

**ENHANCING YIELD POTENTIAL OF HARD RED WINTER WHEAT
(*TRITICUM AESTIVUM* L.) VIA USE OF IMPROVED SYNTHETIC
BACKCROSSES**

A Dissertation

by

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Submitted to the Office of Graduate and Professional Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

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December 2015

Major Subject: Plant Breeding

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ABSTRACT

Wheat (*Triticum aestivum* L.) is the most-widely cultivated and third most-produced grain crop in the world. Wheat contributes 19% calories and 21% protein of the global population diet. With an astounding increase in this global population, that is projected to reach 9 billion by 2050, demand for wheat is expected to reach 900 million tons by 2050. However, narrow genetic base and continued pressure from abiotic and biotic stresses pose a tough challenge to achieve the expected increase in grain yield.

Research leading to the evolution of synthetic hexaploid wheat (*Triticum durum* x *Aegilops tauschii*) and synthetic derived wheat (SDW) (elite bread wheat X synthetic hexaploid wheat) provided a tremendous opportunity to improve wheat production. Preliminary studies showed that SDW had the potential to increase grain yield due to larger seed size and weight. However, heads per square meter and seeds per head are also major determinants of grain yield. Single seed weight was found to be highly heritable in SDW populations in our previous studies. Therefore, we hypothesized that indirectly selecting for heads per square meter and seeds per head, while maintaining single seed weight, will boost yield further.

Multi location yield trials were conducted in 2013 and 2014 to determine grain yield and its components, morphological traits, resistance to green bug (*Schizaphis graminum*, Rond), leaf rust (*Puccinia triticina*), stripe rust (*Puccinia striiformis* f.sp. *Tritici*), and powdery mildew (*Erysiphe graminis* f. sp. *Tritici*). We estimated quantitative genetic parameters including variance components, heritability, and genetic

gain. In addition, we determined response to direct selection and correlated response to an indirect selection using heads per square meter and seeds per head as the indirect selection components. We further estimated the efficiency of indirect selection.

Multi-location yield trials indicated certain SDW produced higher grain yield than their recurrent parents and common check varieties. Comparison of the top ten yielding SDW lines mean with the mean of recurrent parents showed SDW lines produced 11.7% higher grain yield than recurrent parents. The SDW lines maintained a similar number of seeds per head and heads per square meter as recurrent parents but had 10% higher single seed weight. Also, SDW showed higher levels of leaf and stripe rust, greenbug, and powdery mildew resistance compared to their recurrent parents. There were certain indications to show that some resistance was transmitted from primary synthetics. Genetic analyses, such as the genotypic coefficient of variation, heritability, and genetic gain showed that there is tremendous scope for grain yield improvement by utilizing SDW. Genetic gain results indicated that grain yield can be improved by 15.6% per cycle at 10% selection intensity ($i = 1.76$). The efficiency of indirect selection for yield, using heads per meter square, was only 0.41. Similarly, seeds per head and single seed weight had an efficiency of 0.46 and 0.21, respectively.

These results indicate that SDW contributed some favorable alleles for yield, biotic stress resistance, and abiotic stress tolerance. These results also showed that SDW contributions were advantageous under both rainfed and irrigated conditions, which makes them an invaluable source for increasing genetic diversity and improving performance of Texas A&M AgriLife wheat germplasm.

DEDICATION

I dedicate this dissertation to my Guru Raghavendra Swamy and the three most influential people in my life, Dr. Amir M.H. Ibrahim, Dr. Jackie C. Rudd, and Dr. Srirama Reddy.

ACKNOWLEDGEMENTS

Foremost, I would like to express my sincere gratitude to my advisers, Dr. Amir M.H. Ibrahim and Dr. Jackie C. Rudd for their guidance and mentoring me more like parents than teachers. A special thanks Dr. Ibrahim for his help in compiling the dissertation in an appropriate shape with constructive comments on the results and for his persistent guidance throughout my Ph.D. program. His dedication to students was more than I ever expected and his ability to prepare me for a career in plant breeding was immense. I extend my thanks to Dr. Rudd for his valuable guidance during my Ph.D. career, which helped me to think critically and solve problems in an efficient way.

I thank Dr. Shuyu Liu for his advice during the field and lab work and for his valuable reviews in compiling the dissertation. I also like to express my sincere gratitude to my committee members, Dr. Seth Murray and Dr. Ravi Singh, for their valuable time and guidance during my Ph.D. career.

I am grateful to the financial support from the Monsanto's Beachell-Borlaug International Scholarship, Pathways to Doctoral Studies, Texas A&M AgriLife Research and Soil and Crop Sciences Department.

I own my deepest gratitude to my brother, Dr. Srirama Reddy, and my parents for their never-ending support at both personal and professional levels.

Finally, I would like to acknowledge all the member of Texas A&M Wheat Breeding Team for their help in successfully conducting these experiments.

NOMENCLATURE

CAS	Castroville, TX
CH	Chillicothe, TX
CS	College Station, TX
DYB	Diyarbakir, Turkey
SHW	Synthetic Hexaploid Wheat (Primary synthetics)
SDW	Synthetic Derived Wheat lines
GY	Grain yield
TW	Test weight
HT	Height
HS	Heading score
GG	Genetic gain

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1. INTRODUCTION AND LITERATURE REVIEW

1.1 Introduction

Wheat (*Triticum aestivum* L.) is the most-widely cultivated and third most-produced grain crop (Pradhan et al. 2012). Wheat contributes 19% of the calories and 21% of the protein of the global population diet (Pradhan et al. 2012; FAO 2011a). Wheat usually occupies more than 240 million hectares worldwide annually. This is 1.4 times larger than the total area under rice (*Oryza sativa*) and maize (*Zea Mays* L.) combined (FAO 2011b). Hubert et al. (2010) indicated that the world's population might reach 9 billion by 2050. They projected the demand for all cereal crops to reach about 56% with 26% of this increase coming from wheat alone. This alarming demand for an increase in wheat production has posed a big challenge for crop breeders and agronomists and is keeping them actively searching for novel approaches to increase productivity. In order to meet the growing demand for food, we need to either increase the area under cultivation or increase the total production. Constraints such as the declining arable land area due to urban sprawl and marginalization of cultivated soils, lack of access to irrigation, etc. makes it difficult to achieve a horizontal increase in the production area of field crops. Therefore, vertical increase of yield potential (Reynolds and Borlaug, 2006) and precision agriculture technologies are essential. Improvement in grain yield potential requires a broad genetic diversity for yield and related traits and responsiveness to good crop management practices.

1.2 Wheat origin and domestication

Wheat belongs to the family *Poaceae* (=Gramineae) of Angiosperms and the tribe *Triticeae*. This tribe contains 25 recognized annual and perennial genera, among them *Triticum*, *Aegilops* and *Einkorn* are the most commonly grown (Huang et al., 2002). Depending on number of chromosomal set (ploidy level), cultivated wheat is classified into three groups: diploid ($2n=2x=14$; AA), tetraploid ($2n=4x=28$; AABB) and hexaploid wheat ($2n=6x=42$, AABBDD). Man domesticated diploid wheat, a.k.a., wild Einkorn, about 10,000 years ago in the Karaca Dag Mountains in southeast Turkey (Feuillet et al., 2008). There are two species within wild einkorn, namely, *Triticum monococcum* and *Triticum urartu*. Similarly, *Triticum timopheevii* and *Triticum turgidum* are the two major species within cultivated tetraploid wheat. Overall, there are seven subspecies within tetraploid wheat, namely emmer (*Triticum turgidum* ssp. *dicoccum*), macaroni (*Triticum turgidum* ssp. *durum*), persian (*Triticum turgidum* ssp. *carthlicum*), georgian (*Triticum turgidum* ssp. *paleocolchicum*), polish (*Triticum turgidum* ssp. *plonicum*), and khorassan (*Triticum turgidum* ssp. *turanicum*), and *Triticum timopheevii*. ssp. *timopheevii* (Dubcovsky and Dvorak, 2007). Within hexaploid wheat, *Triticum aestivum* and *Triticum zhukovskyii* are the major cultivated species (Dubcovsky and Dvorak, 2007). The former is further classified into five subspecies, namely bread wheat (*Triticum aestivum* ssp. *aestivum*), dinkel or large spelt (*Triticum aestivum* ssp. *spelta*), club (*Triticum aestivum* ssp. *compactum*), shot (*Triticum aestivum* ssp. *sphaerococcum*), and *Triticum aestivum* ssp. *macha*. (Schuber, 2009).

Triticum aestivum (bread wheat) is the most commonly cultivated species, occupying 90% of the total wheat area (Faris et al., 2002). Bread wheat wide adaptation to different climatic conditions, elevations, stresses and presence of special gluten protein are the reasons behind its large acreage and acceptance within the *Triticum* genus. The origin and domestication of bread wheat occurred nearly 10,000 years ago on the banks of river Tigris and Euphrates in the fertile-crescent region that includes present day Turkey, Syria, Iraq, Iran, and Israel (Schuber, 2009). Convergence of three diploid genomes A, B and D resulted in today's cultivated bread wheat. Each of these genomes has seven chromosomes. These seven chromosomes form seven homologous groups with three closely related genomes (Gupta et al., 2008). Recent studies have shown that *Triticum urartu* is the possible donor of the A genome (Gustafson et al., 2009) in hexaploid wheat. The first polyploidization between A genome donor *Triticum urartu* and currently unknown/known B genome source produced tetraploid wheat *Triticum turgidum* L. ($2n=4x=28$; AABB) (Nevo et al., 2002). Some studies have reported *Aegilops speltoides* to be the closest existing species of the B genome in polyploid wheats (Dvořák and Zhang, 1990). Eventually, a second polyploidization that included natural hybridization and chromosomal doubling between *Triticum turgidum* L. and *Aegilops tauschii* Coss. (syn. *Aegilops squarrosa*; $2n=2x=14$; DD) resulted in present day cultivated Bread wheat (*Triticum aestivum* L.; $2n=6x=42$; genome AABBDD) (Huang et al., 2002) (Figure 1.1).

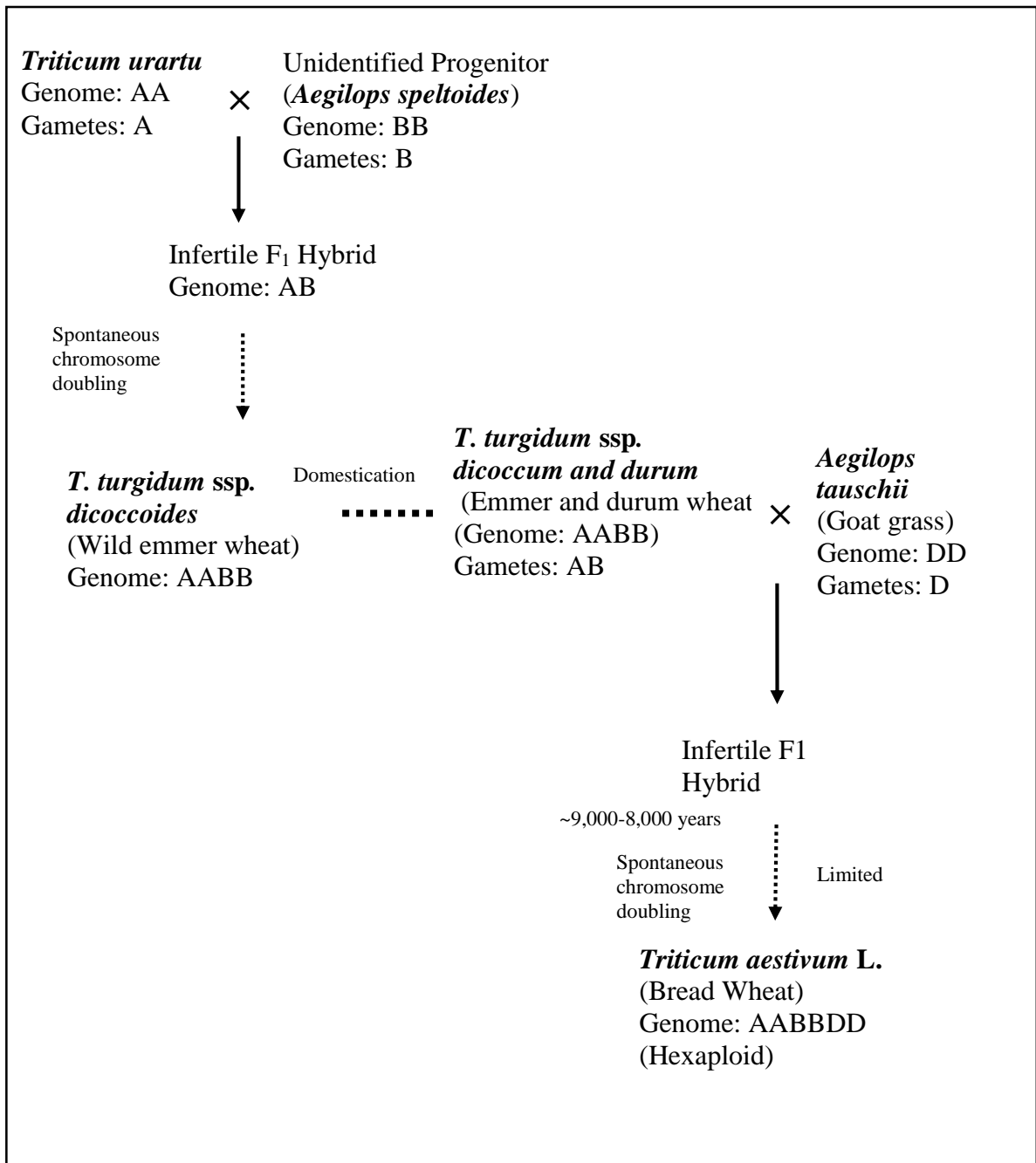


Figure 1.1 Evolutionary diagram of bread wheat (*Triticum aestivum* L.)

