp</b>	<b>val</b>	<b>dwt</b>	<b>ein</b>	
1	MELROSE	148	41	49	3	13	9	1	32	15	49	112	25.3	2249
2	L 805/5g	133	44	51	3	14	10	2	27	19	71	117	25.2	2198
3	GRANGER	148	50	40	2	14	4	2	24	28	121	134	25.3	1776
4	PI 517926	157	34	31	3	14	14	2	30	17	58	102	25.8	1751
5	W6 12713	118	38	20	4	13	14	2	26	29	116	119	25.7	1700
6	KALOFER	159	51	51	5	11	12	2	31	23	73	113	25.2	1674
7	SPECTER	132	52	31	3	13	11	1	30	13	41	143	23.9	1499
8	PI 377693	143	25	43	2	14	10	1	26	27	105	120	26.6	1061
9	RENN	169	33	33	2	12	14	2	34	12	34	103	25.8	1061
10	PS0310126	99	50	25	3	8	6	1	26	23	87	153	23.9	1022
11	W6 12723	127	29	41	3	13	9	2	32	13	40	124	26.2	1022
12	P-226	161	31	46	3	11	12	2	24	15	64	133	25.7	1010
13	FENN	164	59	39	3	17	8	1	33	26	78	117	25.9	971
14	PI 639981	127	35	34	2	11	12	1	30	6	21	121	25.6	895
15	PI 574505	138	35	35	2	10	11	1	30	30	100	120	25.6	869
16	WINTER	137	34	37	4	9	10	2	22	18	91	129	25.1	843
17	PI 560968	86	39	27	5	10	8	2	27	19	72	122	24.3	716

18	KENEJA	130	36	55	4	8	8	2	30	17	58	123	24.9	677
19	ROMACK	122	40	44	1	12	8	1	27	33	126	121	25.4	626
20	Aa135		20		1				27	13	49	110	25.1	588
21	LYNX	91	36	24	2	12	11	2	28	18	63	140	25.9	486
22	MIR	165	27	49	1	15	17	2	25	16	65	136	24.2	473
23	PI 517924	78	20	18	1	10	11	1	27	26	95	138	24.9	422
24	PI 517925	66	32	26	1	12	15	1	21	12	58	124	24.7	319
25	PI 343998	88	27	33	1	14	7	2	27	19	63	117	24.2	294
26	PI 517922	80	53	21	3	11	10	2	33	29	90	116	23.4	281
27	G 10946	102	33	33	4	9	9	2	26	13	46	125	25.3	236
28	PI 517923	72	21	23	2	11	6	2	22	22	99	160	25.8	179
29	GLACIER	73	28	23	3	10	12	2	30	23	81	115	25.2	166
30	ONTOFO	76	20	23	2	11	9	1	30	23	64	195	24.0	153
31	WINDHA	74	42	25	2	13	12	1	28	20	74	147	24.5	153
32	LINE 179	111	20	30	1	8	13	2	29	25	86	147	24.6	141
33	LINE	49	11	25	1	10	11	2	28	8	31	182	24.9	77
34	VARDIM 1	48	24	11	2	7	7	1	32	16	52	275	24.7	77
35	WINTER 1	92	7	31	1	9	10	1	27	10	36	188	23.7	77
36	PI 639979	113	13	28	1	12	11	1	26	15	63	167	24.5	64
	<b>mean</b>	<b>113.5</b>	<b>32.8</b>	<b>32.8</b>	<b>2.2</b>	<b>11.3</b>	<b>10.1</b>	<b>1.4</b>	<b>27.7</b>	<b>19.1</b>	<b>70.0</b>	<b>136.2</b>	<b>25.0</b>	<b>772.4</b>
	<b>sd</b>	<b>35.4</b>	<b>12.4</b>	<b>10.7</b>	<b>1.1</b>	<b>2.2</b>	<b>2.7</b>	<b>0.4</b>	<b>3.1</b>	<b>6.7</b>	<b>25.9</b>	<b>32.9</b>	<b>0.8</b>	<b>631.6</b>
	<b>min</b>	<b>48.0</b>	<b>7.0</b>	<b>11.0</b>	<b>1.0</b>	<b>7.0</b>	<b>4.0</b>	<b>1.0</b>	<b>21.0</b>	<b>6.0</b>	<b>21.0</b>	<b>102.0</b>	<b>23.4</b>	<b>64.0</b>
	<b>max</b>	<b>169.0</b>	<b>59.0</b>	<b>55.0</b>	<b>5.0</b>	<b>17.0</b>	<b>17.0</b>	<b>2.0</b>	<b>34.0</b>	<b>33.0</b>	<b>126.0</b>	<b>275.0</b>	<b>27.0</b>	<b>2249.</b>
	<b>cv (%)</b>	<b>31</b>	<b>38</b>	<b>33</b>	<b>50</b>	<b>19</b>	<b>27</b>	<b>28</b>	<b>11</b>	<b>35</b>	<b>37</b>	<b>24</b>	<b>3</b>	<b>82</b>
	<b>error variance</b>	<b>1251.0</b>	<b>153.</b>	<b>114.</b>	<b>1.2</b>	<b>4.8</b>	<b>7.2</b>	<b>0.2</b>	<b>9.9</b>	<b>44.9</b>	<b>673.3</b>	<b>1079.</b>	<b>0.6</b>	<b>39888</b>
			<b>2</b>	<b>8</b>								<b>4</b>		<b>1.9</b>

Table A11. Data summary of germplasm lines Moccasin 2023.

S	name	vinelen	cano	pod	podht	flwrn	reprn	podp	standc	standc	survi	thsee	prot	yld
N		gth	py	ht	mat	ode	ode	ed	ntfl	ntsp	val	dwt	ein	
1	PI 517925	87	39	28	12	13	9	2	23	16	80	134	24.2	4737
2	PI 517922	70	58	50	10	16	9	2	10	12	124	123	24.0	4473
3	PI 517923	80	64	32	13	11	6	2	8	23	268	153	23.6	4378
4	PI 517924	72	32	28	4	10	9	2	23	19	96	137	23.8	4043
5	WINDHA	76	63	37	11	13	12	2	14	15	91	150	23.6	3995
6	LYNX	89	47	28	6	12	8	2	15	25	171	138	23.5	3804
7	LINE	74	52	30	5	9	6	2	18	30	174	169	23.3	3732
8	GLACIER	70	43	28	11	10	8	2	6	7	156	137	23.7	3684
9	PI 377693	150	51	58	8	16	9	2	16	15	92	111	24.1	3660
10	PI 574505	129	38	65	6	15	7	2	20	9	49	128	24.7	3636
11	FENN	167	43	59	9	15	9	2	11	21	210	118	23.8	3469
12	SPECTER	150	53	59	4	12	8	2	18	26	169	141	23.1	3365
13	GRANGER	152	43	78	8	14	10	2	9	16	159	137	24.1	3301
14	P-226	145	59	35	9	15	7	2	12	17	122	144	23.8	3277
15	ROMACK	129	21	86	8	13	6	2	15	28	192	130	24.2	3277
16	W6 12723	176	68	102	13	18	8	2	12	13	115	130	24.3	2954
17	MELROSE	159	27	69	7	16	14	2	9	20	210	115	24.0	2918
18	RENN	168	47	88	6	13	9	2	13	12	100	111	24.3	2918
19	KALOFER	164	28	104	10	14	7	2	7	15	228	113	24.3	2871
20	KENEJA	189	53	92	7	13	11	2	15	6	100	132	23.0	2847
21	PI 639981	130	50	81	11	14	6	1	15	16	109	127	23.8	2847
22	PS0310126	174	66	51	8	14	11	2	14	13	83	164	24.2	2775
23	MIR	96	60	47	16	10	5	2	17	2	10	138	23.2	2751
24	PI 560968	118	61	42	5	10	10	2	18	11	58	124	21.8	2440
25	W6 12713	154	54	82	5	13	13	2	13	17	152	128	24.1	2440

26	PI 517926	149	49	73	9	13	15	2	16	22	129	108	24.6	2297
27	WINTER	131	51	59	8	16	7	2	11	9	82	128	22.5	1938
28	LINE 179	84	62	28	2	13	6	2	17	10	73	177	23.6	1818
29	G 10946	168	60	53	4	16	12	2	13	17	108	135	23.0	1698
30	L 805/5g	136	45	53	8	12	12	2	16	17	117	128	23.7	1651
31	PI 343998	91	41	26	3	12	7	1	10	17	151	105	23.1	1483
32	PI 639979	145	41	64	3	16	10	1	12	26	220	191	24.3	1316
33	WINTER 1	98	30	41	2	10	7	2	6	25	456	173	22.6	742
34	ONTOFO	136	39	46	4	13	11	2	11	22	203	199	23.8	646
35	Aa135	86	26	26	10	7	12	2	15	18	132	91	24.8	478
36	VARDIM 1	69	28	27	2	11	10	2	13	16	134	266	24.3	407
	<b>mean</b>	<b>123.7</b>	<b>46.8</b>	<b>54.1</b>	<b>7.2</b>	<b>12.8</b>	<b>8.8</b>	<b>1.8</b>	<b>13.4</b>	<b>16.6</b>	<b>142.3</b>	<b>139.7</b>	<b>23.7</b>	<b>2751.8</b>
	<b>sd</b>	<b>37.62</b>	<b>12.6</b>	<b>23.4</b>	<b>3.49</b>	<b>2.38</b>	<b>2.45</b>	<b>0.32</b>	<b>4.16</b>	<b>6.44</b>	<b>77.64</b>	<b>31.95</b>	<b>0.65</b>	<b>1149.52</b>
	<b>Min</b>	<b>69</b>	<b>21</b>	<b>26</b>	<b>2</b>	<b>7</b>	<b>5</b>	<b>1</b>	<b>6</b>	<b>2</b>	<b>10</b>	<b>91</b>	<b>21.8</b>	<b>407</b>
	<b>max</b>	<b>189</b>	<b>68</b>	<b>104</b>	<b>16</b>	<b>18</b>	<b>15</b>	<b>2</b>	<b>23</b>	<b>30</b>	<b>456</b>	<b>266</b>	<b>24.8</b>	<b>4737</b>
	<b>cv (%)</b>	<b>30</b>	<b>27</b>	<b>43</b>	<b>49</b>	<b>19</b>	<b>28</b>	<b>18</b>	<b>31</b>	<b>39</b>	<b>55</b>	<b>23</b>	<b>3</b>	<b>42</b>
	<b>error variance</b>	<b>1415.3</b>	<b>160.4</b>	<b>548.3</b>	<b>12.2</b>	<b>5.7</b>	<b>6.0</b>	<b>0.1</b>	<b>17.3</b>	<b>41.5</b>	<b>6027.2</b>	<b>1021.0</b>	<b>0.4</b>	<b>132140.6</b>

APPENDIX B

TABLES OF BREEDING LINES INCLUDED IN THE  
EXPERIMENT

Table B1. List of breeding lines included in experiment in 2021.

SN	Name	Yield (kg/ha)	TSW (g)	Protein (%)	Pedigree)
1	LYNX	1980	134	23.9	
2	MTP190186	974	194	22.2	LIFTER/CHEROKEE
3	MTP190188				LIFTER/CHEROKEE
4	MTP190193	964	200	24.8	LIFTER/CHEROKEE
5	MTP190201				LIFTER/EFB333
6	MTP190203				LIFTER/EFB333
7	MTP190210	280	200	24.6	LIFTER/IGLOO
8	MTP190211				LIFTER/IGLOO
9	MTP190212				LIFTER/IGLOO
10	MTP190215	236	220	24.9	LIFTER/IGLOO
11	MTP190219	254	210		LIFTER/IGLOO
12	MTP190220	814	170		LIFTER/IGLOO
13	MTP190222				LIFTER/IGLOO
14	MTP190223	746	180	24.1	LIFTER/IGLOO
15	MTP190267	554	250	24.5	APACHE/LIFTER
16	MTP190270	1477	182	21.2	APACHE/LIFTER
17	MTP190272	1676	183	25.2	APACHE/LIFTER
18	MTP190278	823	230		APACHE/LIFTER
19	MTP190279	394	200	25.0	APACHE/LIFTER
20	MTP190280				APACHE/LIFTER
21	MTP190281	530	193		APACHE/LIFTER
22	MTP190295	378	250	24.5	ARAVIS/LIFTER
23	MTP190296	283	260	24.8	ARAVIS/LIFTER
24	MTP190331	1731	182	25.4	ASSAS/LIFTER
25	MTP190332	960	220	24.3	ASSAS/LIFTER
26	MTP190345	965	200		CHEROKEE/LIFTER
27	MTP190346	384	200		CHEROKEE/LIFTER
28	MTP190355	447	220	24.4	CHEYENNE/LIFTER
29	MTP190356	495	190	24.3	CHEYENNE/LIFTER
30	MTP190359	312	200		CHEYENNE/LIFTER
31	MTP190362	907	175	23.7	CHEYENNE/LIFTER
32	MTP190363				CHEYENNE/LIFTER
33	MTP190368	233	270	24.9	CHEYENNE/LIFTER
34	MTP190376				DOVE/LIFTER
35	MTP190377	904	170	25.1	DOVE/LIFTER
36	MTP190379				DOVE/LIFTER
37	MTP190382	455	200		DOVE/LIFTER
38	MTP190383	771	173	25.1	DOVE/LIFTER

39	MTP190389	1284	175	23.4	DOVE/LIFTER
40	MTP190390	386	220	24.3	DOVE/LIFTER
41	MTP190392	299	200	24.5	DOVE/LIFTER
42	MTP190401				DOVE/LIFTER
43	MTP190403	895	180	23.8	DOVE/LIFTER
44	MTP190409	187	230	25.0	DOVE/LIFTER
45	MTP190410	615	230	24.6	DOVE/LIFTER
46	MTP190415				GRANA/LIFTER
47	MTP190423				GRANA/LIFTER
48	MTP190424				GRANA/LIFTER
49	MTP190438				ISARD/LIFTER
50	MTP190441	673	230	24.4	ISARD/LIFTER
51	MTP190443	543	220	24.5	ISARD/LIFTER
52	MTP190444	1783	161	22.0	ISARD/LIFTER
53	MTP190446	2007	160	24.9	ISARD/LIFTER
54	MTP190453	404	200	24.9	ISARD/LIFTER
55	MTP190456	668	230	24.8	ISARD/LIFTER
56	MTP190458	1416	210	19.9	ISARD/LIFTER
57	MTP190460				NATURA/LIFTER
58	MTP190464				NATURA/LIFTER
59	MTP190465				NATURA/LIFTER
60	MTP190469				NATURA/LIFTER
61	MTP190477	148	280	25.1	NATURA/LIFTER
62	MTP190479	515	260		NATURA/LIFTER
63	MTP190482	457	270	25.0	NATURA/LIFTER
64	MTP190487				NATURA/LIFTER
65	MTP190489				NATURA/LIFTER
66	MTP190503				NATURA/LIFTER
67	MTP190507				NATURA/LIFTER
68	MTP190508				NATURA/LIFTER
69	MTP190514				NATURA/LIFTER
70	MTP190516				NATURA/LIFTER
71	MTP190520				NATURA/LIFTER
72	MTP190521				NATURA/LIFTER
73	MTP190522	760	236	26.2	NATURA/LIFTER
74	MTP190533				NATURA/LIFTER
75	MTP190546				NATURA/LIFTER
76	MTP190548				NATURA/LIFTER
77	MTP190558	841	170		CDC MEADOW/DOVE
78	MTP190563	319	180	24.0	CDC MEADOW/DOVE