1.3 Present status of yield potential in bread wheat

Grain yield improvement is one of the main objectives of U.S. and global wheat breeding programs (Graybosch and Peterson, 2010). Wheat grain yield has increased substantially in the 20th Century due to genetic improvement in the plant ideotypes and tolerance to biotic and abiotic stresses, enhanced agronomic practices, and use of fertilizers, pesticides, fungicides, and herbicides. Many studies have credited one-half of grain yield improvement in the past century to genetic improvement of crops alone (Rudd, 2009; Feyerherm et al., 1984).

Battenfield et al., (2013) reported an average of 0.43 to 1.3% global increase of grain yield per year. Similarly, Rudd (2009) has reported that in the last century grain yield increased at an average of 1% per year. Genetic gain studies in the U.S. Great Plains by Graybosch and Peterson (2010) have shown yield improvement of 1.1 to 1.3% per year (from 1959 to 2008) in the Southern Regional Performance Nursery (SRPN) and 0.79 to 0.85% in the Northern Regional Performance Nursery (NRPN). Based on this study, they concluded that a yield plateau might have been reached in the U.S. Great Plains hard winter wheats. Wheat breeders in the region might be able to make further improvement in yield potential only via the adoption of new breakthrough technologies or introduction of new biological material and diversity, according to this study. In contrast to Graybosch and Peterson, Battenfield et al. (2013) did not point to a yield plateau in U.S. Great Plains hard winter wheat germplasm; however, they indicated that genetic gain for grain yield was low around 0.40% per year. Similar studies conducted by Patrignanai et al. (2014) showed that hard red winter wheat grain yield in the

southern U.S. Great Plains has increased at the rate of 34 kg ha⁻¹ year⁻¹ from 1955 to 1980 and only 6 kg ha⁻¹ year⁻¹ from 1980 to 2012. A number of similar studies pointed to the need for increasing wheat grain yield potential to meet the growing food demand.

Wheat grain yield can be divided into two major categories, grains m⁻² and grain weight (single seed weight) (Slafer et al., 1996). The first Green Revolution resulted from an increase in grains m⁻² attributable to gibberellic acid sensitive dwarfing genes (Rht1 and Rht2). These dwarfing genes not only reduced the plant height but also played a significant role in partitioning the photosynthates to the reproductive tissue; in this case grains m⁻². As a result, semi-dwarf wheats set more grains in the head compared to tall wheats (Miralles et al. 1998; Calderini and Reynolds 2000). While the grains m⁻² component has increased over the time, grain weight remained unchanged or reduced since the beginning of the Green Revolution. Therefore, recent studies have focused on improving genetic gain for single seed weight while maintaining other traits intact. Furthermore, lack of genetic diversity in the bread wheat genome for yield potential and adaptation to biotic and abiotic stresses has also resulted in a lower genetic gain for grain yield. Hence, there is a need for creating new diversity in the bread wheat genome, and it is hypothesized that such diversity can come from wheat relatives and synthetic wheat.

1.4 Importance of genetic diversity in bread wheat

Genetic diversity is essential for plant breeders in developing high yielding, climate-resilient crops (FAO 2011a). Wheat has a tremendous amount of genetic diversity. However, because of 10,000 years of domestication accompanied by recurrent

selection for agronomically superior genotypes we have lost access to more than 69% of genetic diversity present in the wheat genomes (Brubaker, 2013). *Aegilops tauschii* contributed the D genome and *Aegilops speltoides* could be the possible donor of the B genome (Gill et al., 2006). Together, *Aegilops* species contributed two of the three genomes in the present day bread wheat. This has narrowed the genetic base further. All of these factors have collectively contributed to the low genetic gain values in wheat. Gill et al. (2004) have reported wheat grain yield should increase at the rate of 2% per year to meet the growing global food demand. Therefore, addressing the issues of genetic bottleneck present in the bread wheat genome has become the top most priority for wheat breeders.

Wild relatives, landraces, traditional and modern wheat varieties are the important sources for introducing genetic variation. A broad range of novel genetic diversity within the A, B and D genome has persuaded breeders to accept wild relatives as a choice for introducing genetic diversity into the bread wheat genome (Metakovsky et al., 1984). The close genetic proximity of wild relatives *Aegilops tauschii* (D genome) to cultivated bread wheat D genome makes it one of the best choices of a hybridizing source for enriching diversity. Many studies have reported a substantial level of novel genetic variability for abiotic and biotic stresses in wild relatives and landraces of wheat. Oliver et al. (2005) have reported that there is more genetic diversity for disease and insect resistance, endosperm proteins, gliadins and glutenins in *Aegilops tauschii* than in *Triticum aestivum*.

Wild relatives of wheat are good sources for biotic and abiotic stress tolerance in wheat. However, there are no known wild relatives for hexaploid wheat. Therefore, new sources of genetic diversity can be introduced into hexaploid wheat by reproducing hexaploid wheat original cross (*Triticum turgidum* X *Aegilops tauschii*). The hexaploid wheat produced by artificial synthesis is called as synthetic hexaploid wheat (SHW) (Zhang et al., 2008), which represents a promising source for improving qualitative and quantitative traits in present day bread wheat (McFadden and Sears 1944 cited in Feldman and Levy 2005).

The International Maize and Wheat Improvement Center (CIMMYT) in Mexico developed more than 1,100 SHW lines. A number of studies have shown that there is ample amount of genetic diversity for biotic and abiotic stress tolerance in these SHW lines (Mujeeb-Kazi et al. 2000a, 2000b, 2001a, 2001c, 2001b). A number of studies showed that SDW produced higher grain yield than elite varieties (Warburton et al., 2006, Mujeeb-Kazi et al., 1996). This is probably due to improvement in yield components (Calderini and Reynolds, 2000), resistance to insects and diseases (Mujeeb-Kazi et al. 2001c, 2001b, Hartel et al., 2004), and tolerance to abiotic stresses, such as heat, drought and salinity (Trethowan and Mujeeb-Kazi, 2008; Reynolds et al., 2005).

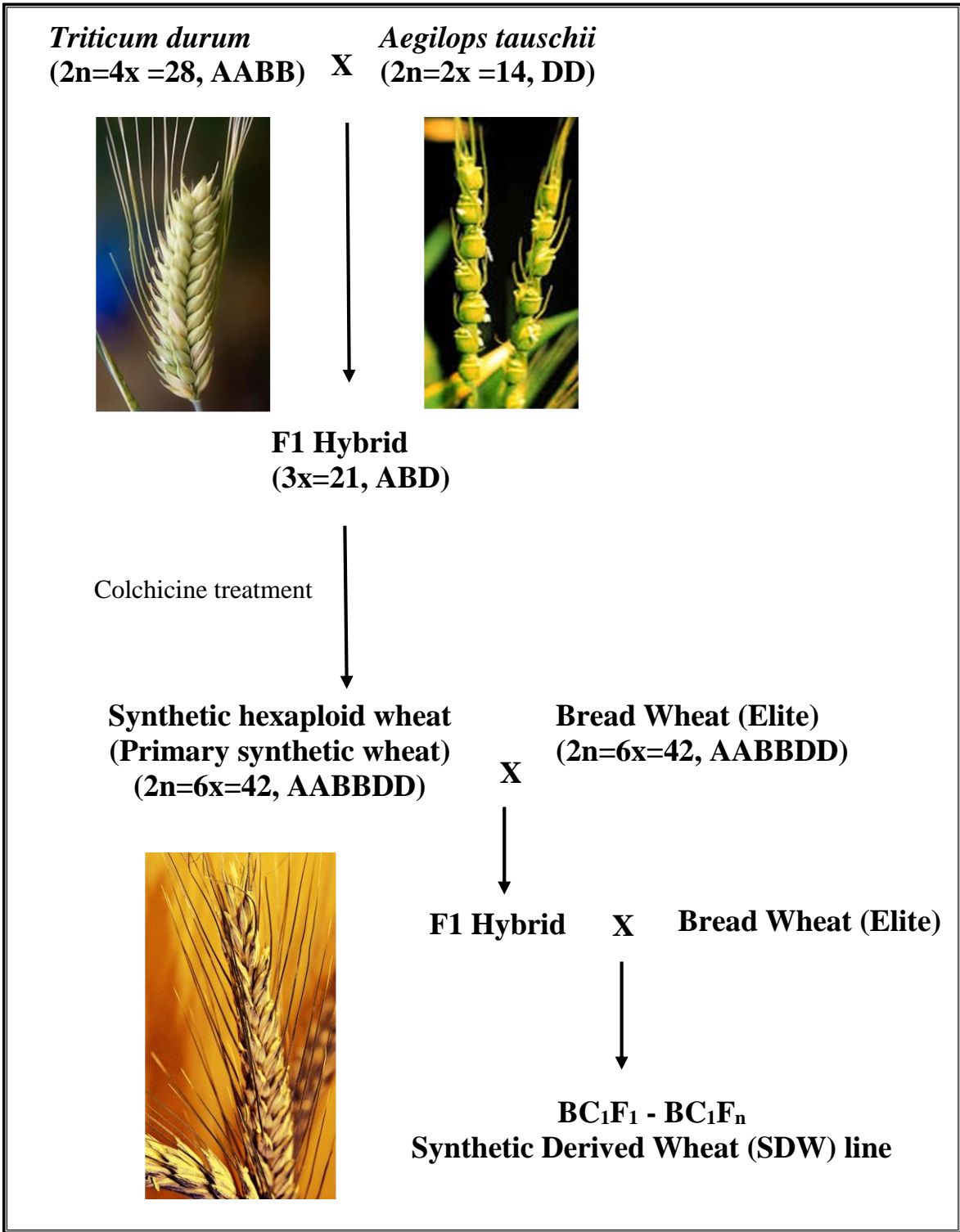


Figure 1.2 Schematic representation of development of primary synthetic wheat and synthetic derived wheat lines

1.5 Definition and history of synthetic wheat

The pioneering work of hybridizing Emmer (*Triticum turgidum* ssp. *dicoccum*) with bread wheat by McFadden in the 1930's resulted in first artificial synthetic wheat. Artificial synthetic wheat had a good level of resistance to stem rust. Eventually, it led to the release of 'Hope' wheat, which saved the U.S. wheat industry when it was succumbing to severe stem rust epidemics. Through this process, McFadden successfully introduced the *Sr2* gene as a source for adult plant resistance, which wheat breeders continue to utilize today. McFadden and Sears (1946) coined the term SHW to represent synthesis of allopolyploid.

Decades later, in 1980's, Mujeeb Kazi and other researchers at CIMMYT started developing synthetic wheat to obtain new genetic diversity for karnal bunt (*Tilletia indica*) and other traits (Mujeeb-Kazi et al., 1996; Warburton et al., 2006). As mentioned earlier, there are two subspecies within *Aegilops tauschii*; *Aegilops tauschii* ssp. *tauschii* and *Aegilops tauschii* ssp. *strangulata* (Hammer, 1980). *Aegilops tauschii* ssp. *strangulata* considered as the direct donor of D genome to common wheat (Pestsova et al., 2000). However, only a few accessions of ssp. *strangulata* believed to be involved in the evolution of present day hexaploid wheat. Therefore, hybridizing either with ssp. *tauschii* or ssp. *strangulata* will contribute a good amount of genetic diversity for bread wheat improvement. There have been efforts to develop SHW by hybridizing with both wild emmer and durum wheat (Niwa et al., 2010, Mujeeb Kazi 2000a). Ultimately, the durum parent was the primary choice as a tetraploid parent because of it's agronomic and free threshing characteristics (Trethowan and van Ginkel, 2009).

In CIMMYT, the primary synthetics (PS) or SHW were developed by hybridizing female *Triticum turgidum* ssp. *durum* (AABB) with male *Aegilops tauschii* (DD) (Mujeeb-Kazi et al., 1996). This hybridization event resulted in sterile triploid embryos, which were rescued and cultivated on culture media until they differentiated into plantlets with roots and shoots. Mujeeb-Kazi et al., (2008) treated these differentiated plantlets with colchicine to induce chromosomal doubling and to facilitate hexaploid seeds set upon self-fertilization (2008) (Figure 1.2).

Ogbonnaya et al. (2013) reported that more than 1500 SHW were produced from 1940 to 2010. Out of these, CIMMYT-Mexico produced around 1300 SHW from 1988 to 2010 using 900 *Aegilops tauschii* accessions. These *Aegilops tauschii* accessions were characterized and grouped according to breeding objectives and were randomly hybridized with tetraploid wheats (Mujeeb-Kazi et al., 2001) as pointed above. Based on agronomic performance and disease resistance, 128 SHW out of the 1300 core collection, grouped into Elite-I and Elite-II categories were selected and distributed globally. The Elite-I had 95 SHW with good agronomic characteristics, biotic and abiotic stress tolerance (Mujeeb-Kazi et al., 2000); whereas the Elite-II had 33 SHW lines with good resistance to leaf rust, stripe rust, stem rust and other diseases (Mujeeb-Kazi and Delgado, 2001). Most of the SHW's in this core collection of 128 SHW were spring types (>1000) with very few winters (186) (Cox et al., 1995). Apart from CIMMYT-Mexico, ICARDA-Syria, CSIRO-Canberra, University of Sydney-Australia, NIAB-United Kingdom, USDA-ARS-USA, and the University of Melbourne-Australia also produced different sets of SHW (Gatford, 2004).

Overall, SHWs are low yielding and have poor agronomic and quality traits (Trethowan and van Ginkel, 2009). Linkage drag and a high degree of F₁ hybrid necrosis are the major constraints in using SHW at a large scale. Therefore, backcrossing these SHW to adapted wheat varieties is essential to reap the maximum benefit. Backcrossing one or two times would introduce desired alleles and traits into the adapted wheat background with minimal linkage drag (Trethowan and van Ginkel, 2009). The hexaploid wheat resulting from backcrossing SHW with adapted wheat varieties is often regarded as SDW (Figure 1.2).

1.6 Impact of synthetic wheat in wheat improvement

In the last two decades, the need and enthusiasm for utilizing SHW in wheat breeding programs has significantly increased. An estimated one-third of the advanced lines in the CIMMYT global wheat-breeding program are synthetic derivatives (Imtiaz et al., 2007). Many novel useful traits and genes from these SHW have been identified and utilized to improve disease and insect resistance, abiotic stress (drought, heat and salinity) tolerance, high biomass, large root system, micronutrient (Fe and Zn) content, and yield and yield components (large kernel size) (Ogbonnaya et al., 2007, Trethowna and Mujeeb-Kazi, 2008). A survey of seven years of data from 2005 to 2012 showed that 17% of all entries in CIMMYT advanced lines and spring bread wheat observation nursery (SBWON) in ICARDA were synthetic derivatives (Ogbonnaya et al., 2013). However, the percentage varied among the nurseries. About 35% of the entries of

CIMMYT's Semiarid Wheat Yield Trials (SAWYT) and 46% of ICARDA's SAWYT in were synthetic derivatives during that period.

In 2006, China released its first documented SDW variety, 'Chuanmai 42'. This variety had higher grain yield and grain weight and better stripe rust resistance than some of the elite varieties in few provinces (Yang et al., 2006, 2009). Chuanmai 42, which was cultivated in more than 100,000 ha, out-yielded all commercial varieties in Sichuan province for two years, including "Chuanmai 107" by 22.7% (Yang et al., 2009). Following the success of Chuanmai 42, China released four more SDW varieties (Yang et al. 2009; Ogonnaya et al., 2013). Similarly, a number of SDW varieties have been released by many other countries around the world. A private company in Uruguay and Argentina has released commercial variety 'NOGAL' and Instituto Nacional de Investigacion Agropecuaria (INIA) in Uruguay has released two more varieties, namely, "Genesis 2354" and "Genesis 2359". Similarly, Spain has released commercial variety 'Carmona' (Ogonnaya et al., 2007).

1.6.1 Disease and insect resistance

As mentioned earlier, bread wheat D genomes came from *Aegilops tauschii* (Gill et al., 2006). *Aegilops* is an important source of disease and insect resistance (Friebe et al., 1991; Gill et al., 2006). Therefore, SHW has shown a broad range of genetic diversity for diseases and insect resistance. A number of studies have documented resistance to leaf rust (LR; *Puccinia triticina* Erikss. & Henn.) (Cox et al., 1997; Assefa and Fehrman, 2000), stripe/yellow rust (YR; *Puccinia striiformis* Westend. f. sp.

Tritici) (Ogbonnaya et al., 2008, Badebo and Ferhmann, 2005), and stem rust (SR; *Puccinia graminis* Pers. f. sp. *Tritici*) (Maraisa et al., 1994). Resistance was also documented for other diseases such as Fusarium head blight (*Fusarium graminearum*) (Mujeeb-Kazi et al. 2001b), Karnal bunt (Mujeeb-Kazi et al. 2001c, 2008), Septoria tritici leaf blotch (*Mycosphaerella graminicola*) (Arraiano et al., 2001), Spot blotch (*Cochliobolus sativus*) (Mujeeb-Kazi et al., 2001b), Tan spot (*Pyrenophora tritici-repentis*) (Tadesse et al., 2007) and Powdery mildew (*Erysiphe graminis* f. sp. *Tritici*) (Hu and Xin, 2001). Similarly, resistance among SHW exists for insect pests such as, Greenbug (*Schizaphis graminum* Rondani) (Weng et al., 2005; Gill et al., 1991), Hessian fly (*Mayetiola destructor*) (Friesen et al., 2008; Yu et al., 2010, 2012), and Cereal Cyst Nematode (CCN; *Heterodera avenae*) (Eastwood et al., 1991). Gill et al., (2006) have documented a number of diseases and insect resistant genes transferred from wild relatives into cultivated wheat.

Among wheat diseases, LR is the most prevalent around the world (Roelfs et al., 1992). In the U.S. and Canada, it causes yield loss of 25 to 95% on susceptible varieties and 10 to 28% on resistant lines (Peturson et al., 1945). Many major LR resistance genes have been deployed in many varieties around the world. However, rapidly evolving races of *Puccinia triticina* Eriks. have developed virulence to many of these genes and defeated resistance. To date, more than 90 LR (standard designations: *Lr1* to *Lr68*) and 89 YR resistance genes (standard designations: *Yr1* to *Yr49*) have been reported (McIntosh et al., 2010). Eight of these LR genes come from *Aegilops tauschii* background. The *Lr40* gene was found to be allelic to *Lr21* (Huang and Gill, 2001) and

Lr41 was found to be allelic to *Lr39* (Singh et al., 2004). The other major *Lr* genes that came from *Aegilops tauschii* background are *Lr32*, *Lr42*, *Lr22* (*Lr22a*), and *Lr21*. Among these, *Lr42* is widely used in U.S. and worldwide for leaf rust resistance in breeding programs. This gene is most common in high yielding rust resistant lines from CIMMYT (Martin et al., 2003). To date, the gene *Yr28* is the only stripe rust resistance gene that comes from the D genome of *Aegilops tauschii* (Singh et al., 2000).

A number of studies have reported the introduction of a new source of resistance from SHW. In the U.S., one of the most promising results of SHW was the introduction of greenbug resistance gene *Gb3* into common wheat. This gene was first identified in ‘Landon’-derived SHW line Largo and was eventually introduced into famous winter wheat varieties such as TAM 110 and TAM 112 (Lu et al., 2012).

Yu et al. (2012) reported that 52 out of 118 CIMMYT SHW lines were resistant or moderately resistant to Hessian fly. As the tetraploid parent in the original cross was a susceptible durum parent, the resistance in these SHW lines should have come from *Aegilops tauschii*. The molecular studies on these resistant lines using PCR-based markers showed resistant genes closely linked to *H13*, *H22*, *H23*, *H26*, and *H32*. However, remaining 19 lines had different haplotypes suggesting these lines might contain new genes for Hessian fly resistance. Eastwood et al. (2006) identified SDW lines with high grain yield potential in environments with high terminal moisture stress and low yield level and these lines came from *Aegilops tauschii* accessions with CCN resistance.

1.6.2 Abiotic stress tolerance

Synthetics have also contributed genes for tolerance to abiotic stresses such as drought, heat, salinity, as well as nutrient use efficiency. Heat and drought are major constraints for wheat production in Texas specifically and the U.S. Great Plains in general. A number of experiments were conducted to study the response of SHW and SDW to drought and heat stresses. These studies showed that SDW lines generally had better tolerance to heat and drought than their recurrent parents did (Yang et al., 2002; Trethowan and Mujeeb-Kazi, 2008; Yang et al., 2009). Some of these studies attributed this enhanced tolerance to heat and drought to greater root biomass in deeper layers and better water extraction capacity (Reynolds et al., 2007). Similarly, Ogonnaya et al. (2013) pointed out that thicker and deeper root systems facilitated better performance of SDW under drought conditions. Studies conducted by Lopez and Reynolds (2011) indicated that early flowering and increased water use efficiency at anthesis improved terminal drought tolerance in SDW. Some wild accessions of *Aegilops* such as *Aegilops tauschii*, *Aegilops speltoides* and *Aegilops geniculata* have shown the capacity to withstand better drought (Zaharieva et al., 2001). Yang et al. (2002) documented some of the SHW and SDW lines showed better tolerance to frequent heat spells than their recurrent parents. Zaharieva et al. (2001) reported that few accessions of *Aegilops* such as *Aegilops geniculata*, *Aegilops speltoides*, *Aegilops searsii* have shown better heat tolerance than common wheat check cultivars. *Aegilops tauschii* has also shown a good level of tolerance to many nutrient deficiencies. Cakmak et al. (1999), for instance, have

documented many accessions of *Aegilops tauschii* that possess tolerance to zinc deficiency.

1.6.3 Enhancing yield potential

A number of studies have shown a high level of genetic variation for yield and its components in SHW (Calderini and Reynolds, 2000; Mujeeb-Kazi and Hettel, 1995). Further studies also demonstrated potential for yield improvement in SDW (Ogbonnaya et al., 2013). Molecular analysis of these SDW lines has indicated improvement in the genetic diversity of bread wheat genome (Dreisigacker et al., 2008). Ogbonnaya et al. (2007) and Dreccer et al. (2007) reported SDW lines produced up to 30% and 11% higher grain yield than famous Australian varieties in northern and southern Australia, respectively.

Studies conducted at ICARDA showed that SDW had 25% higher grain yield than their recurrent parent (Cham-6) under stressed and non-stressed conditions. The best SDW line in this set had 124% and 128% higher grain yield than Cham-6 under stressed and non-stressed conditions, respectively (Ogbonnaya et al., 2013). Mean grain yield in stressed sites was 0.89-1.66 t ha⁻¹ and in non-stressed sites was 2.64 - 6 t ha⁻¹. In China, 'Chuanmai 42' out-yielded commercial cultivar 'Chuanmai 107' by 22.7% (0.45 to 0.75 t ha⁻¹). Preliminary results of BC₁F₅ at the National Institute of Agricultural Botany (NIAB), UK suggested that use of SHW in breeding programs enhanced yield potential. These results showed that the best SDW lines had 113 and 119% higher grain yield than their recurrent parents and elite control lines, respectively (Ogbonnaya et al.,

2013). Based on studies conducted in southern Queensland, Australia Christopher et al. (2006) reported that SDW had better stay-green phenotype and out-yielded commercial varieties 'Hartog', 'Banks', and 'Baxter' by 55%, 31% and 15%, respectively. The yield of these SDW lines was 12% higher than that of CIMMYT stay-green variety Seri-M82. Under terminal water stress conditions, SDW lines yielded 25% higher grain yield than their recurrent parents in Mexico yield trials (Lopez and Reynolds, 2011). Many studies were conducted to elucidate this increase in grain yield of SDW. For instance, at CIMMYT-Mexico, Dreccer et al. (2006) reported that increased yield was associated with an increase in water use efficiency and root length density. However, these results were not consistent with studies conducted in Victoria, Australia. Rattey et al. (2008) attributed this increase in grain yield of SDW to greater grain size under both high and low yielding conditions. Similarly, Yang et al. (2009) attributed this increase to larger kernels and better resistance to stripe rust. Reynolds (2007) and Lopez and Reynolds (2011) accredited it to high root biomass and better water extraction capacity, especially in places where wheat is grown on stored soil moisture. They also reported that under terminal water stress conditions, early flowering and increased water use efficiency at anthesis were critical for this increase.

1.7 Impact of genotype-by-environment (G*E) interaction on grain yield and yield components in synthetic wheat

Grain yield is a poorly heritable and complex quantitative trait. Environment and GE interaction play a significant role in determining overall grain yield in wheat (Wu et

al., 2012). Grain yield and stability of a genotype play a decisive role in determining the release of a variety (Zafarnaderi et al., 2013). The literature is full of studies that worked on elucidating the relationship between yield and its components (single seed weight, heads per square meter, and seeds per head) in bread wheat and SDW (Cooper et al., 2013). A number of these studies have shown that synthetics increased grain yield under both low and high-yielding environmental conditions. A larger advantage was witnessed in low-yielding (185 gm^{-2}) than high-yielding environments (429 gm^{-2}) (Rathey et al., 2010, 2011; Gororo et al., 2002). Yield components such as increased seed size and single seed weight (Gororo et al., 2002, Cooper et al., 2012, 2013), and heads per square meter (Dreccer et al., 2008) were accredited for this increase. Similarly, physiological and morphological traits such as cooler canopies (Reynolds et al., 2007), improved water extraction characteristics with an increased partitioning of root biomass to deeper layers (Reynolds et al., 2007), and increased biomass at maturity (Reynolds et al., 2007) were accredited for improved performance of SDW. Along with physiological, morphological and yield components, improved resistance to biotic stresses and tolerance to abiotic stresses were accredited for this increase (Lopes and Reynolds, 2011; Ogbonnaya et al., 2007; Trethowan et al., 2005; Cooper et al., 2012; Narasimhamoorthy et al., 2006). In Australia, SDW lines yielded 8-30% higher grain yield than adapted varieties. The average rainfall in these areas ranges from 275-700 mm (Ogbonnaya et al., 2007). Rathey et al. (2011) reported that SDW lines had 16-18% higher single seed weight than broadly adopted Australian lines and some studies recorded single seed weight as high as 67 mg when tested in CIMMYT, Mexico (Calderini and Reynolds, 2000)

1.8 Indirect selection for grain yield in wheat

Indirect selection for a trait is practiced when selecting for the primary trait is less efficient than a secondary one that is highly correlated with the primary trait. For instance, selecting for primary traits can be very expensive and difficult especially when the traits are expressed late in the plant life cycle. Gallais (1984) has reported that indirect selection for a secondary trait is never as effective as direct selection for the primary trait, unless the former has high narrow-sense heritability and the additive genetic correlations between two traits is high. Under stress conditions, indirect selection for yield is very effective even if the heritability of primary and secondary traits are almost equal (Hill et al., 1999). Greater effectiveness was related to the high genetic correlation between two traits and high narrow sense heritability of the secondary trait.

A number of studies were conducted to study the relationship between yield and its components. These studies suggested that yield components seeds per head, heads per square meter, and single seed weight have significant impact on overall grain yield (Cooper et al., 2012, Gorjanovic and Balalic, 2006).