79	MTP190566	568	170		CDC MEADOW/DOVE
80	MTP190568	773	170		CDC MEADOW/DOVE
81	MTP190570	1168	174	25.0	LIFTER/CHEROKEE
82	MTP190571	406	215	25.1	LIFTER/CHEROKEE
83	MTP190573	419	205	24.7	LIFTER/CHEROKEE
84	MTP190574	1721	170	21.4	LIFTER/CHEROKEE
85	MTP190577	1260	193	22.6	LIFTER/IGLOO
86	MTP190578	948	179	24.5	LIFTER/IGLOO
87	MTP190582	1530	193	22.7	STIRLING/CHEROKEE
88	MTP190583	1021	191	24.8	STIRLING/CHEROKEE
89	MTP190584	880	202	25.4	STIRLING/CHEROKEE
90	MTP190587	346	205	23.7	APACHE/HAMPTON
91	MTP190588				APACHE/HAMPTON
92	MTP190591	804	215		APACHE/LIFTER
93	MTP190596	2151	181	20.5	APACHE/LIFTER
94	MTP190597	200	220	24.4	APACHE/LIFTER
95	MTP190619	653	205		DOVE/LIFTER
96	MTP190624	759	257	25.0	DOVE/STABIL
97	MTP190625	360	210		DOVE/STABIL
98	MTP190629	983	135		EFB333/WINDHAM
99	MTP190633	2532	198	24.5	GRANA/LIFTER
100	MTP190635	905	206	25.6	GRANA/LIFTER
101	MTP190636	743	210	24.5	GRANA/ARAVIS
102	MTP190638				GRANA/ARAVIS
103	MTP190641	481	235	24.6	GRANA/ISARD
104	MTP190642	972	190	23.6	GRANA/ISARD
105	MTP190646				GRANA/NATURA
106	MTP190650				NATURA/LIFTER
107	MTP190660				NATURA/ARAVIS
108	MTP190662	554	210		NATURA/ARAVIS
109	MTP190664	2905	139	24.2	WINDHAM/ASSAS
110	MTP190665	1246	190		APACHE/DS ADMIRAL
111	MTP190666				APACHE/DS ADMIRAL
112	MTP190667	970	167	24.5	APACHE/DS ADMIRAL
113	MTP190669	1208	185	24.3	APACHE/DS ADMIRAL
114	MTP190674	943	185	23.9	APACHE/HAMPTON
115	MTP190678	1962	174	20.4	APACHE/HAMPTON
116	MTP190681	1600	208	21.3	APACHE/STABIL
117	MTP190684	1133	199	25.0	APACHE/STABIL
118	MTP190694	869	160		APRIL/DS ADMIRAL
119	MTP190696				APRIL/DS ADMIRAL

120	MTP190701	1299	175	21.9	ARAVIS/HAMPTON
121	MTP190711	1972	190	24.6	ARAVIS/LYNX
122	MTP190712	814	195	24.3	ARAVIS/LYNX
123	MTP190717				ARAVIS/LYNX
124	MTP190720				ASSAS/STABIL
125	MTP190724	1090	182	23.5	CHEROKEE/HAMPTON
126	MTP190733	898	191	24.5	CHEROKEE/DS
127	MTP190734	972	210	24.2	CHEROKEE/DS
128	MTP190739	1232	231	24.8	CHEROKEE/STABIL
129	MTP190740	1200	210	22.5	CHEROKEE/STABIL
130	MTP190742	1517	202	21.1	CHEROKEE/STABIL
131	MTP190746				CHEROKEE/STIRLING
132	MTP190747	1194	181	24.0	CHEROKEE/STIRLING
133	MTP190749	1281		24.9	CHEROKEE/STIRLING
134	MTP190751	3013	130	22.5	CHEROKEE/WINDHAM
135	MTP190752	2906	164	20.2	CHEROKEE/WINDHAM
136	MTP190753	3613	155	21.5	CHEROKEE/WINDHAM
137	MTP190760				CHEYENNE/HAMPTON
138	MTP190762				CHEYENNE/STABIL
139	MTP190763				CHEYENNE/STIRLING
140	MTP190766	2266	171	22.6	CHEYENNE/WINDHAM
141	MTP190767	2560	162	22.2	CHEYENNE/WINDHAM
142	MTP190769	2544	167	20.8	CHEYENNE/WINDHAM
143	MTP190774	1795	169	21.7	DOVE/HAMPTON
144	MTP190776	2450	186	22.3	DOVE/DS ADMIRAL
145	MTP190782	1388	181	21.9	DOVE/STIRLING
146	MTP190786	1184	189	24.0	DOVE/STIRLING
147	MTP190787	1276	174	23.8	DOVE/STIRLING
148	MTP190792	1120	193	25.3	GRANA/HAMPTON
149	MTP190794	1515	223	24.5	GRANA/STABIL
150	MTP190796	1753	212	23.9	GRANA/STABIL
151	MTP190797	1402	237	24.4	GRANA/STABIL
152	MTP190798	1161	223	24.1	GRANA/STABIL
153	MTP190803	905	212	24.7	GRANA/STIRLING
154	MTP190805				GRANA/STIRLING
155	MTP190806	575	210	24.6	IGLOO/DS ADMIRAL
156	MTP190809				IGLOO/DS ADMIRAL
157	MTP190810	1145	178	26.2	IGLOO/DS ADMIRAL
158	MTP190812	1352	197	23.2	IGLOO/STABIL
159	MTP190818				IGLOO/STIRLING
160	MTP190820	2858	150	24.6	IGLOO/WINDHAM

161	MTP190821	2943	154	25.4	IGLOO/WINDHAM
162	MTP190822	2903	131	25.6	IGLOO/WINDHAM
163	MTP190823	959	220	24.2	ISARD/HAMPTON
164	MTP190825	649	215	24.2	ISARD/HAMPTON
165	MTP190826	487	210	24.0	ISARD/HAMPTON
166	MTP190833	2252	159	23.7	ISARD/DS ADMIRAL
167	MTP190836	994	210		ISARD/DS ADMIRAL
168	MTP190837	911	181	24.8	ISARD/DS ADMIRAL
169	MTP190838	1704	171	24.3	ISARD/DS ADMIRAL
170	MTP190840	575	200		ISARD/DS ADMIRAL
171	MTP190843	724	205	24.2	ISARD/STABIL
172	MTP190844	1034	220		ISARD/STABIL
173	MTP190847				ISARD/STABIL
174	MTP190849	951	201	24.1	ISARD/STABIL
175	MTP190850	1512	190	21.4	ISARD/STABIL
176	MTP190853	1420	192	21.2	ISARD/STIRLING
177	MTP190854	2787	106	22.8	ISARD/STIRLING
178	MTP190855				ISARD/STIRLING
179	MTP190857	930	208	25.5	NATURA/DS ADMIRAL
180	MTP190861	791	180	25.7	NATURA/DS ADMIRAL
181	MTP190868	1227	228	24.8	NATURA/STABIL
182	MTP190871	1119	215	24.9	NATURA/STABIL
183	MTP190872	1767	218	25.7	NATURA/STABIL
184	MTP190875	742	208	24.1	NATURA/STIRLING
185	MTP190877	679	199	24.1	NATURA/STIRLING
186	MTP190882				NATURA/STIRLING
187	MTP190886				NATURA/STIRLING
188	MTP190887	2596	159	25.0	NATURA/WINDHAM
189	MTP190891	2533	165	24.2	NATURA/WINDHAM
190	MTP190918	994	186	25.0	M17P017/LIFTER
191	MTP190953	1081	217	25.2	M17P044/LYNX
192	MTP190960	843	223	24.3	M17P044/LYNX
193	MTP190966				M17P061/LYNX
194	MTP190968				M17P061/LYNX
195	MTP190972	900	176	24.5	M17P061/LYNX
196	MTP190975	1006	194	24.5	M17P061/LYNX
197	MTP190978	753	188	24.7	M17P062/WINDHAM
198	MTP190980				M17P062/WINDHAM
199	MTP191028	2845	151	25.5	M17P083/WINDHAM
200	MTP191029	2662	141	25.2	M17P083/WINDHAM
201	MTP191030	2831	165	25.1	M17P083/WINDHAM

202	MTP191031	2614	163	22.3	M17P083/WINDHAM
203	SPECTER	2007	125	23.6	

Table B2. List of breeding lines included in experiment in 2022.