Savil and Nedelea (2012) studied the impact of spike length, spikelet number, seed per head, seed weight per head, and plant height on yield. Based on these studies, they reported that indirect selection for seed weight per head and plant height were the most efficient methods for increasing grain yield. Seed weight per head (71.62%) had the highest influence on grain yield followed by plant height (14%) and spikelet number (7%) in their study. Bahadur and Lodhi (1995) indicated that indirect selection for seeds per head might increase grain yield. Some other studies argued that single seed weight

and seed per head as the most efficient indirect selection yield components (McNeal et al., 1978). Austin (1994) reported selection for heads per unit area and seeds per head were the main contributors to yield improvement. Iftikhar et al. (2012) reported that single seed weight had the highest positive direct effect on yield (0.970). This suggests that single seed weight might be a suitable selection criterion for developing high yielding wheat genotypes for rainfed areas. The rationale for this assumption is plant available soil moisture during grain filling period plays an important role in determining single seed weight and hence the overall grain yield. They also reported positive indirect effect between yield and plant height (0.153), peduncle length (0.066) and head length (0.104), grain per head (0.137) and indirect negative effect via days to heading (-0.212) and tiller per plant (-0.135).

1.9 Correlations and path-coefficient analysis

Correlation coefficients and path-coefficients help to determine the interrelationship between yield and its components. Correlation coefficients only determine the interrelationships among the different traits. However, path-coefficient is a standardized partial regression coefficient that suggests direct and indirect relationship among variables and also partitions correlations coefficients into direct and indirect components (Dewey and Lu, 1959). Therefore, correlations and path-coefficients can be great resources for indirectly selecting for a complex trait (Pordel and Maragheh, 2013).

Path-coefficient analysis for yield and its components such as heads per square meter, seeds per head, and single seed weight, can determine how each component

influences yield as well as other components. Based on studies conducted under field conditions in India, Khan et al. (1999) found a high correlation between grain yield and single seed weight, seeds per head, and harvest index. Similar to previous studies, Mondal et al. (1997) reported tillers per plant, seeds per head, and single seed weight had a direct positive effect on grain yield. Gupta and Chaturvedi (1995) reported that plant height and days to maturity had direct negative effects on grain yield. Some durum wheat studies conducted in Egypt reported that tillers per plant had the highest direct effect on grain yield (Bakhit et al., 1989). Dakioku and Akaya (1999) reported that heads per square meter and seed weight per head had a direct effect on grain yield and seeds per head had a positive indirect effect on grain yield through grain weight. Under terminal drought stress conditions, seeds per head, single seed weight, and total biomass had direct positive effects on grain yield (Mollasadeghi et al., 2011). Similarly, Zakaizadesh et al. (2010) have reported that heads per square meter, seed weight per head, and total biological yield had direct effects on grain yield. Under heat stress conditions, heads per square meter had a highest positive direct effect on grain yield in their study. Path-coefficient studies conducted by Pordel and Maragheh (2013) reported peduncle length had a highest direct effect on grain yield and infertile tillers had an indirect effect on grain yield.

Based on studies conducted at multiple locations in Texas, Cooper et al. (2012) reported heads per unit area, seeds per head, and single seed weight had a direct positive effect on grain yield. Among these components, heads per unit area had the greatest effect on grain yield followed by seeds per head. Seed weight consistently remained less

affected by indirect effects from other yield components. Based on studies conducted on SDW lines, Mohsin et al. (2009) reported seeds per head and head length had a positive direct effect on grain yield.

In the U.S. Great Plains, the wheat crop faces the dual menace of biotic and abiotic stresses, these stresses cumulatively contributing to lower yields. In order to increase grain yields under these circumstances, a breeder need to have access to good genetic diversity, have better understanding of the germplasm he has, and genetic relatedness among breeding materials. In an effort to increase the available genetic diversity in the Texas A&M wheat breeding program, breeders at Texas A&M University hybridized and backcrossed synthetic hexaploid wheat with two of their elite wheat cultivars. Thus, resulting populations are called synthetic derived wheat (SDW) populations. These SDW populations believed to have novel sources of alleles for yield, biotic stress resistance, and abiotic stress tolerance. The following studies, which emphasized on characterizing these SDW populations for grain yield, biotic stress resistance, and abiotic stress tolerance will help us to address some of the common challenges

2. GENOTYPE-BY-ENVIRONMENT INTERACTION IN SYNTHETIC DERIVED HARD RED WINTER WHEAT LINES

2.1 Introduction

Wheat (*Triticum aestivum* L.) is the most widely cultivated and third most produced grain crop (Pradhan et al. 2012). Wheat contributes 19% of the calories and 21% of the protein of the global population diet (Pradhan et al. 2012; FAO 2011a). Wheat usually occupies more than 240 million hectares worldwide annually. This is 1.4 times larger than the total area under rice (*Oryza sativa*) and maize (*Zea Mays* L.) combined (FAO 2011b). Due to its wide global distribution, wheat is subjected to many biological and environmental challenges. However, because of domestication and repeated selection by nature and human, we have lost accesses to more than 69% of the genetic diversity that is actually present in the wheat genome (Brubaker, 2013). Globally wheat grain yield has been increasing at an annual rate of 1%. However, wheat workers need to double this rate to meet the needs of a growing population (Gill et al., 2004). A narrow genetic base and continued pressure from abiotic and biotic stresses pose a tough challenge to achieve the needed gain in wheat grain yield.

New approaches and technologies, including broadening the genetic diversity for grain yield and biotic and abiotic stresses tolerance, are highly needed. Synthetic hexaploid wheat, developed by interspecific hybridization between durum wheat (*Triticum turgidum* L.) and accessions of wild goat grass (*Aegilops tauschii* L.) is one of the most efficient methods to introduce lost genetic diversity into the bread wheat

genome. The primary synthetics (PS), or synthetic hexaploid wheat (SHW) per se, have a very poor agronomic performance and weak end-use quality attributes. Therefore, SHWs are repeatedly backcrossed to elite common wheat genotypes. The developed lines from this repeated backcrossing are called synthetic derived wheat (SDW) lines and they have a better agronomic performance and usually better end-use quality attributes. Many studies have shown that SDW contributed to increasing grain yield in spring wheat backgrounds (Ogbonnaya et al., 2013). However, only a few studies have been conducted to study contributions to winter wheat backgrounds. The objectives of this study were to 1) study the genetic variability of yield and its components in SDW lines and to identify superior genotypes for advanced yield trials and 2) study the extent of genotype-by-environment interaction (GE) in SDW lines.

2.2 Materials and methods

2.2.1 Germplasm

The SDW lines used in this study were developed by backcrossing selected CIMMYT (International Center for Maize and Wheat Improvement) SHW from Elite-I (8 SHW) and Elite-II (2 SHW) sets to Texas A&M AgriLife Research hard red winter wheat varieties, ‘TAM 111’ and ‘TAM 112’ (Tables 2.1). Elite I set consists of 95 primary synthetics with better morphological characteristics and abiotic and biotic stress tolerance (Mujeeb-Kazi et al., 2000). Similarly, Elite II set consists of 33 selected primary synthetics that had better resistance to biotic stresses such as leaf rust, stripe rust, stem rust and other common wheat diseases (Mujeeb Kazi and Delgado, 2001a).

Breeders at Texas A&M AgriLife Research had developed 40 SDW populations by hybridizing and backcrossing ten primary synthetics with TAM 111 and TAM 112 and evaluated them across various locations in Texas. Based on agronomic, morphological, and biotic stress tolerance, heads were carefully chosen from selected SDW populations and advanced to head-rows ($BC_1 F_{5:6}$) and later generations. Data from $BC_1 F_{5:8}$ and $BC_1 F_{5:9}$ is presented in this chapter.

2.2.2 Experimental design

In 2011, head-rows ($BC_1 F_{5:6}$) were planted in Chillicothe (CH), TX (latitude = 34.2°N, longitude = 99.5°W) under rainfed conditions. Based on heads per square meter, seeds per head and other agronomic traits, 213 lines were advanced to the $BC_1 F_{5:7}$ generation. These 213 lines were laid out in a randomized complete block design (RCBD) with two replications of 1 meter (m) rows at Bushland (BD), TX (latitude = 35.2°N, longitude = 102.1°W) and Castroville (CAS), TX (latitude = 29.35°N, longitude = 98.88°W). In 2013, the same set of 213 lines along with the check varieties TAM 111, TAM 112, and ‘TAM 401’ were planted as yield plots ($BC_1 F_{5:8}$) at College Station (CS), TX (latitude = 30.5°N, longitude = 96.4°W) under irrigated conditions. This trial was laid out in an augmented design with TAM 112 as the repeated check and TAM 111 and TAM 401 as random checks. We determined yield and its components using plot yield and 50 heads.

Table 2.1 Primary Synthetic wheat and their associated *Aegilops tauschii* accession, synthetic derived wheat populations, pedigrees and number of lines that were evaluated in each of these families in field trials in 2013 and 2014

Pop ID	<i>Aegilops tauschii</i> accession	Name (Synthetic hexaploid wheat)	Name (Synthetic derived wheat)	Pedigree (Synthetic derived wheat)	Selection history	No. of lines
1	WX198	CIMMYT E95Syn4152-5	X05VSBC01	TAM 111*2/CIMMYT E95Syn4152-5	BC ₁ F ₅	11
2	WX198	CIMMYT E95Syn4152-5	X05VSBC49	TAM 112*2/CIMMYT E95Syn4152-5	BC ₁ F ₅	8
3	WX219	CIMMYT E95Syn4152-16	X05VSBC07	TAM 111*2/CIMMYT E95Syn4152-16	BC ₁ F ₅	8
4	WX219	CIMMYT E95Syn4152-16	X05VSBC51	TAM 112*2/CIMMYT E95Syn4152-16	BC ₁ F ₅	4
5	WX629	CIMMYT E95Syn4152-37	X05VSBC17	TAM 111*2/CIMMYT E95Syn4152-37	BC ₁ F ₅	5
6	WX629	CIMMYT E95Syn4152-37	X05VSBC57	TAM 112*2/CIMMYT E95Syn4152-37	BC ₁ F ₅	13
7	WX408	CIMMYT E95Syn4152-61	X05VSBC31	TAM 111*2/CIMMYT E95Syn4152-61	BC ₁ F ₅	11
8	WX408	CIMMYT E95Syn4152-61	X05VSBC60	TAM 112*2/CIMMYT E95Syn4152-61	BC ₁ F ₅	1
9	WX314	CIMMYT E95Syn4152-78	X05VSBC35	TAM 111*2/CIMMYT E95Syn4152-78	BC ₁ F ₅	5
10	WX314	CIMMYT E95Syn4152-78	X05VSBC65	TAM 112*2/CIMMYT E95Syn4152-78	BC ₁ F ₅	9
11	WX417	CIMMYT E2Syn4153-31	X05VSBC46	TAM 111*2/CIMMYT E2Syn4153-31	BC ₁ F ₅	6
12	.	CIMMYT E95Syn4152-51	X05VSBC24	TAM 111*2/CIMMYT E95Syn4152-51	BC ₁ F ₅	12

Based on heads per square meter, seeds per head, overall grain yield, and biotic stress tolerance, 93 lines were advanced to the BC₁F_{5;9} generation (2014), which were laid out in an alpha lattice design with two replications at CAS, CH, and CS and Diyarbakir (DYB), Turkey (latitude = 38.1422 °N, longitude = 40.2711 °E). Wheat varieties ‘TAMW101’, TAM 111, TAM 112, ‘TAM 113’, ‘TAM 304’, ‘TAM 305’, and TAM 401 were planted as checks along the SDW lines in 2014 yield trials. Based on location and year, each trial was given a unique name. Yield trials conducted in 2013 were named as CS2013 and those conducted in 2014 were named as CAS2014, CH2014, CS2014, and DYB2014 respective of location. Trials in CAS2014 and CS2014 were planted at a seed rate of 1582 seeds plot⁻¹ in a plot of 5.11 m² area (3.35 m X 1.52 m). In the CH2014 trial, yield plots were planted with a similar seedling rate equivalent to 340 seeds m⁻². Each yield plot in CH2014 trial measured a total area of 4.64 m² (3.04 m X 1.52 m) after removing alley ways. In the DYB2014 trial, plots were planted at a seeding rate of 350 seeds m⁻² in an area of 3 m² (3.04 m X 0.91 m). In 2014, except CAS2014, all trials were planted under rainfed conditions. Temperature and rainfall data for these locations are reported in Table 2.2. Plots in CAS2014 were supplemented with 10 cm of irrigation using a linear irrigation system. Depending on previous season cropping pattern and irrigation method, nutrient supply was determined for each environment. Trials in each location were fertilized at standard rates for optimum crop production in the location. In CH2014, plots were supplemented with N at the rate of 30 kg ac⁻¹. Similarly, trials in CS2014 and CAS2014 were supplemented with N at the rate of 18.2 kg ac⁻¹ and 49 kg ac⁻¹, respectively. Plots in DYB2014 were supplemented with nutrients at two stages. At

the time of planting, nutrients were supplemented with 20N-20P fertilizer at the rate of 30 kg ac⁻¹. During tillering, plots were supplemented with 18 kg ac⁻¹ of ammonium nitrate.

2.2.3 Environments

Trials at DYB2014, CS2014 and CH2014 were grown under rainfed and the one at CAS2014 was grown under irrigated conditions. The growing season (Oct 31st, 2013 – June 2nd, 2014) precipitation for CH2014 was 148.8 mm, for CS2014 (Nov 25th, 2013- May 26th, 2014) was 319.2 mm, and for DYB2014 (Jan 7th, 2014 - June 24th, 2014) was 163 mm (Table 2.2) (weatherunderground.com).

2.2.4 Measurements

2.2.4.1 Pre-harvest measurements

Observations on agronomic traits, such as heading date (HS), plant height (HT), lodging score, and agronomic score (Agscore), were recorded during the cropping season. Feekes scale was used to determine the heading date (Large, 2007). The heading date was recorded when plants reached Feekes scale 10.1 and 50% of the heads in the plot had emerged from the boots. The number of Julian days each genotype took to reach 50% heading from Jan 1st was recorded as approximate heading date. Heading dates changed from location to location depending on climatic pattern and number of days taken to accumulate certain heat units to reach the heading stage. To be able to

Table 2.2 Maximum (Max), mean (Ave), and minimum (Min) temperature (Temp) and precipitation (rain) data for every month during cropping season in each environment

Month	CH2014				CS2014				CAS2014				DYB2014			
	Rain (mm)		Temp (° C)		Rain (mm)		Temp (° C)		Rain (mm)		Temp (° C)		Rain (mm)		Temp (° C)	
	Total	Max	Ave	Min	Total	Max	Ave	Min	Total	Max	Ave	Min	Total	Max	Ave	Min
Nov	8.6	17.2	10.6	3.3	34.0	11.7	6.7	1.7	15.5	18.9	13.3	7.8
Dec	25.9	10.6	3.9	-2.8	19.3	15.0	9.4	3.9	9.9	16.1	10.0	3.9
Jan	0.0	13.3	5.6	-2.8	33.0	15.6	8.9	2.2	1.3	16.7	8.9	0.6	25.4	10.0	4.4	-1.7
Feb	3.0	11.1	5.0	-1.7	22.6	16.7	11.7	5.6	4.8	19.4	12.8	5.6	18.5	13.3	5.6	-2.2
Mar	19.8	18.9	10.6	2.8	40.9	20.0	13.9	7.8	18.8	22.2	15.0	8.3	37.8	16.7	10.6	4.4
Apr	24.4	26.1	17.8	10.0	31.2	25.6	20.0	14.4	22.1	28.3	21.1	13.9	31.5	21.7	13.9	6.7
May	63.0	31.1	23.3	15.6	138.2	28.9	22.8	16.7	8.6	31.7	23.3	14.4	38.4	27.8	19.4	10.6
	10.2	32.8	24.4	16.1

Codes for environments are CH2014 = Chillicothe, CS2014 = College Station, CAS2014 = Castroville, DYB2014 = Diyarbakir

compare the heading at different locations, we generated new scoring pattern called heading score. In this new format, depending on the location, we classified heading dates into a scale of one to five, with one being very early, two being early, three medium, four late, and five very late. Heading dates of TAM 112, TAM 111, and TAM 401 were considered as standards for early, early-medium, and medium heading, respectively.

Measurements for HT were done when plants reached physiological maturity (loss of the green color in the last internode below the head). Measurements were recorded on representative plants in the center of the plot by measuring height from the soil surface to the tip of the head, excluding awns.

Straw strength, a.k.a. lodging-resistance score was estimated using a scale of one to five, with one being erect and five completely lodged. The Agscore was documented using a scale of one to nine, with one being the poorest and nine being the best looking genotypes, overall.

2.2.4.2 Harvest and post-harvest measurements

A representative sample of 50 heads was collected from each yield plot. These heads were re-counted and bulk threshed, using an Almaco belt thresher model BT14, and weighed. Apart from the 50 heads sample, yield plots were bulk harvested using a Winterstieger combine. Total harvested grain weight (TGW) was determined by adding 50 head sample weight (SW) and yield plot weight (YW). Grain yield (GY; t ha⁻¹) per plot was calculated by entering the TGW weight in the formula 2.1

Formula 2.1

$$\text{Grain Yield (t ha}^{-1}\text{)} = \left\{ \frac{\left[\left(\frac{\text{TGW}}{0.454} \right) \left(\frac{43560}{\text{total area of plot in ft}^2} \right) \right]}{60} \right\} \times 0.0673$$

Yield component such as single seed weight (SeedWt), seeds per head (SeedsHead⁻¹) and head per square meter (HeadNo; heads m⁻²) were estimated based on the 50 head samples. One thousand seeds from the 50 head samples were counted, using a Data Count S-25 Plus seed counter, and thousand seed weight was recorded. Based on thousand seed weighed and TGW we estimated SeedWt, SeedsHead⁻¹, and HeadNo using the formulas 2.2 to 2.4. Test weights were also determined.

Formula 2.2

$$\text{Single seed weight (SeedWt)} = \left(\frac{\text{Thousand seed weight}}{1000} \right)$$

Formula 2.3

$$\text{Seeds per head (SeedsHead}^{-1}\text{)} = \left[\frac{\left(\frac{\text{SW}}{\text{SeedWt}} \right)}{50} \right]$$

Formula 2.4

$$\text{Heads per square meter (HeadNo)} = \left\{ \frac{\left[\frac{\left(\frac{\text{TGW}}{\text{SeedWt}} \right)}{\text{total plot area in m}^2} \right]}{\text{SeedsHead}^{-1}} \right\}$$

Where, SW= 50 head sample weight, TGW = total harvested grain weight.

2.2.5 Statistical analysis

Statistical analysis was performed using SAS 9.4 (SAS Institute Inc., 2008), SAS based META, AgroBase Gen II and Microsoft Excel. Individual and combined analyses for alpha lattice experiments were performed using PROC GLM procedure. Data from CS2013 was analyzed in AgroBase Gen II using moving mean analysis. Analysis for covariance (ANCOVA) was conducted to adjust for some of the post-planting errors in the field. Plots in the CH2014 were affected by unevenly distributed root rot. Uneven plant stand was observed in the DYB2014 yield trials. Heavy rains after physiological maturity resulted in plant lodging in the CS2014 trial. Stability analysis was conducted using AgroBase Gen II.

2.3 Results and discussion

Trials from CAS2014, CH2014, DYB2014, and CS2014 were analyzed for homogeneity of error variance and normality using Leven's and Shapiro-Wilkins's tests, respectively (Grassini et al., 2013). Leven's test showed there was no significant difference between the error variances and hence assumption of homogeneity of error variance was fulfilled for combined analysis. Shapiro-Wilkin test showed that the (W) statistic was close to unity; however, the p-value ($P < W$) was significantly different at 0.05 significance level. This suggests that errors were not normally distributed. Violation of the normality assumption should not cause major problems with a population size of

more than 30 or 40 lines (Ghasemi and Zahediasl, 2012; Pallant, 2007) and the sampling distribution tended to be normal regardless the shape of the data (Field, 2009; Elliott, 2007). Therefore, the assumption of normality was ignored in these analyses.

Combined analysis of variance (ANOVA) across four environments showed that there was a significant difference among the genotypes for GY, TW, SeedWt, SeedsHead⁻¹, HeadNo, HT, and HS (Tables 2.3). Significant differences for the aforementioned traits were attributed to the differences in the genotypes and the way they responded to each environment. We also observed there were highly significant differences in the environments and genotype-by-environment interaction (G*E) for all the aforementioned traits.

Environment (32%) formed the major portion of the total variation, followed by G*E (6.5%), and genotype (2.2%) (Table 2.3). The large sum of squares for environments indicates that environments in which trials were conducted were diverse and large differences among their means might have contributed to differences for these traits. As the trials were conducted in sub-tropical to semi-arid climatic locations under extreme drought to optimum irrigation conditions, a large difference was expected. The presence of significant G*E interactions illustrates that genotypes performed differently across the test environments. Kang and Pham (1991) indicated that presence of significant G*E interaction affects the selection of genotypes.

The ANOVA for GY, TW, SeedWt, SeedsHead⁻¹, HeadNo, HT, and HS was also performed for individual environments. The ANOVA for GY showed significant differences among the genotypes at CH2014 and DYB2014 but not at CAS2014 and

CAS2014 (Table 2.4, Table 2.5, Table 2.6, and Table 2.7). Yield trials in CAS2014 were planted under irrigated conditions with severe and unevenly distributed leaf and stripe rusts. Heavy rains around physiological maturity caused lodging in CS2014. Furthermore, plots at CAS2014 and CS2014 had high CV%. These factors combined might have made it difficult to detect differences among genotypes in the CAS2014 and CS2014 environments.

The ANOVA indicated significant differences among the genotypes for TW, SeedWt, SeedsHead⁻¹, HT, and HS traits at all four individual environments. Significant differences were observed among genotypes for HeadNo at CH2014 and DYB2014 but not at CS2014.

Table 2.3 Mean square values of GY (grain yield), TW (test weight), SeedWt (single seed weight), SeedsHead⁻¹, HeadNo (heads m⁻²), HT (plant height) and HS (heading score) for alpha lattice combined analysis of variance (ANOVA)

Source	df	Mean Squares						
		GY	TW	SeedWt 10 ⁵	SeedsHead ⁻¹	HeadNo	HT	HS
Environment (E)	3	277.13*	5260.24**	65.25**	5495.49**	1854963.89**	4436.23**	2.86**
Genotype (G)	99	0.59*	8.54**	5.42**	77.41**	5653.44**	169.25**	3.07**
Rep	1	0.0017	33.66**	6.12**	156.13**	46204.97**	89.65*	.
Block(Rep)	18	0.88*	1.87	1.42**	36.91*	5858.49	.	0.17
G*E	297	0.58**	6.03**	1.42**	30.65**	5533.41**	65.01*	0.79*
Error	271-376	0.44	1.31	0.53	20.37	3877.17	20.07	0.20

*, ** Significant at 0.05 and 0.01, respectively

G*E = Genotype by environment interaction, Rep = Replication, Environment = Location

Table 2.4 Mean squares values of GY (grain yield), TW (test weight), SeedWt (single seed weight), SeedsHead⁻¹, HeadNo (heads m⁻²), HT (plant height) and HS (heading score) for alpha lattice trial in Castroville, TX-2014

Source	df	Mean Squares						
		GY	TW	SeedWt 10 ⁵	SeedsHead ⁻¹	HeadNo	HT	HS
Genotype	99	0.64	8.27**	2.84**	41.13**	4894.37	74.83**	1.84**
Rep	1	0.37	5.88*	0.18	1.81	5907.85	0.01	3.65**
Block(Rep)	18	0.53	1.07	0.40	16.89	5084.57	2.47	0.19
Error	79-81	0.73	0.89	0.34	12.31	5436.35	1.7	0.19

*, ** Significant at 0.05 and 0.01, respectively
Rep = Replication

Table 2.5 Mean squares values of GY (grain yield), TW (test weight), SeedWt (single seed weight), SeedsHead⁻¹, HeadNo (heads m⁻²), HT (plant height) and HS (heading score) for alpha lattice trial in Chillicothe, TX-2014

Source	df	Mean Squares						
		GY	TW	SeedWt 10 ⁵	SeedsHead ⁻¹	HeadNo	HT	HS
Genotype	99	0.13**	8.07**	1.03**	19.50**	3470.54**	36.40**	0.42**
Rep	1	12.68**	156.42**	2.14*	236.72**	282817.22**	10.96	0.1
Block(Rep)	18	0.08**	4.23**	0.85**	45.86**	2293.44	40.67**	0.14*
Covariate	1	0.82**	2.58	0.03	113.40**	3084	2.68	0.12
Error	78	0.03	1.69	0.39	8.73	1439.42	14.81	0.07

*, ** Significant at 0.05 and 0.01, respectively
 Rep = Replication, Covariate = Root rot

Table 2.6 Mean squares values of GY (grain yield), TW (test weight), SeedWt (single seed weight), SeedsHead⁻¹, HeadNo (heads m⁻²), HT (plant height) and HS (heading score) for alpha lattice trial in Diyarbakir, Turkey -2014

Source	df	Mean Squares					
		GY	SeedWt 10 ⁵	SeedsHead ⁻¹	HeadNo	HT	HS
Genotype	99	0.73*	2.74**	56.91**	8597.80**	142.59**	1.29**
Rep	1	12.81**	27.91**	26.96	17888.62	581.47**	2.43*
Block(Rep)	18	1.85**	1.93**	23.21	8271.4	98.95*	0.47
Covariate	1	5.34	0.69	9.38	51741.41**	0.02	0.02
Error	78	0.48	0.68	29.54	5169.25	53.62	0.44

*, ** Significant at 0.05 and 0.01, respectively

Rep = Replication, Covariate = Plant Stand

Table 2.7 Mean squares values of GY (grain yield), TW (test weight), SeedWt (single seed weight), SeedsHead⁻¹, HeadNo (heads m⁻²), HT (plant height) and HS (heading score) for alpha lattice trial in College Station, TX -2014

Source	df	Mean Squares						
		GY	TW	SeedWt 10 ⁵	SeedsHead ⁻¹	HeadNo	HT	HS
Genotype	99	0.29	8.69**	1.84**	45.86**	2005.27	59.02**	1.50**
Rep	1	0.78	1.12	3.29**	73.42	1944.91	15.74**	0.08
Block(Rep)	18	0.26	0.43	0.87*	30.43	2653.91	1.77	0.09
Covariate	1	22.16**	0.13	0.59	17.17	149476.10	5.97	0.07
Error	75-79	0.22	1.06	0.43	24.18	1949.46	1.91	0.1

*, ** Significant at 0.05 and 0.01, respectively

Rep = Replication, Covariate = Harvestable plant stand

2.3.1 Grain yield characteristics

Combined analysis of GY across environments showed a wide range of diversity from 2.14 t ha⁻¹ - 3.54 t ha⁻¹ and a mean of 2.92 t ha⁻¹. The least significant difference (LSD_{0.05}) between genotypes for GY was 0.82 t ha⁻¹. There was a fivefold difference in GY between high yielding and low yielding environments. The highest mean GY was observed in DYB2014 and lowest mean GY was observed in CH2014. In DYB2014, mean GY of 5.33 t ha⁻¹ and a range of 3.86 - 6.76 t ha⁻¹ was observed. In CH2014 mean GY of 1.01 t ha⁻¹ and a range of 0.29 - 1.59 t ha⁻¹ was observed (refer to page 44).