SN	Name	Yield (kg/ha)	TSW (g)	Protein (%)	Pedigree
1	LYNX	2062	125	22.4	
2	MTP190186	769	205	19.8	LIFTER/CHEROKEE
3	MTP190270	1806	199	18.6	APACHE/LIFTER
4	MTP190272	1755	190	24.8	APACHE/LIFTER
5	MTP190331	1532	170	25.3	ASSAS/LIFTER
6	MTP190389	1605	175	21.4	DOVE/LIFTER
7	MTP190444	1966	161	18.8	ISARD/LIFTER
8	MTP190446	2194	163	24.1	ISARD/LIFTER
9	MTP190458	1744	210	17.7	ISARD/LIFTER
10	MTP190570	1121	175	25.2	LIFTER/CHEROKEE
11	MTP190574	2086	177	17.9	LIFTER/CHEROKEE
12	MTP190577	1533	207	19.8	LIFTER/IGLOO
13	MTP190582	1704	202	20.7	STIRLING/CHEROKEE
14	MTP190583	1056	191	24.7	STIRLING/CHEROKEE
15	MTP190596	2427	185	18.9	APACHE/LIFTER
16	MTP190664	2244	137	24.0	WINDHAM/ASSAS
17	MTP190678	1754	175	19.1	APACHE/HAMPTON
18	MTP190681	1396	220	19.4	APACHE/STABIL
19	MTP190684	1562	203	24.5	APACHE/STABIL
20	MTP190701	1610	177	20.1	ARAVIS/HAMPTON
21	MTP190739	1063	249	24.0	CHEROKEE/STABIL
22	MTP190740	1014	224	19.3	CHEROKEE/STABIL
23	MTP190742	1299	206	18.9	CHEROKEE/STABIL
24	MTP190747	1478	185	23.8	CHEROKEE/STIRLING
25	MTP190751	1927	127	19.9	CHEROKEE/WINDHAM
26	MTP190752	2178	168	18.6	CHEROKEE/WINDHAM
27	MTP190753	2448	161	19.9	CHEROKEE/WINDHAM
28	MTP190766	1699	173	21.5	CHEYENNE/WINDHAM
29	MTP190767	2152	157	19.6	CHEYENNE/WINDHAM
30	MTP190769	2467	169	19.0	CHEYENNE/WINDHAM
31	MTP190774	1940	172	20.5	DOVE/HAMPTON
32	MTP190782	1698	180	20.6	DOVE/STIRLING
33	MTP190786	1540	186	24.2	DOVE/STIRLING
34	MTP190787	1364	174	23.9	DOVE/STIRLING

35	MTP190812	1431	211	20.8	IGLOO/STABIL
36	MTP190820	1666	150	25.3	IGLOO/WINDHAM
37	MTP190821	2052	148	25.6	IGLOO/WINDHAM
38	MTP190822	1676	126	25.8	IGLOO/WINDHAM
39	MTP190850	1743	195	19.4	ISARD/STABIL
40	MTP190853	1819	203	19.9	ISARD/STIRLING
41	MTP190854	1889	103	20.5	ISARD/STIRLING
42	MTP190887	1882	150	25.5	NATURA/WINDHAM
43	MTP190891	1802	164	24.7	NATURA/WINDHAM
44	MTP190918	987	186	24.7	M17P017/LIFTER
45	MTP190960	798	234	24.0	M17P044/LYNX
46	MTP190972	880	190	24.5	M17P061/LYNX
47	MTP191028	1852	145	25.3	M17P083/WINDHAM
48	MTP191029	2220	137	25.4	M17P083/WINDHAM
49	MTP191030	1985	159	25.2	M17P083/WINDHAM
50	MTP191031	2101	162	20.0	M17P083/WINDHAM
51	SPECTER	1920	122	21.9	

Table B3. List of breeding lines included in experiment in 2023.

SN	Name	Yield (kg/ha)	TSW (g)	Protein (%)	Pedigree
1	LYNX	3626	135	24.1	
2	MTP190186	884	197	22.5	LIFTER/CHEROKEE
3	MTP190270	1328	193	22.5	APACHE/LIFTER
4	MTP190272	1824	197	24.9	APACHE/LIFTER
5	MTP190331	2284	187	25.2	ASSAS/LIFTER
6	MTP190389	1174	181	23.7	DOVE/LIFTER
7	MTP190444	2252	167	22.3	ISARD/LIFTER
8	MTP190446	2326	168	24.6	ISARD/LIFTER
9	MTP190570	982	176	25.0	LIFTER/CHEROKEE
10	MTP190574	1863	180	22.3	LIFTER/CHEROKEE
11	MTP190577	906	197	23.3	LIFTER/IGLOO
12	MTP190582	1598	197	23.2	STIRLING/CHEROKEE
13	MTP190583	445	189	24.6	STIRLING/CHEROKEE
14	MTP190596	2286	188	21.6	APACHE/LIFTER
15	MTP190664	3801	146	24.1	WINDHAM/ASSAS
16	MTP190678	2094	183	21.4	APACHE/HAMPTON
17	MTP190681	1834	213	21.9	APACHE/STABIL
18	MTP190684	902	213	24.9	APACHE/STABIL
19	MTP190701	951	179	22.5	ARAVIS/HAMPTON
20	MTP190739	1054	232	24.8	CHEROKEE/STABIL

21	MTP190740	1153	208	22.8	CHEROKEE/STABIL
22	MTP190742	1550	212	22.2	CHEROKEE/STABIL
23	MTP190747	1003	190	23.9	CHEROKEE/STIRLING
24	MTP190751	4513	138	22.9	CHEROKEE/WINDHAM
25	MTP190752	3528	170	21.4	CHEROKEE/WINDHAM
26	MTP190753	4779	161	22.4	CHEROKEE/WINDHAM
27	MTP190766	2848	174	22.9	CHEYENNE/WINDHAM
28	MTP190767	3481	166	22.9	CHEYENNE/WINDHAM
29	MTP190769	2846	170	21.3	CHEYENNE/WINDHAM
30	MTP190774	1512	174	22.3	DOVE/HAMPTON
31	MTP190782	958	191	22.7	DOVE/STIRLING
32	MTP190786	482	199	24.1	DOVE/STIRLING
33	MTP190787	955	181	23.8	DOVE/STIRLING
34	MTP190812	1134	201	23.7	IGLOO/STABIL
35	MTP190820	3828	159	24.4	IGLOO/WINDHAM
36	MTP190821	3830	159	24.8	IGLOO/WINDHAM
37	MTP190822	4582	138	25.0	IGLOO/WINDHAM
38	MTP190850	1261	204	22.3	ISARD/STABIL
39	MTP190853	1012	201	22.2	ISARD/STIRLING
40	MTP190854	3687	111	23.8	ISARD/STIRLING
41	MTP190887	3537	162	24.2	NATURA/WINDHAM
42	MTP190891	3131	172	24.0	NATURA/WINDHAM
43	MTP190918	780	183	24.9	M17P017/LIFTER
44	MTP190960	507	220	24.3	M17P044/LYNX
45	MTP190972	728	176	24.5	M17P061/LYNX
46	MTP191028	3880	157	24.8	M17P083/WINDHAM
47	MTP191029	3628	147	24.8	M17P083/WINDHAM
48	MTP191030	4149	170	24.7	M17P083/WINDHAM
49	MTP191031	3819	167	22.8	M17P083/WINDHAM
50	SPECTER	3749	126	23.5	

Table B4. Overall ranking of breeding lines.

SN	Name	Yield (kg/ha)	TSW (g)	Protein (%)
1	MTP190753	3662	161	24.2
2	MTP190822	3296	141	25.0
3	MTP190664	3285	148	24.1
4	MTP190820	3129	159	24.4
5	MTP191028	3087	158	24.8
6	MTP190821	2975	158	24.8

7	MTP190752	2906	170	23.0
8	MTP191030	2901	170	24.7
9	MTP190769	2895	170	23.0
10	MTP190751	2867	138	24.5
11	MTP191031	2820	167	24.6
12	MTP191029	2808	147	24.8
13	MTP190854	2787	111	25.8
14	MTP190766	2440	174	23.9
15	MTP190887	2369	159	24.2
16	MTP190891	2345	172	24.0
17	MTP190767	2325	166	24.6
18	MTP190596	2242	188	23.9
19	MTP190446	2183	170	24.6
20	SPECTER	2109	124	24.6
21	LYNX	2056	132	25.5
22	MTP190678	1962	183	23.3
23	MTP190574	1910	178	24.3
24	MTP190444	1901	167	24.4
25	MTP190774	1795	174	23.3
26	MTP190272	1785	197	24.9
27	MTP190331	1778	186	25.2
28	MTP190458	1737	211	24.6
29	MTP190681	1708	213	23.8
30	MTP190850	1655	205	23.2
31	MTP190582	1621	196	24.2
32	MTP190701	1559	179	23.4
33	MTP190787	1557	184	23.8
34	MTP190812	1532	203	24.6
35	MTP190742	1517	212	24.1
36	MTP190270	1501	193	24.4
37	MTP190853	1494	200	23.2
38	MTP190786	1457	200	24.1
39	MTP190739	1432	226	24.8
40	MTP190389	1417	182	24.7
41	MTP190782	1388	191	23.7
42	MTP190583	1380	186	24.6
43	MTP190570	1359	177	25.0
44	MTP190577	1326	197	25.0
45	MTP190747	1286	189	23.9
46	MTP190740	1285	208	24.8

47	MTP190960	1160	221	24.3
48	MTP190684	1139	212	24.9
49	MTP190186	1058	197	25.4
50	MTP190972	1046	171	24.5
51	MTP190918	994	183	24.9
	<b>Mean</b>	<b>2004</b>	<b>178</b>	<b>24.4</b>
	<b>SD</b>	<b>708</b>	<b>25.1</b>	<b>0.6</b>
	<b>CV (%)</b>	<b>35</b>	<b>14</b>	<b>2</b>
	<b>Min</b>	<b>994</b>	<b>110</b>	<b>23.0</b>
	<b>Max</b>	<b>3662</b>	<b>226</b>	<b>25.8</b>

Table B5. Ranking of breeding lines by year.