Although total precipitation in DYB2014 was almost similar to CH2014, GY average was exceptionally high in DYB2014. Favorable environmental conditions that include ideal temperature along with very good precipitation at the key stages of plant growth and residual soil moisture might be responsible for this. Plots in DYB2014 (81 mm) received two times more precipitation than plots in CH2014 (42.5 mm) during early plant establishment stage (Feekes scale 1-5, Large, 1954). This might have helped plants to produce more tillers and biomass (NDVI data for biomass not presented here). Many studies have shown a positive correlation between HeadNo and GY in winter wheat and between biomass at anthesis and GY (Petcu, 2003; White and Wilson, 2006). In addition, plots in DYB2014 (38.3 mm) received 2/3rd more precipitation than plots in CH2014 (21.08 mm) around anthesis (Feekes scale 10.1-10.5). In addition to precipitation, temperatures also played a critical role in determining GY at these two locations. Studies have shown that for every one degree rise in the temperature above 22 °C preceding anthesis, SeedsHead⁻¹ were reduced by 4%, and when temperature rises

above 30 °C it causes complete sterility (Fischer 1985; Saini and Aspinall, 1982). Temperature data during the cropping season, especially 30 days around anthesis (Feekes scale 10.1 to 10.5), better explains potential reasons for low yields in CH2014. Number of days when average temperature was above 22 °C in DYB2014 (4 days) was smaller than CH2014 (11 days). Furthermore, number of days when maximum temperature was more than 30 °C was lower in DYB2014 (8 days) compared to CH2014 (14 days). All of these factors might have contributed to the higher GY at DYB2014 and thus lower impact on seed set (seeds head⁻¹ and seed filling).

2.3.2 Yield components characteristics

2.3.2.1 Single seed weight (SeedWt)

A combined analysis showed there was a wide range of diversity for SeedWt. A range of 25.9 g - 40.9 mg and an average of 30.8 mg was observed. The LSD_{0.05} value for SeedWt was 3.7 g (Table 2.8). The lowest and highest SeedWt values were observed in CH2014 and CAS2014, respectively (Table 2.8).

The period between anthesis and physiological maturity, a.k.a., grain filling period/duration, is very important in determining SeedWt (Singh et al., 2014). Environment and genotype both play an important role in determining grain-filling duration in wheat (Bauer et al., 1985). Singh et al., (2014) reported high temperatures and water stress during grain filling period could significantly reduce grain filling duration and consequently SeedWt. Studies conducted by Jocković et al., (2014) have also shown a positive but not significant correlation between grain filling duration and

thousand seed weight. Some other studies have reported that a 5 °C increase in temperature above 20 °C increased the rate of grain filling and reduced the grain filling duration by 12 days in wheat (Yin et al., 2009). Drought and heat stress might have contributed to the low SeedWt in CH2014 trials. Temperature and precipitation data showed there were around six days when the temperature exceeded 25 °C in CAS2014 and around seven days in CH2014.

2.3.2.2 Seeds per head (SeedsHead⁻¹)

Combined analysis of SeedsHead⁻¹ showed an average of 35 SeedsHead⁻¹, a range of 27 - 43 SeedsHead⁻¹ and an LSD_{0.05} of 6 SeedsHead⁻¹ (Table 2.8). There was indeed a broad range of diversity within synthetic germplasm for this trait to warrant more progress through selection in the future.

Seed set in wheat is influenced by several factors, including temperature and water stresses. These two environmental stresses cause pollen sterility and ovary abortion and consequently lower overall seed set (Boyer and Westgate 2004). Bauder (2001) reported that water stress during the jointing stage significantly affects SeedHead⁻¹ in winter wheat. Based on studies conducted on stem reserves in wheat, Blum (1998) stated that pre-anthesis stem reserves are important for determining flowering and grain development under terminal drought and heat stress conditions. Fischer (1985) has shown that for every one degree rise in the temperature above 22 °C preceding anthesis SeedsHead⁻¹ are reduced by 4%, and when temperature rises above 30 °C it causes sterility (Saini and Aspinall, 1982). Plots in CH2014 received around 21 mm of

Table 2.8 Mean, range, LSD (least significant differences) for GY (grain yield), TW (test weight), SeedWt (single seed weight), SeedsHead⁻¹, HeadNo (heads m⁻²), HT (height), and HS (heading score) by environment and combined analysis across environments

	GY	TW	SeedWt (mg)	Seed Head⁻¹	HeadNo	HT	HS
<u>Combined</u>							
Mean	2.92	75.9	30.8	35	262	82.6	2.7
Range	(2.14-3.54)	(72.6-78.3)	(25.9-40.9)	(27-43)	(201-330)	(73.2-97.4)	(1.7-4.3)
LSD	0.82	2.7	3.7	6	74	6.5	0.9
<u>CAS2014</u>							
Mean	3.45	78.6	34.3	36	289	90.6	2.9
Range	(1.96-4.84)	(72.2-83.4)	(23.3-45.4)	(25-47)	(162-617)	(79.5-105.4)	(1.5-5.5)
LSD	1.66	1.9	3.7	8	160	2.7	1
<u>CH2014</u>							
Mean	1.01	71.1	26.1	22	181	58.8	2.1
Range	(0.29-1.59)	(60.7-74.4)	(18.5-36.0)	(14-30)	(95-600)	(44.2-71.0)	(0.96-4)
LSD	0.35	2.7	4.3	7	149	7.5	0.54
<u>DYB2014</u>							
Mean	5.33	83.3	34.0	38	431	93.8	2.9
Range	(3.86-6.76)	(78.1-87.3)	(26.7-48.8)	(25-56)	(280-673)	(72.9-126.8)	(1.5-5)
LSD	1.47	.	5.5	12	195	15.1	1.3
<u>CS2014</u>							
Mean	1.88	70.6	28.8	43	158	87.2	2.9
Range	(0.68-2.66)	(64.6-76.1)	(20.9-40.7)	(30-55)	(65-248)	(71.9-100.0)	(1.5-5)
LSD	0.97	1.9	4.4	11	94	2.6	0.6

Codes for environments are CAS2014 = Castroville, CH2014 = Chillicothe, Combined = across four environments, CS2014 = College Station, and DYB2014 = Diyarbakir

precipitation during anthesis. The average temperature exceeded 22 °C for 11 out of 30 days around anthesis and grain filling; also, we recorded 14 days when the maximum temperature exceeded 30 °C. In addition, there was root rot incidence in the field. All these factors combined might have reduced the seed set in CH2014 trials. Although, plants in CS2014 had much higher SeedsHead⁻¹ than CH2014, there was not great difference in SeedWt. Fischer et al., (1977) have reported SeedsHead⁻¹ and SeedWt were negatively correlated. Plants in CS2014 were also exposed to water (26 mm) and temperature stresses (11 days above 22 °C) around anthesis, but the intensity of stress was less than that at CH2014. Plants in CS2014 could potentially have good pre anthesis stem reserves because of good early season precipitation (108 mm).

2.3.2.3 Heads per square meter (HeadNo)

HeadNo is one of the most important traits that influence overall GY in wheat. The combined environment analysis for HeadNo showed a mean of 262 heads m⁻² and a range of 201 - 330 head m⁻², and an LSD_{0.05} of 74 heads m⁻² (Table 2.8). The highest and lowest values for this trait were observed in DYB2014 and CS2014, respectively.

Studies by Zhong-hu and Rajaram (1994) and Simanae (1993) revealed that HeadNo and SeedsHead⁻¹ are the most sensitive yield components under drought. Very good early season precipitation accompanied by ideal temperatures for plant growth and development in DYB2014 might have contributed to high tiller number and maintenance (supported by NDVI data, which is not presented here). Moreover, ideal temperature and precipitation at frequent intervals during grain filling in DYB2014 helped in efficient

mobilization of nutrients from source to sink. Trials in CS2014 and CH2014 had relatively low HeadNo. Low precipitation around anthesis might have resulted in lower HeadNo in CS2014. Studies conducted by Moayedi et al. (2010) best support the above statement. They have reported that HeadNo is very sensitive to drought stress and a drastic reduction in HeadNo might occur during reproductive stage with the onset of water stress.

2.3.3 Morphological characteristics

2.3.3.1 Plant height (HT)

Overall plants were shortest in CH2014 and taller in CAS2014 and DYB2014 (Table 2.8 and Table 2.9). Water stress seems to have a negative impact on overall HT. These results are in agreement with studies conducted by Richard et al., (1996) who reported water stress during cropping season reduced HT, which resulted in decrease in total biomass accumulated and GY. Similar results were also reported by Guendouz et al. (2012) who pointed out that plants were relatively shorter under water stressed conditions.

2.3.3.2 Heading score/heading date (HS)

There was a great range of genetic diversity for heading date/score (HS) in the current germplasm. The HS varied from very early to very late. However, most of the genotypes were classified as medium (Table 2.8 and Table 2.9). TAM 401, TAM 112 and TAM 111 were used as a reference for determining very early to very late heading

genotypes. TAM 401, TAM 112 and TAM 111 were classified as early, early to medium, and medium heading, respectively. Except CH2014, HS for most other locations HS was around 2.9. Plots in CH2014 seem to have headed earlier than other locations. Bauder (2001) reported that drought stress in winter wheat stimulates early heading and make plants head seven to ten days earlier than normal. Intensity and duration of stress also plays a vital role in determining how early a plant will be heading. Early heading eventually results in shortened growth period, HT, biomass accumulation and consequently lower GY. Precipitation and temperature data has shown that plots in CH2014 were consistently exposed to both water and temperature stress. This might have played a role in preponing the heading date in CH2014 compared to other locations.

Table 2.9 Best linear unbiased estimation (BLUEs) values of GY (grain yield), TW (test weight), SeedWt (single seed weight), SeedsHead⁻¹, HeadNo (heads m⁻²), HT (height), and HS (heading score) for all genotypes and their corresponding stability estimates (b_i) and regression deviation (S_{di}^2) for combined analysis

ID	Genotype	GY	TW	SeedWt	Seed	HeadNo	HT	HS	b_i	S_{di}^2
		t ha ⁻¹	kg hl ⁻¹	mg	Head ⁻¹ count					
1	TX11Vsyn0101	3.24	77.1	32.4	36	251	84.8	2.7	1.27	0.24
2	TX11Vsyn0103	2.65	73.5	25.9	36	271	78.4	2.7	0.77	-0.14
3	TX11Vsyn0110	2.96	76.5	30.9	33	277	87.2	2.6	1.00	0.09
4	TX11Vsyn0111	2.77	77.8	35.1	30	252	82.0	1.9	1.01	-0.10
5	TX11Vsyn0112	2.71	76.2	30.1	36	238	83.6	3.1	1.01	0.09
6	TX11Vsyn0113	2.95	78.1	33.7	34	237	87.6	2.4	1.04	0.18
7	TX11Vsyn0116	2.95	75.1	30.3	33	279	77.8	3.0	1.02	0.29
8	TX11Vsyn0118	3.28	76.2	31.9	36	279	81.6	2.4	0.95	-0.19
9	TX11Vsyn0119	2.86	76.8	31.4	36	242	79.9	3.6	1.09	-0.20
10	TX11Vsyn0120	2.97	75.0	33.1	39	209	88.3	3.0	1.06	0.09
11	TX11Vsyn0122	3.51	77.0	30.3	41	259	85.0	3.3	1.23	-0.21
12	TX11Vsyn0123	3.28	77.2	31.9	37	255	86.1	2.1	1.14	-0.22
13	TX11Vsyn0124	2.61	75.1	29.7	35	258	83.3	4.0	0.93	-0.23
14	TX11Vsyn0127	2.63	75.7	32.5	37	203	90.2	3.8	0.84	0.17
15	TX11Vsyn0130	3.01	76.8	27.1	37	288	76.7	3.1	1.08	0.09
16	TX11Vsyn0131	2.50	77.1	32.4	30	248	84.1	2.4	0.93	0.11
17	TX11Vsyn0133	3.03	76.6	31.8	36	247	83.3	2.6	1.18	-0.09
18	TX11Vsyn0134	2.75	76.6	31.8	32	255	80.7	2.2	0.95	0.03
19	TX11Vsyn0135	2.86	76.2	32.9	36	223	78.8	3.0	1.16	-0.01
20	TX11Vsyn0136	3.04	74.0	29.7	33	301	93.5	4.0	1.06	0.62
21	TX11Vsyn0137	3.33	76.2	33.1	33	283	83.0	4.2	1.17	-0.06
22	TX11Vsyn0138	2.73	75.4	36.6	32	244	87.4	2.2	0.89	0.12
23	TX11Vsyn0140	3.24	75.7	40.9	28	273	89.7	2.1	0.94	-0.11