SN	Name	2021	2022	2023	Sum of ranks	Overall rank
1	MTP190769	1	1	19	21	<b>1</b>
2	MTP190751	5	15	3	23	<b>2</b>
3	MTP191029	6	5	12	23	<b>3</b>
4	MTP190753	21	2	2	25	<b>4</b>
5	MTP191028	2	19	7	28	<b>5</b>
6	MTP191030	11	13	5	29	<b>6</b>
7	MTP190664	17	4	9	30	<b>7</b>
8	MTP191031	19	9	6	34	<b>8</b>
9	MTP190767	15	8	15	38	<b>9</b>
10	MTP190752	18	7	14	39	<b>10</b>
11	MTP190822	13	27	1	41	<b>11</b>
12	SPECTER	16	16	10	42	<b>12</b>
13	LYNX	23	11	13	47	<b>13</b>
14	MTP190766	4	26	18	48	<b>14</b>
15	MTP190446	24	6	20	50	<b>15</b>
16	MTP190820	20	28	4	52	<b>16</b>
17	MTP190821	32	12	8	52	<b>17</b>
18	MTP190891	25	22	16	63	<b>18</b>
19	MTP190596	43	3	21	67	<b>19</b>
20	MTP190854	39	17	11	67	<b>20</b>
21	MTP190681	7	41	25	73	<b>21</b>
22	MTP190850	3	37	33	73	<b>22</b>
23	MTP190678	29	24	23	76	<b>23</b>
24	MTP190444	41	14	22	77	<b>24</b>
25	MTP190574	40	10	27	77	<b>25</b>
26	MTP190812	8	39	31	78	<b>26</b>

27	MTP190774	22	29	30	81	<b>27</b>
28	MTP190701	14	30	40	84	<b>28</b>
29	MTP190577	9	34	42	85	<b>29</b>
30	MTP190887	50	18	17	85	<b>30</b>
31	MTP190331	28	35	24	87	<b>31</b>
32	MTP190272	47	23	26	96	<b>32</b>
33	MTP190270	44	21	32	97	<b>33</b>
34	MTP190740	10	46	41	97	<b>34</b>
35	MTP190389	33	31	34	98	<b>35</b>
36	MTP190853	42	20	36	98	<b>36</b>
37	MTP190582	49	25	28	102	<b>37</b>
38	MTP190742	34	43	29	106	<b>38</b>
39	MTP190583	12	45	50	107	<b>39</b>
40	MTP190782	30	40	38	108	<b>40</b>
41	MTP190786	26	33	49	108	<b>41</b>
42	MTP190787	27	42	39	108	<b>42</b>
43	MTP190684	38	32	43	113	<b>43</b>
44	MTP190739	31	48	35	114	<b>44</b>
45	MTP190570	45	44	37	126	<b>45</b>
46	MTP190747	48	36	44	128	<b>46</b>
47	MTP190972	35	50	48	133	<b>47</b>
48	MTP190960	37	51	47	135	<b>48</b>
49	MTP190186	46	49	46	141	<b>49</b>
50	MTP190918	51	47	45	143	<b>50</b>
51	MTP190458	36	38		74	<b>51</b>

Table B6. Ranking of breeding lines by location.

SN	Name	Bozeman	Havre	Huntley	Moccasin	Sum of ranks	Overall rank
1	MTP190753	1	11	1	2	15	<b>1</b>
2	MTP190820	6	4	4	6	20	<b>2</b>
3	MTP190822	2	9	5	14	30	<b>3</b>
4	MTP190664	15	7	6	4	32	<b>4</b>
5	MTP190751	3	17	11	3	34	<b>5</b>
6	MTP190821	12	2	9	12	35	<b>6</b>
7	MTP190752	4	21	7	7	39	<b>7</b>
8	MTP191030	7	8	14	10	39	<b>8</b>
9	MTP191028	13	10	8	9	40	<b>9</b>
10	MTP190854	8	19	13	1	41	<b>10</b>
11	MTP190891	10	3	16	23	52	<b>11</b>

12	MTP191031	9	18	3	24	54	<b>12</b>
13	MTP190767	22	13	10	11	56	<b>13</b>
14	MTP191029	5	1	41	13	60	<b>14</b>
15	MTP190887	14	14	12	21	61	<b>15</b>
16	MTP190766	16	30	2	28	76	<b>16</b>
17	MTP190678	17	29	27	5	78	<b>17</b>
18	MTP190769	11	12	49	8	80	<b>18</b>
19	MTP190446	20	6	42	20	88	<b>19</b>
20	LYNX	26	20	22	22	90	<b>20</b>
21	SPECTER	25	31	20	16	92	<b>21</b>
22	MTP190331	19	23	25	34	101	<b>22</b>
23	MTP190774	32	37	23	15	107	<b>23</b>
24	MTP190444	42	22	29	17	110	<b>24</b>
25	MTP190272	36	5	46	26	113	<b>25</b>
26	MTP190681	33	25	15	42	115	<b>26</b>
27	MTP190742	37	32	35	18	122	<b>27</b>
28	MTP190812	29	42	18	36	125	<b>28</b>
29	MTP190850	30	24	33	39	126	<b>29</b>
30	MTP190574	40	15	45	29	129	<b>30</b>
31	MTP190853	18	43	31	37	129	<b>31</b>
32	MTP190582	21	35	48	32	136	<b>32</b>
33	MTP190782	31	44	32	31	138	<b>33</b>
34	MTP190458	39	33	40	27	139	<b>34</b>
35	MTP190701	28	39	30	46	143	<b>35</b>
36	MTP190270	41	26	47	30	144	<b>36</b>
37	MTP190739	38	40	17	49	144	<b>37</b>
38	MTP190570	49	27	28	41	145	<b>38</b>
39	MTP190787	27	49	39	35	150	<b>39</b>
40	MTP190583	24	50	37	40	151	<b>40</b>
41	MTP190786	34	51	24	43	152	<b>41</b>
42	MTP190577	43	28	36	47	154	<b>42</b>
43	MTP190740	48	38	26	44	156	<b>43</b>
44	MTP190747	46	34	44	33	157	<b>44</b>
45	MTP190389	45	46	50	19	160	<b>45</b>
46	MTP190684	44	36	43	38	161	<b>46</b>
47	MTP190186	51	41	21	50	163	<b>47</b>
48	MTP190960	50	47	19	51	167	<b>48</b>
49	MTP190972	35	48	38	48	169	<b>49</b>
50	MTP190918	47	45	34	45	171	<b>50</b>

51      MTP190596                      23              16                                      25

Table B7. Yield (kg/ha) of breeding lines across locations averaged for three years.

Rank	Name	Bozeman	Havre	Huntley	Moccasin	Average
1	MTP190753	4621	3066	3577	3186	3613
2	MTP190822	4455	3345	3178	2171	3287
3	MTP190820	3366	3774	3190	2479	3202
4	MTP190751	3983	2697	2746	2760	3047
5	MTP190821	3021	3839	2890	2432	3046
6	MTP190664	2756	3531	3141	2580	3002
7	MTP190752	3681	2423	3049	2474	2907
8	MTP190854	3246	2658	2420	3230	2889
9	MTP191030	3311	3392	2395	2455	2888
10	MTP191028	2920	3104	2974	2473	2868
11	MTP190891	3220	3792	2187	1736	2734
12	MTP191031	3234	2684	3259	1671	2712
13	MTP191029	3449	3896	954	2317	2654
14	MTP190767	2217	2879	2885	2434	2604
15	MTP190887	2873	2844	2487	1757	2490
16	MTP190766	2734	1920	3277	1406	2334
17	MTP190769	3154	2992	263	2473	2221
18	MTP190596	2188	2757	NA	1490	2145
19	MTP190446	2262	3571	922	1773	2132
20	MTP190678	2496	2002	1264	2560	2081
21	LYNX	2124	2511	1660	1748	2011
22	SPECTER	2125	1869	1947	1910	1963
23	MTP190272	1667	3729	726	1474	1899
24	MTP190331	2301	2402	1528	1183	1854
25	MTP190681	1785	2334	2270	795	1796
26	MTP190774	1820	1563	1630	2067	1770
27	MTP190444	1400	2416	1259	1891	1742
28	MTP190574	1455	2834	769	1386	1611
29	MTP190742	1605	1793	1086	1866	1588
30	MTP190850	1890	2356	1185	910	1585
31	MTP190812	1937	1233	2037	955	1541
32	MTP190853	2338	1184	1201	954	1419
33	MTP190458	1464	1729	1014	1433	1410
34	MTP190782	1841	1148	1189	1340	1380
35	MTP190582	2241	1630	383	1255	1377

36	MTP190270	1413	2243	419	1371	1362
37	MTP190739	1471	1257	2101	601	1358
38	MTP190570	986	2191	1260	863	1325
39	MTP190577	1348	2101	1085	757	1323
40	MTP190701	1953	1257	1220	766	1299
41	MTP190747	1255	1711	822	1243	1258
42	MTP190787	2093	484	1026	1167	1193
43	MTP190684	1336	1600	878	944	1190
44	MTP190740	1072	1504	1309	777	1166
45	MTP190583	2144	475	1058	898	1144
46	MTP190389	1295	978	132	1862	1067
47	MTP190786	1681	201	1571	791	1061
48	MTP190186	558	1249	1768	494	1017
49	MTP190918	1077	1051	1093	769	998
50	MTP190972	1674	509	1048	731	991
51	MTP190960	927	800	1959	232	980
	<b>Mean</b>	<b>2225</b>	<b>2186</b>	<b>1714</b>	<b>1594</b>	
	<b>CV (%)</b>	<b>41.8</b>	<b>45.6</b>	<b>53.9</b>	<b>46.6</b>	
	<b>SD</b>	<b>929</b>	<b>998</b>	<b>924</b>	<b>743</b>	

Table B8. Data summary of breeding lines Bozeman 2023.

SN	name	vinelen	cano	pod	podht	flwrn	reprn	podp	standc	standcn	survi	thseed	prote	yld
		gth	py	ht	mat	ode	ode	ed	ntfl	tsp	val	wt	in	
1	MTP19082	79	68	25	18	13	12	2.5	11	10	98	153	24.4	5622
2	MTP19075	87	60	20	16	15	10	2	11	10	89	167	24.8	5335
3	MTP19076	104	78	18	16	14	21	2	8	2	80	161	23.8	4832
4	MTP19103	91	83	20	22	11	18	2	9	8	126	177	24.2	4665
5	MTP19102	94	83	19	18	9	15	1.5	12	11	99	158	24.4	4545
6	MTP19103	81	67	19	14	11	14	2	9	12	156	178	24.6	4498
7	MTP19075	90	66	20	17	11	16	2	8	10	120	150	24.4	4474
8	LYNX	89	44	21	11	11	12	2	12	11	115	137	25.0	4043
9	MTP19088	93	78	28	16	11	14	1.75	9	9	107	165	23.9	3708
10	MTP19102	77	49	25	12	14	13	2	8	9	103	161	24.7	3636
11	MTP19082	73	69	14	9	13	16	2	5	9	170	168	24.6	3541
12	MTP19076	99	68	21	14	13	11	1.75	14	9	60	163	24.2	3517
13	SPECTER	142	55	20	9	13	17	2	10	10	101	134	23.8	3469
14	MTP19085		44		9				16	16	106	123	25.6	3397
15	MTP19082	79	70	18	20	12	11	1.5	13	9	72	165	24.1	3349
16	MTP19075	75	77	15	15	10	14	2	7	12	186	173	24.1	3277
17	MTP19033	105	46	25	7	10	8	2	10	7	77	206	25.1	3206
18	MTP19089	104	84	25	29	11	15	1.5	13	12	96	179	24.6	3086
19	MTP19066	128	56	14	9	11	16	1.5	4	7	214	153	24.1	2895
20	MTP19044	66	54	11	11	8	8	1	13	13	106	185	24.7	2249
21	MTP19076	77	65	23	13	11	15	2	12	13	106	168	24.3	2010
22	MTP19067	63	59	15	7	9	10	1.5	12	10	87	208	24.5	1938
23	MTP19059	87	43	15	6	8	12	1.25	8	7	98	197	24.5	1794
24	MTP19058	65	47	11	12	10	11	1.75	9	10	111	196	24.4	1746

25	MTP19078	51	41	13	5	7	8	1	11	12	126	191	24.1	1220
26	MTP19085	66	38	13	9	9	15	1.75	6	7	106	201	24.3	1124
27	MTP19074	51	57	12	16	8	9	1	13	11	90	233	25.0	981
28	MTP19081	61	58	12	8	10	10	1.5	7	10	152	220	25.8	909
29	MTP19085	60	51	12	3	10	10	1.25	9	11	122	228	24.4	790
30	MTP19078	68	36	17	5	9	11	1.75	13	15	111	207	24.6	766
31	MTP19057	74	29	12	2	10	10	1.75	10	7	158	209	24.6	718
32	MTP19038	66	36	19	3	11	10	1.25	10	7	73	199	24.6	574
33	MTP19070	46	62	13	5	10	8	1	9	7	83	184	24.1	526
34	MTP19027	73	33	19	6	9	11	1.25	12	5	38	226	24.9	502
35	MTP19068	52		10		11	9	1	8	11	141	232	25.1	502
36	MTP19078	63	44	9	4	10	10	1	9	10	115	221	24.6	455
37	MTP19077	59	50	15	4	12	11	1.5	6	4	60	186	24.6	359
38	MTP19044	49		9		8	9	1	9	10	109	195	24.7	287
39	MTP19027	68	36	10	1	9	14	1.5	4	6	160	201	24.8	287
40	MTP19057	42	32	7	1	7	9	1	6	8	140	209	25.0	239
41	MTP19073	54		13		11	9	1.25	8	9	106	240	24.8	239
42	MTP19057	68		11		8	13	1	12	8	77	204	24.8	215
43	MTP19074	48	60	9	26	11	9	1.75	8	10	114	213	24.7	215
44	MTP19096	44	80	12	16	9	11	1	5	10	193	260	24.8	168
45	MTP19018	64		9		9	14	1.5	8	9	119	224	25.1	144
46	MTP19068	59		15		11	14	1.25	8	10	136		24.6	96
47	MTP19074	58		11		13	11	1	7	7	169		25.0	96
48	MTP19058	53		9		8	12	1.25	9	4	71			
49	MTP19091								11	3	57			
50	MTP19097	41.5		10.5	7	8	8	1	7	4	61			
	<b>mean</b>	<b>72.5</b>	<b>56.2</b>	<b>15.4</b>	<b>10.8</b>	<b>10.3</b>	<b>11.8</b>	<b>1.5</b>	<b>9.0</b>	<b>9.0</b>	<b>111.0</b>	<b>189.0</b>	<b>24.6</b>	<b>2047.7</b>
	<b>std</b>	<b>21.7</b>	<b>15.9</b>	<b>5.4</b>	<b>6.6</b>	<b>1.8</b>	<b>2.9</b>	<b>0.4</b>	<b>2.6</b>	<b>2.9</b>	<b>37.4</b>	<b>30.6</b>	<b>0.4</b>	<b>1733.4</b>
	<b>min</b>	<b>41.5</b>	<b>29.0</b>	<b>6.5</b>	<b>1.0</b>	<b>7.0</b>	<b>7.8</b>	<b>1.0</b>	<b>3.8</b>	<b>1.5</b>	<b>38.3</b>	<b>123.0</b>	<b>23.8</b>	<b>95.7</b>
	<b>max</b>	<b>141.8</b>	<b>84.3</b>	<b>28.0</b>	<b>28.5</b>	<b>14.8</b>	<b>20.5</b>	<b>2.5</b>	<b>15.5</b>	<b>15.8</b>	<b>214.4</b>	<b>260.0</b>	<b>25.8</b>	<b>5621.9</b>

<b>cv (%)</b>	<b>30</b>	<b>28</b>	<b>35</b>	<b>61</b>	<b>18</b>	<b>25</b>	<b>27</b>	<b>28</b>	<b>32</b>	<b>34</b>	<b>16</b>	<b>2</b>	<b>80</b>
<b>error_variance</b>	<b>470.4</b>	<b>251.5</b>	<b>28.7</b>	<b>44.2</b>	<b>3.4</b>	<b>8.6</b>	<b>0.2</b>	<b>6.8</b>	<b>8.4</b>	<b>1396.3</b>	<b>936.0</b>	<b>0.2</b>	<b>3004531.2</b>

Table B9. Data summary of breeding lines Havre 2023.