Table 2.9 Continued

ID	Genotype	GY	TW	SeedWt	Seed Head ⁻¹	HeadNo	HT	HS	b_i	S_{di}^2
24	TX11Vsyn0146	3.02	76.8	33.1	31	286	87.5	3.4	0.96	0.02
25	TX11Vsyn0153	2.59	75.0	27.5	34	279	73.2	2.5	0.82	-0.20
26	TX11Vsyn0154	2.94	76.7	27.6	38	268	81.0	2.5	0.80	-0.02
27	TX11Vsyn0156	2.96	74.3	30.9	36	227	85.5	3.2	1.03	-0.18
28	TX11Vsyn0158	2.85	77.5	34.5	34	231	84.0	3.3	0.99	0.11
29	TX11Vsyn0159	2.86	73.8	27.5	40	235	78.6	3.1	1.09	1.05
30	TX11Vsyn0160	3.24	77.4	32.6	35	272	82.4	3.7	1.01	0.58
31	TX11Vsyn0161	2.74	75.6	26.4	36	293	84.4	3.8	0.98	0.04
32	TX11Vsyn0164	2.96	75.3	31.2	36	266	81.3	2.7	0.59	0.27
33	TX11Vsyn0165	3.46	75.1	32.8	37	278	87.1	2.4	1.06	0.12
34	TX11Vsyn0167	3.18	77.3	32.1	33	277	80.6	2.8	0.98	0.07
35	TX11Vsyn0168	2.72	74.0	28.6	35	252	80.7	2.5	0.82	0.14
36	TX11Vsyn0169	2.82	77.4	34.3	35	225	84.3	2.9	0.92	0.28
37	TX11Vsyn0174	2.55	75.5	33.8	31	244	85.7	3.6	0.90	0.23
38	TX11Vsyn0175	2.81	74.3	28.4	33	285	81.0	2.9	1.03	0.18
39	TX11Vsyn0178	2.99	75.8	29.4	34	279	79.7	2.8	0.97	0.65
40	TX11Vsyn0179	2.87	76.6	29.8	31	317	77.9	2.9	1.23	-0.16
41	TX11Vsyn0180	2.85	74.5	27.9	39	232	81.2	3.6	1.12	-0.17
42	TX11Vsyn0182	3.35	76.9	33.4	35	262	90.0	2.0	1.22	-0.09
43	TX11Vsyn0185	3.28	76.5	30.4	35	275	81.2	2.9	1.41	0.02
44	TX11Vsyn0188	3.18	73.4	30.6	34	290	82.8	4.1	0.88	0.22
45	TX11Vsyn0189	3.24	76.1	30.9	32	313	80.2	2.1	1.01	0.47
46	TX11Vsyn0190	2.85	73.6	26.8	37	282	80.3	2.7	0.84	-0.23
47	TX11Vsyn0191	3.08	78.4	30.3	35	281	84.2	2.4	1.08	-0.12
48	TX11Vsyn0195	3.24	76.6	33.7	37	236	89.4	2.4	1.20	-0.23
49	TX11Vsyn0196	2.75	74.4	35.7	33	219	86.3	2.3	0.85	0.06
50	TX11Vsyn0197	3.02	76.4	34.5	35	240	89.5	2.2	1.09	-0.23

Table 2.9 Continued

ID	Genotype	GY	TW	SeedWt	Seed Head ⁻¹	HeadNo	HT	HS	b_i	S_{di}^2
51	TX11Vsyn0199	3.29	76.1	31.0	34	306	89.2	4.1	0.94	-0.02
52	TX11Vsyn0201	2.64	75.6	34.2	29	253	83.7	2.7	0.95	-0.21
53	TX11Vsyn0208	2.68	76.7	32.1	33	231	88.4	2.4	1.08	-0.17
54	TX11Vsyn0211	2.72	76.6	31.5	36	236	80.4	2.8	0.83	-0.20
55	TX11Vsyn0212	2.80	73.3	28.5	43	221	76.9	2.4	0.73	0.05
56	TX11Vsyn0213	3.10	75.9	30.8	39	246	79.7	2.3	1.08	0.34
57	TX11Vsyn0216	2.95	76.2	27.0	36	286	78.6	3.3	0.92	0.12
58	TX11Vsyn0217	2.90	76.1	35.5	32	245	90.2	2.3	1.11	-0.09
59	TX11Vsyn0219	3.07	74.2	32.4	33	264	78.3	2.6	1.05	0.17
60	TX11Vsyn0225	3.19	76.0	32.9	33	285	75.7	2.2	1.11	-0.06
61	TX11Vsyn0226	2.85	76.3	33.0	31	247	79.4	2.3	1.03	0.11
62	TX11Vsyn0228	2.82	77.5	30.3	33	272	80.2	2.1	1.01	-0.06
63	TX11Vsyn0229	2.50	74.4	31.1	32	240	83.0	2.0	0.81	-0.01
64	TX11Vsyn0230	3.13	77.5	33.0	32	270	80.5	2.2	0.98	-0.19
65	TX11Vsyn0232	2.38	76.1	31.2	29	236	87.4	2.0	0.97	0.32
66	TX11Vsyn0234	3.14	77.4	30.3	34	291	78.8	1.9	1.13	-0.01
67	TX11Vsyn0238	2.86	74.8	30.8	35	250	77.4	2.2	0.97	-0.17
68	TX11Vsyn0240	2.52	76.2	30.9	29	262	86.2	2.0	0.94	-0.19
69	TX11Vsyn0241	2.88	74.8	28.5	32	297	75.8	2.0	1.02	-0.04
70	TX11Vsyn0243	2.72	75.7	35.8	29	247	88.4	2.2	0.78	-0.11
71	TX11Vsyn0253	2.49	75.7	27.8	29	319	83.6	1.9	1.02	-0.12
72	TX11Vsyn0261	2.80	76.3	30.7	40	221	77.3	2.5	0.92	-0.05
73	TX11Vsyn0263	2.98	75.3	30.6	34	266	80.2	2.6	1.14	0.01
74	TX11Vsyn0264	3.05	74.1	30.5	34	272	80.2	2.4	0.94	0.17
75	TX11Vsyn0265	3.05	76.0	31.6	31	298	90.5	2.0	0.96	-0.04
76	TX11Vsyn0266	2.86	75.3	30.1	37	249	81.8	2.1	0.87	0.46
77	TX11Vsyn0267	2.53	75.4	28.2	34	239	78.1	2.1	1.04	0.12
78	TX11Vsyn0271	3.16	76.6	30.8	32	283	83.9	2.0	1.23	0.29

Table 2.9 Continued

ID	Genotype	GY	TW	SeedWt	Seed Head ⁻¹	HeadNo	HT	HS	b_i	S_{di}^2
79	TX11Vsyn0272	3.19	73.6	30.4	32	307	78.5	2.2	1.14	0.10
80	TX11Vsyn0275	2.88	77.7	30.4	33	271	79.1	1.7	0.99	0.21
81	TX11Vsyn0277	3.07	75.6	31.3	36	257	82.2	3.0	1.14	-0.16
82	TX11Vsyn0279	2.52	75.6	27.3	33	259	76.2	2.4	0.92	0.62
83	TX11Vsyn0280	2.26	72.5	27.6	29	268	83.8	2.0	1.02	0.50
84	TX11Vsyn0282	2.64	75.0	28.1	31	285	87.3	4.0	0.80	0.27
85	TX11Vsyn0294	2.97	73.9	27.5	38	250	75.6	2.3	0.97	0.21
86	TX11Vsyn0300	3.42	75.1	28.4	43	280	85.1	2.5	1.19	-0.12
87	TX11Vsyn0303	2.14	77.6	28.2	36	202	93.9	4.2	0.66	-0.02
88	TX11Vsyn0305	2.53	74.1	28.2	35	253	80.9	3.3	0.60	0.79
89	TX11Vsyn0306	2.70	76.3	30.5	36	235	82.8	3.2	0.93	0.03
90	TX11Vsyn0308	2.90	76.1	31.2	37	231	80.5	2.9	0.95	0.15
91	TX11Vsyn0309	2.82	74.8	30.5	36	235	87.3	2.1	1.13	-0.07
92	TX11Vsyn0312	3.16	75.3	26.2	37	330	83.1	3.0	1.13	-0.13
93	TX11Vsyn0313	2.82	76.8	29.4	37	237	97.4	4.2	0.86	-0.10
94	TAM112	3.19	76.6	28.3	33	324	74.6	2.0	1.18	0.00
95	TAM111	2.80	77.6	29.1	39	231	80.5	2.9	0.81	-0.03
96	TAM113	3.34	78.8	29.6	35	302	78.8	2.9	1.25	-0.15
97	TAM304	3.20	74.8	26.5	40	279	73.4	2.4	1.09	0.43
98	TAM305	2.60	77.0	28.9	36	243	74.9	2.4	0.89	0.59
99	TAM401	3.54	75.3	27.1	40	304	82.3	2.4	1.46	1.35
100	TAMW101	2.78	77.7	35.2	27	288	75.9	3.1	0.72	-0.11
	Mean	2.92	75.87	30.8	34	262	82.6	2.7		
	Minimum	2.14	72.55	25.9	27	202	73.2	1.7		
	Maximum	3.54	78.83	40.9	43	330	97.3	4.3		
	LSD	0.82	2.72	3.7	6	74	6.5	0.9		
	CV (%)	22.8	1.6	7.5	13.1	23.7	5.4	16.7		

2.3.4 Interrelationships between traits

2.3.4.1 Correlations among yield and other traits

Correlation coefficient values help us to understand the nature and magnitude of the relationship between two variables. Phenotypic correlation coefficients (r_p) are reported in table 2.10. Except for seed weight, all yield components had a positive and significant correlation with overall GY. Many other studies conducted on synthetic, spring, and winter wheat also showed similar results (Khan and Naqvi, 2012; Mohsin et al., 2009; Gupta et al., 1999). Seed weight had positive but no significant correlation with GY ($r_p = 0.14$, $P < 0.05$). HeadNo had the highest positive correlation with GY ($r_p = 0.44$, $P < 0.001$) followed by SeedsHead⁻¹ ($r_p = 0.33$, $P < 0.001$). Studies conducted by Khan and Naquvi (2012) also showed that HeadNo had a positive and significant correlation with GY under different water levels and stresses imposed at different growth stages. Similar results were also reported for SeedsHead⁻¹ (Khan and Naquvi, 2001). Morphological traits such as HS ($r_p = -0.029$) and HT ($r_p = -0.022$) had negative but non-significant correlation with GY. Hossain et al., (2012a) reported that heat and water stresses play a critical role in determining HT and heading date in wheat, especially in dryland areas. Three out of four trials in our study were planted under rainfed conditions. Therefore, temperature and precipitation are believed to have played an important role in determining HT and heading date in our study. A number of studies have shown a negative correlation between heading date and GY under extreme temperature and water stress conditions (Hossain et al., 2012b; Mohammadi et al., 2012). Our results showed that trials in most locations were exposed to high

temperatures, and in some locations water stress was also present. Although, three out of four trials were planted under rainfed conditions, only one had extreme water stress during the vegetative stage. Water stress from jointing to heading stages plays a critical role in determining HT in wheat (Khokhar et al., 2010). Many studies have shown that when water stress is very low during the vegetative stage, HT had a negative correlation with GY depending on germplasm used in the study (Khokhar et al., 2010).

Among the yield components, SeedWt had negative and highly significant correlation with SeedsHead⁻¹ ($r_p = -0.43$) and HeadNo ($r_p = -0.30$). Similarly, SeedsHead⁻¹ had negative and significant correlation with HeadNo ($r_p = -0.32$). Non-significant negative correlation was observed between HS and HeadNo ($r_p = -0.070$) and between HS and SeedWt ($r_p = -0.14$). However, the positive significant correlation between HS and SeedsHead⁻¹ ($r_p = 0.21$) was observed (Table 2.10).

Table 2.10 Pearson correlation coefficients among GY (grain yield), TW (test weight), SeedWt (Single seed weight), SeedsHead⁻¹, HeadNo (heads m⁻²), HS (heading score), and HT (height) based on means of combined analysis

	GY	TW	SeedWt	SeedsHead ⁻¹	HeadNo	HS	HT
GY		0.17 ^{ns}	0.14 ^{ns}	0.32**	0.44**	-0.029 ^{ns}	-0.02 ^{ns}
TW			0.33**	-0.12 ^{ns}	-0.03 ^{ns}	-0.03 ^{ns}	0.12 ^{ns}
SeedWt				-0.43**	-0.30**	-0.14 ^{ns}	0.40**
SeedsHead ⁻¹					-0.28**	0.21*	-0.13 ^{ns}
HeadNo						-0.07 ^{ns}	-0.25*
HS							0.25*
HT							

* Significant at the 0.05 probability level; ** Significant at the 0.01 probability level; ns not significant

2.3.4.2 Path coefficient analysis among yield and yield components

Simple correlation coefficients cannot explain the importance of each factor and interrelationship among the factors in producing a given correlation coefficient (Ibrahim et al., 2012). Therefore, tools such as path coefficient analysis that partitions the correlation coefficient into direct and indirect effects can be more effective than simple correlations in examining the effect of a specific factor and hence to understand the relationship between two traits. This information on direct and indirect effects will help in formulating an effective selection strategy for improvement of a particular trait. Grain yield is a product of three yield components such as SeedWt, SeedsHead⁻¹, and HeadNo. The values that are bold and in the diagonal direction are the direct effect and the values that are horizontal in the same row are indirect effects (Table 2.11). The combination of direct and indirect effects results in total effect due to that trait on GY. For example, in the case of combined analysis, direct effect of HeadNo on GY is 0.95 but because of indirect negative effect of similarly strong SeedWt (-0.25) and SeedsHead⁻¹ (-0.26) total effect due to HeadNo is limited at 0.44. Many studies have shown that an increase in one yield component might result in decreasing other component because of yield compensatory effects to maintain a balance between the source and the sink (Cooper et al., 2012; Fischer et al., 1985).

In the combined environment analysis, the highest and most significant positive correlation was observed between HeadNo (0.44) and GY followed by SeedsHead⁻¹ (0.33) and GY. Lowest and the non-significant correlation was observed between SeedWt (0.14) and GY (Table 2.11). The values of path coefficients are in accordance

with Pearson correlation coefficients. Similar results were reported by Cooper et al. (2012). This suggests that HeadNo followed by SeedsHead⁻¹ was the best indirect selection criteria for increasing GY in SDW. Any gain in HeadNo and SeedsHead⁻¹ can positively improve GY. The same rules applied to other environments as well. Path coefficient analysis for single environments showed similar results as combined analysis (Table 2.11).