<b>SN</b>	<b>name</b>	<b>vinelength</b>	<b>canopy</b>	<b>standcntsp</b>	<b>thseedwt</b>	<b>protein</b>	<b>yld</b>
1	MTP190751	42	27	25	143	23.9	3913
2	MTP191029	46	33	22	147	24.4	3896
3	MTP190753	48	31	25	149	23.7	3853
4	MTP190821	41	32	22	160	25.1	3839
5	MTP190891	39	32	19	177	24.4	3792
6	MTP190820	47	33	22	151	24.4	3774
7	MTP190272	61	44	23	184	25.8	3729
8	MTP190769	50	38	22	163	22.9	3670
9	MTP191031	42	25	19	165	24.7	3639
10	MTP190446	45	38	13	152	26.4	3571
11	MTP190664	68	63	31	136	24.8	3531
12	MTP190681	44	34	15	201	22.8	3460
13	MTP190752	42	28	23	161	22.8	3444
14	MTP191030	41	32	15	178	24.7	3392
15	MTP190822	36	31	19	148	24.8	3345
16	MTP190596	53	40	25	165	23.9	3341
17	MTP190574	51	39	18	148	24.5	3315
18	MTP190854	59	42	30	116	27.1	3313
19	MTP190767	46	35	16	166	24.9	3183
20	MTP191028	38	30	17	156	26.1	3104
21	MTP190444	50	30	17	161	26.7	3071
22	SPECTER	59	42	25	127	28.0	2989

23	MTP190850	59	46	20	188	21.1	2942
24	LYNX	34	23	9	138	26.6	2879
25	MTP190887	39	32	16	165	24.1	2844
26	MTP190577	48	35	18	179	26.3	2812
27	MTP190678	44	37	17	175	22.0	2734
28	MTP190270	54	38	15	153	23.9	2669
29	MTP190766	39	31	15	172	24.6	2611
30	MTP190742	41	36	12	198	24.9	2530
31	MTP190331	68	37	11	192	27.5	2402
32	MTP190582	40	33	10	175	23.9	2204
33	MTP190570	51	36	12	164	25.8	2191
34	MTP190740	49	32	11	188	26.7	2163
35	MTP190812	45	31	15	179	26.3	2085
36	MTP190701	38	33	5	179	23.0	1850
37	MTP190739	48	34	9	204	26.3	1726
38	MTP190747	38	28	9	163	23.9	1711
39	MTP190774	42	37	8	170	24.1	1679
40	MTP190186	44	30	12	182	27.3	1673
41	MTP190684	45	35	9	179	26.1	1600
42	MTP190853	50	32	9	169	23.0	1320
43	MTP190782	39	38	11	180	23.3	1177
44	MTP190960	34	32	5	198	24.2	1140
45	MTP190918	38	35	12	190	26.5	1051
46	MTP190389	45	28	6	173	25.8	1048
47	MTP190972	33	22	5	181		728
48	MTP190583	35	28	2	191		632
49	MTP190787	38	25	2	176	26.6	484
50	MTP190786	43	29	1	193		201
	<b>mean</b>	<b>45.4</b>	<b>33.9</b>	<b>14.8</b>	<b>168.9</b>	<b>25.9</b>	<b>2564.9</b>
	<b>std</b>	<b>8.3</b>	<b>6.6</b>	<b>7.3</b>	<b>19.5</b>	<b>1.6</b>	<b>1048.9</b>

<b>min</b>	<b>33.0</b>	<b>21.9</b>	<b>0.8</b>	<b>115.6</b>	<b>21.1</b>	<b>200.8</b>
<b>max</b>	<b>68.0</b>	<b>63.0</b>	<b>30.8</b>	<b>203.8</b>	<b>28.0</b>	<b>3913.3</b>
<b>cv (%)</b>	<b>18</b>	<b>20</b>	<b>50</b>	<b>12</b>	<b>6</b>	<b>41</b>

Table B10. Data summary of breeding lines Huntley 2023.

SN	Name	vinelen	cano	pod	podht	flwrno	reprn	podp	standc	standcn	survi	thseed	prote	yld
		gth	py	ht	mat	de	ode	ed	ntfl	tsp	val	wt	in	
1	MTP19103	85	79	42	19	16	9	2	31	7	22	165	23.8	7655
2	MTP19082	84	49	32	14	12	13	2	33	11	33	148	23.8	7129
3	MTP19082	72	58	16	12	10	13	2	29	8	27	176	24.3	6435
4	MTP19066	130	55	38	13	11	15	1	32	11	34	162	23.9	5191
5	MTP19075	74	62	22	12	11	11	2	29	13	47	157	23.0	5191
6	MTP19102	77	59	16	14	10	13	2	29	10	36	159	23.5	5167
7	MTP19076	79	72	16	21	14	12	2	30	11	35		23.8	4880
8	MTP19085	159	51	47	17	13	14	2	36	9	25	114	26.1	4809
9	MTP19075	84	61	35	5	10	13	2	26	8	31	180	23.8	4713
10	SPECTER	143	51	56	9	11	15	2	27	11	40		23.2	4569
11	MTP19082	69	62	26	15	9	11	2	35	11	31	162	24.1	4545
12	MTP19103	68	65	26	10	10	10	2	31	10	34	171	23.4	4163
13	MTP19088	69	83	29	14	11	9	2	33	6	18	164	22.1	3971
14	MTP19075	85	52	37	12	13	15	2	29	10	34	180	23.3	3493
15	MTP19089	92	62	29	20	11	14	2	31	10	31	170	23.4	3301
16	MTP19076	78	52	21	18	10	13	2	26	11	43	184	22.8	3277
17	MTP19033	159	48	64	10	14	12	1	21	8	38	195	24.5	2608
18	LYNX	76	38	23	14	9	12	2	32	8	24	151	25.1	2584
19	MTP19085	56	30	25	5	11	8	1	27	7	24	219	23.6	550
20	MTP19068	64	28	21	8	9	11	2	35	11	30	237	24.8	502

21	MTP19027	78	22	21	7	12	12	2	31	6	18	207	24.4	419
22	MTP19058	72	27	26	6	11	14	2	27	15	55	193	24.2	383
23	MTP19102								27	4	16		23.8	335
24	MTP19074	65	38	24	13	11	11	2	22	11	54		23.6	287
25	MTP19076	44	33	17	7	10	12	2	28	6	20		21.3	263
26	MTP19067	62	27	28	5	9	9	2	28	10	35	195	23.1	191
27	MTP19057	63	21	25	5	10	11	2	30	10	36	177	24.8	144
28	MTP19078	47	14	21	5	8	7	2	30	10	34	195	23.9	144
29	MTP19038	60	21	19	3	8	11	1	25	9	40	184	24.6	132
30	MTP19044	66	17	16	2	9	12	2	30	8	28		24.6	96
31	MTP19074								26	8	33	178	24.7	96
32	MTP19058								28	6	21	164	23.7	72
33	MTP19027	74	15	20	7	12	13	2	28	7	27		24.8	48
34	MTP19057	55	20	19	5	9	9	1	32	12	37		22.5	48
35	MTP19077	66	41	24	10	10	8	2	32	7	21	165	23.1	48
36	MTP19078	51	28	15	2	13	8	1	28	17	59		24.8	24
37	MTP19085	38	15	15	1	10	7	1	29	9	31		24.9	24
38	MTP19018								22	8	35			
39	MTP19044	44	15	4	8	9	7	1	28	8	27			
40	MTP19057	124	27	47	14	12	9	1	23	11	46			
41	MTP19059	79		23		11	14	2	26	9	36			
42	MTP19068	52	18	18	6	8	10	1	28	7	25			
43	MTP19070								26	3	12			
44	MTP19073								17	10	59			
45	MTP19074	50	10	14	2	11	10	2	24	6	26			
46	MTP19078								32	12	38			
47	MTP19081	67	32	26	9	9	11	1	28	10	35			
48	MTP19091								30	9	29			
49	MTP19096								19	4	20			
50	MTP19097								27	7	23			

<b>mean</b>	<b>76.4</b>	<b>39.8</b>	<b>25.9</b>	<b>9.6</b>	<b>10.4</b>	<b>11.0</b>	<b>1.5</b>	<b>28.2</b>	<b>8.9</b>	<b>32.2</b>	<b>175.8</b>	<b>23.9</b>	<b>2364.5</b>
<b>std</b>	<b>28.9</b>	<b>20.1</b>	<b>12.0</b>	<b>5.3</b>	<b>1.8</b>	<b>2.4</b>	<b>0.3</b>	<b>3.9</b>	<b>2.6</b>	<b>10.6</b>	<b>24.1</b>	<b>0.9</b>	<b>2454.9</b>
<b>min</b>	<b>38.0</b>	<b>9.5</b>	<b>4.0</b>	<b>1.0</b>	<b>7.5</b>	<b>6.5</b>	<b>1.0</b>	<b>16.8</b>	<b>3.0</b>	<b>11.8</b>	<b>113.5</b>	<b>21.3</b>	<b>23.9</b>
<b>max</b>	<b>159.3</b>	<b>82.8</b>	<b>64.0</b>	<b>21.0</b>	<b>15.5</b>	<b>15.0</b>	<b>2.0</b>	<b>36.3</b>	<b>16.8</b>	<b>59.3</b>	<b>236.8</b>	<b>26.1</b>	<b>7655.3</b>
<b>cv (%)</b>	<b>38</b>	<b>51</b>	<b>46</b>	<b>55</b>	<b>17</b>	<b>22</b>	<b>22</b>	<b>14</b>	<b>30</b>	<b>33</b>	<b>14</b>	<b>4</b>	<b>95</b>
<b>error_variance</b>	<b>834.0</b>	<b>405.3</b>	<b>143.1</b>	<b>28.0</b>	<b>3.3</b>	<b>5.8</b>	<b>0.1</b>	<b>14.9</b>	<b>6.9</b>	<b>111.6</b>	<b>578.6</b>	<b>0.8</b>	<b>602634.2.6</b>

Table B11. Data summary of breeding lines Moccasin 2023.

SN	name	vinelen	cano	pod	podht	flwrno	reprno	podp	standc	standcn	survi	thseed	prote	yld
		gth	py	ht	mat	de	de	ed	ntfl	tsp	val	wt	in	
1	MTP19075	80	59	35	7	10	7	2	23	11	46	178	23.4	5215
2	LYNX	85	56	38	8	11	5	2	25	16	64	151	24.2	5000
3	MTP19103	78	64	37	11	11	7	2	25	7	27	192	24.2	4545
4	MTP19075	87	58	35	9	11	8	2	20	10	49	163	23.8	4474
5	MTP19082	69	64	30	10	6	9	2	28	11	38	156	23.8	4402
6	MTP19102	86	75	36	9	8	9	2	22	8	38	167	23.9	4091
7	SPECTER	126	61	56	6	11	8	2	17	9	49	138	23.4	3971
8	MTP19075	76	66	34	9	7	6	2	21	11	51	188	22.7	3899
9	MTP19076	84	73	37	12	8	6	2	23	11	46	187	24.0	3852
10	MTP19082	77	72	30	4	9	9	2	22	7	33	176	23.8	3732
11	MTP19076	88	66	30	7	8	9	2	22	12	55	188	22.5	3612
12	MTP19102	68	60	26	7	9	9	2	25	11	44	180	23.9	3612
13	MTP19066	130	67	40	9	10	9	2	20	7	37	158	24.0	3588
14	MTP19082	75	65	28	14	8	8	2	19	14	72	175	23.6	3588
15	MTP19077	59	53	22	5	6	6	1	25	10	42	188	22.4	3230
16	MTP19085	157	54	73	10	13	9	2	21	11	52	117	25.0	3230

17	MTP19038	85	58	38	1	7	8	2	22	9	42	197	23.4	2943
18	MTP19088	92	77	40	12	6	10	2	24	11	46	177	23.1	2919
19	MTP19067	68	48	33	4	8	7	1	28	11	40	199	23.0	2560
20	MTP19044	71	55	18	7	8	7	1	25	9	35	194	23.0	2416
21	MTP19089	83	78	32	13	9	9	2	18	10	58	203	23.0	2344
22	MTP19103	76	66	29	5	11	9	2	23	9	40	186	23.7	2321
23	MTP19044	64	54	29	4	10	7	2	30	11	37	196	23.0	2273
24	MTP19057	68	53	21	6	8	9	2	24	10	39	209	24.1	2201
25	MTP19027	71	49	36	2	10	7	2	26	10	40	243	23.1	2129
26	MTP19058	53	48	25	7	8	6	1	21	8	39	221	23.8	2057
27	MTP19076	69	66	18	8	8	9	2	19	12	60	191	23.3	1986
28	MTP19027	57	39	25	3	9	7	1	23	14	59	227	24.4	1938
29	MTP19074	59	52	23	5	9	7	2	21	14	65	261	23.4	1866
30	MTP19074	58	54	25	5	6	7	2	18	7	36	214	23.0	1794
31	MTP19059	51	46	22	1	8	6	1	22	14	64	215	23.8	1722
32	MTP19078	63	54	26	6	7	8	1	23	13	58	210	23.4	1627
33	MTP19078	61	47	19	2	9	7	1	23	10	46	219	23.7	1340
34	MTP19068	47	43	15	5	8	7	2	24	11	47	242	24.0	1077
35	MTP19085	63	39	23	4	10	8	2	24	15	60	230	23.6	1053
36	MTP19068	54	49	20	4	9	7	1	25	8	32	251	24.2	1005
37	MTP19033	129	35	58	10	10	7	2	17	11	63	192	25.3	981
38	MTP19073		27		2				24	11	46		24.7	789
39	MTP19078	65	46	16	6	7	8	1	23	7	31	243	23.9	789
40	MTP19081	65	37	23	1	10	4	2	18	9	54	217	24.2	718
41	MTP19085	59	33	20	2	10	8	1	23	9	43	251	23.1	670
42	MTP19070	56	33	22	1	9	7	2	18	16	94	230	24.4	478
43	MTP19057	58	34	16	3	8	7	2	19	10	56	218	24.5	431
44	MTP19091		38		8				18	6	31	153	23.9	239
45	MTP19096		15		1				16	9	59	154	23.8	215
46	MTP19074		19						21	6	27	185	24.5	191

47	MTP19018		32						21	5	24			48
48	MTP19057	57	21	21	1	5	8	2	21	5	22	162	24.7	
49	MTP19058								30	6	22			
50	MTP19097								24	7	28			
	<b>mean</b>	<b>75</b>	<b>51</b>	<b>30</b>	<b>6</b>	<b>9</b>	<b>7</b>	<b>2</b>	<b>22</b>	<b>10</b>	<b>46</b>	<b>195</b>	<b>23.7</b>	<b>2322.6</b>
	<b>std</b>	<b>22.9</b>	<b>15.4</b>	<b>11.7</b>	<b>3.6</b>	<b>1.6</b>	<b>1.3</b>	<b>0.3</b>	<b>3.2</b>	<b>2.7</b>	<b>14.3</b>	<b>32.3</b>	<b>0.6</b>	<b>1444.2</b>
	<b>min</b>	<b>47</b>	<b>15</b>	<b>15</b>	<b>1</b>	<b>5</b>	<b>4</b>	<b>1</b>	<b>16</b>	<b>5</b>	<b>22</b>	<b>117</b>	<b>22.4</b>	<b>47.8</b>
	<b>max</b>	<b>157</b>	<b>78</b>	<b>73</b>	<b>14</b>	<b>13</b>	<b>10</b>	<b>2</b>	<b>30</b>	<b>16</b>	<b>94</b>	<b>261</b>	<b>25.3</b>	<b>5215.2</b>
	<b>cv (%)</b>	<b>31</b>	<b>30</b>	<b>39</b>	<b>61</b>	<b>19</b>	<b>17</b>	<b>18</b>	<b>14</b>	<b>28</b>	<b>31</b>	<b>17</b>	<b>3</b>	<b>62</b>
	<b>error_variance</b>	<b>525.4</b>	<b>236.5</b>	<b>136.</b>	<b>12.8</b>	<b>2.7</b>	<b>1.7</b>	<b>0.1</b>	<b>10.3</b>	<b>7.4</b>	<b>203.5</b>	<b>1041.5</b>	<b>0.4</b>	<b>208558</b>
				<b>6</b>										<b>3.8</b>

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