Table 2.11 Estimates of direct, indirect effect of SeedWt (Single seed weight), SeedsHead⁻¹, and HeadNo (heads m⁻²) on grain yield (GY) and total correlation with GY based on means of combined analysis

Trait	SeedWt	HeadNo	SeedHead ⁻¹	GY
		<u>Combined</u>		
SeedWt	0.83	-0.29	-0.40	<u>0.14^{ns}</u>
HeadNo	-0.25	0.95	-0.26	<u>0.44**</u>
SeedsHead ⁻¹	-0.35	-0.26	0.94	<u>0.33**</u>
		<u>CAS2014</u>		
SeedWt	0.64	-0.29	-0.14	<u>0.21*</u>
HeadNo	-0.20	0.93	-0.19	<u>0.54**</u>
SeedsHead ⁻¹	-0.13	-0.24	0.71	<u>0.34**</u>
		<u>CH2014</u>		
SeedWt	0.30	0.01	-0.06	<u>0.26**</u>
HeadNo	0.01	0.95	-0.14	<u>0.56**</u>
SeedsHead ⁻¹	-0.03	-0.18	0.57	<u>0.36**</u>
		<u>DYB2014</u>		
SeedWt	0.77	-0.24	-0.41	<u>0.12^{ns}</u>
HeadNo	-0.18	1.02	-0.56	<u>0.28**</u>
SeedsHead ⁻¹	-0.29	-0.53	1.08	<u>0.26**</u>
		<u>CS2014</u>		
SeedWt	0.47	-0.23	-0.20	<u>0.04^{ns}</u>
HeadNo	-0.11	0.95	-0.03	<u>0.81**</u>
SeedsHead ⁻¹	-0.18	-0.06	0.53	<u>0.29**</u>

* Significant at the 0.05 probability level; ** Significant at the 0.01 probability level; ns not significant

Codes for environments are CAS2014 = Castroville, CH2014 = Chillicothe, Combined = across four environments, CS2014 = College Station, and DYB2014 = Diyarbakir

2.3.5 Elite genotypes

Combined analysis of environments showed that 8 out of the top 10 lines in the yield trials were SDW lines. We documented a number of SDW lines that had higher GY than their recurrent parents and some other check varieties in individual environments as well as in the combined analysis (refer to page 59 and 60). The SDW line TX11Vsyn0122 ranked consistently high across the different environments. Nine of the top ten SDW lines had TAM 111 as the recurrent parent and only one line (TX11Vsyn0300) had TAM 112 as the recurrent parent. This suggests that TAM 111 might have better combining ability than TAM 112. Combining ability studies showed that among the recurrent parents TAM 111 had better general combining ability than TAM 112 (data not presented).

Apart from mean GY, stability of the genotype is one more factor that needs to be considered when releasing a variety. We used Eberhart and Russell's (type III) regression coefficient method to determine stability. Eberhart and Russell (1966) suggested that both linear regression coefficient (b_i) and deviation (S_{di}^2) from the regression coefficient need to be considered when determining stability. The b_i determines linear response to environmental change by explaining the relationship between the yield of the genotype for each environment and mean yield for the environment. On the other hand, S_{di}^2 determines consistency of this response by explaining the deviation from the regression. When the regression coefficient (b_i) is close to 1 and S_{di}^2 is not different from zero the genotype is considered to have average stability (Eberhart and Russell's, 1966). When the b_i is more than 1 the genotype is

considered to have low stability but high sensitivity to environmental changes and might be better for high yielding environments (Wachira et al., 2002). When the b_i is less than 1 the genotype has high stability and low sensitivity to environmental changes and might be better for low yielding environments (Wachira et al., 2002). Overall, the genotypes with high mean yield, average stability and zero deviation from the regression are considered the best for the trait under consideration. Stability values for each SDW lines are presented in the table on page 48 to 51.

The coefficient of variation (CV) for GY in CAS2014 (24%), CS2014 (26%), and CH2014 (17.8%) was higher than the acceptable range for variety trials (Table 2.12). There might be multiple reasons for the high CV in some of these environments. Uneven distribution of rust across the field along with lodging might have caused excessive CV in the CAS2014 trial. In the CS2014 trial, lodging caused by excessive rain during physiological maturity might have resulted in high CV. Drought stress accompanied by root rot might have resulted in high CV in CH2014.

Comparing the mean of the top ten yielding SDW with the mean of seven check varieties showed that SDW lines (3.34 t ha^{-1}) produced 0.30 t ha^{-1} higher GY than the check varieties average (3.05 t ha^{-1}). A detailed study of the factors that produced higher GY in SDW lines showed that SDW lines maintained similar SeedsHead⁻¹ and HeadNo but had higher SeedWt than check varieties. Our studies showed that SDW lines had an average of 32.0 mg SeedWt , $37 \text{ SeedsHead}^{-1}$, and 273 heads m^{-2} ; whereas the check varieties had 29.0 mg SeedWt , $36 \text{ SeedsHead}^{-1}$, and 281 heads m^{-2} . Although check varieties had 7 more heads m^{-2} on average than the SDW lines, when we considered the

LSD_{0.05} of 74 heads m⁻² this is a negligible value. We also observed that SDW lines (85.3 cm) were relatively taller than check varieties (77.2 cm) by 8 cm. Check varieties and SDW lines were classified as being early to medium heading (score 2.6 to 2.8) (Table 2.13). These results are in accordance with the findings of Narasimhamoorthy et al., (2006).

2.3.6 GGE biplot (5 environments)

GGE biplot is a data visualizing software that helps to understand G*E interactions in a more efficient way (Yan et al., 2003). GGE biplot can classify genotypes into different mega environments and determine the elite and ideal genotype for each one of these environments. In addition, GGE biplot helps to estimate the stability of genotypes and understand the interrelationship among the target environments and target traits. Data from all five environments was used to for GGE biplot analysis. The five environments included CS2013, CAS2014, CS2014, CH2014, and DYB2014.

2.3.6.1 GGE biplot for grain yield

The polygon view of the GGE biplot for GY presents which genotype is best for which mega environments (Figure 2.1). The polygon is formed by connecting the genotypes that are furthest away (good or bad) from the origin of the biplot so that all genotypes are grouped within the polygon. A sector is formed by drawing perpendicular line between two adjacent genotype that form

Table 2. 12 Mean grain yield (BLUEs) of ten top yielding synthetic derived wheat lines determined from combined analysis and performance of these lines in each environment, their respective ranking (R), stability (b_i) and regression deviation (S_{di}^2)

ID	Genotype	CAS	R	CH	R	DYB	R	CS	R	Across	R	b_i	S_{di}^2
		2014 t ha ⁻¹		2014 t ha ⁻¹		2014 t ha ⁻¹		2014 t ha ⁻¹		2014 t ha ⁻¹			
11	TX11Vsyn0122	4.28	11	1.39	9	6.28	6	2.05	31	3.51	2	1.23	-0.21
33	TX11Vsyn0165	3.93	24	0.98	58	6.02	15	2.49	7	3.46	3	1.06	0.12
86	TX11Vsyn0300	4.24	12	0.98	57	6.01	16	1.85	53	3.42	4	1.19	-0.12
42	TX11Vsyn0182	3.48	45	0.78	80	6.18	9	2.02	34	3.35	5	1.22	-0.09
21	TX11Vsyn0137	4.41	8	1.23	22	5.88	20	1.64	73	3.33	7	1.17	-0.06
51	TX11Vsyn0199	3.86	29	0.62	93	4.9	73	2.43	13	3.29	8	0.94	-0.02
12	TX11Vsyn0123	3.89	27	1.24	20	6.03	14	1.94	44	3.28	9	1.14	-0.22
8	TX11Vsyn0118	3.85	30	1.19	27	5.4	48	2.15	25	3.28	10	0.95	-0.19
43	TX11Vsyn0185	4.64	2	0.79	78	6.31	5	1.31	93	3.28	11	1.41	0.02
1	TX11Vsyn0101	3.31	59	1.23	25	6.76	1	1.94	43	3.24	12	1.27	0.24
94	TAM112	3.24	65	1.12	35	6.21	8	1.63	74	3.19	19	1.18	0.00
95	TAM111	3.4	48	1	51	4.77	78	2.23	22	2.80	70	0.81	-0.03
96	TAM113	4.19	14	1.06	46	6.59	2	1.68	65	3.34	6	1.25	-0.15
97	TAM304	4.84	1	1.28	17	5.83	22	1.51	83	3.20	17	1.09	0.43
98	TAM305	1.97	99	0.93	65	5.48	43	1.93	45	2.60	88	0.89	0.59
99	TAM401	4.47	7	1.06	44	6.39	3	2.02	32	3.54	1	1.46	1.35
100	TAMW101	3.32	56	1.13	33	4.33	96	2.06	30	2.78	71	0.72	-0.11
	Mean	3.45		1.01		5.33		1.88		2.92			
	Min	1.97		0.29		3.86		0.68		2.14			
	Max	4.84		1.59		6.76		2.67		3.54			
	LSD	1.66		0.35		1.47		0.97		0.82			
	CV (%)	24.1		17.8		13.8		26.0		22.8			

Table 2.13 Mean GY (grain yield), TW (test weight), SeedWt (single seed weight), SeedsHead⁻¹, HeadNo (heads m⁻²), HT (height), and HS (heading score) of top ten yielding synthetic derived wheat lines and check varieties across four environments

ID	Genotype	GY	TW	SeedWt	Seed Head ⁻¹	HeadNo	HT	HS
		t ha ⁻¹	kg hl ⁻¹	mg	count	heads m ⁻²	cm	scale 1-5
11	TX11Vsyn0122	3.51	77.0	30.3	41	259	85.0	3.3
33	TX11Vsyn0165	3.46	75.1	32.8	37	278	87.1	2.4
86	TX11Vsyn0300	3.42	75.1	28.4	43	280	85.1	2.5
42	TX11Vsyn0182	3.35	76.9	33.4	35	262	90.0	2.0
21	TX11Vsyn0137	3.33	76.2	33.1	33	283	83.0	4.2
51	TX11Vsyn0199	3.29	76.1	31	34	306	89.2	4.1
12	TX11Vsyn0123	3.28	77.2	31.9	37	255	86.1	2.1
8	TX11Vsyn0118	3.28	76.2	31.9	36	279	81.6	2.4
43	TX11Vsyn0185	3.28	76.5	30.4	35	275	81.2	2.9
1	TX11Vsyn0101	3.24	77.1	32.4	36	251	84.8	2.7
94	TAM112	3.19	76.6	28.3	33	324	74.6	2.0
95	TAM111	2.80	77.6	29.1	39	231	80.5	2.9
96	TAM113	3.34	78.8	29.6	35	302	78.8	2.9
97	TAM304	3.20	74.8	26.5	40	279	73.4	2.4
98	TAM305	2.60	77.0	28.9	36	243	74.9	2.4
99	TAM401	3.54	75.3	27.1	40	304	82.3	2.4
100	TAMW101	2.78	77.7	35.2	27	288	75.9	3.1
	Mean	2.92	75.9	30.8	34	262	82.6	2.7
	Min	2.14	72.5	25.9	27	202	73.2	1.7
	Max	3.54	78.8	40.9	43	330	97.4	4.2
	LSD	0.82	2.7	3.7	6	74	7.8	0.9
	CV (%)	22.8	1.6	7.5	13.1	23.7	5.4	16.7

BLUES = Best linear unbiased estimated mean; Codes for environments are CAS2014 = Castroville, CH2014 = Chillicothe, Combined = across four environments, CS2014= college station, and DYB2014 = Diyarbakir

the polygon (Yan et al., 2003). For example, line 3 is drawn between genotype 11 and 56 and this forms sector 3. Nine sectors were formed when a polygon was drawn. Although, there were nine sectors, our testing environments were divided into only four sectors. A genotype that is in the corner of the polygon a.k.a. vertex is the most responsive genotype for that sector and environments within that sector. Sector 3 was formed between perpendicular lines 3 and 4. This sector includes environments CS2013 and CAS2014 and genotype TX11Vsyn0213 (ID = 56) is the best genotype for sector 3. Similarly, genotype TX11Vsyn0294 (ID = 85) is the best for CS2014 and sector 4. Genotype TX11Vsyn0275 (ID = 80) is the best for CH2014 environment and sector 1, and genotype TX11Vsyn0122 (ID = 11) is best for environment DYB2014 and sector 2. The other vertex genotypes TX11Vsyn0174 (ID = 37), TX11Vsyn0303 (ID = 87), TX11Vsyn0282 (ID = 84), and TX11Vsyn0305 (ID = 88), which were located away from all other testing environments, were the poorest among all the environments. The genotypes that are close to the origin of the biplot are less responsive to environmental changes than the vertex genotypes.

Interrelationship among the testing environments for GY is presented in figure 2.2. Vectors or the lines that connect the environments to the origin of the biplot were used to study the interrelationship between these environments. The angle between any two vectors approximates the correlation coefficient between them (Yan et al., 2003). An obtuse ($> 90^\circ$) angle indicates a negative correlation whereas an acute angle indicates a positive correlation. On the other hand, a right angle is indicative of the absence of correlation. The angle between CS2013 and CAS2014 was very narrow, indicating that

these environments were very similar. The same is true for CH2014 and CS2014, which also had an obtuse angle between them. Based on this biplot view, there seem to be three major groups of environments (Figure 2.2): group 1 included DYB2014 and CH2014, group 2 included CS2013 and CAS2014, and group 3 included CS2014 by its own. This clustering of environments matches to a lot of extent the climatic pattern and growing conditions within each group. Group 1 environments DYB2014 (163 mm) and CH2014 (148.8 mm) received similar precipitation and had similar temperatures during the cropping season. Likewise, group 2 included environments CS2013 and CAS2014, which were closer geographically to one another. Although, CS2014 had similar climatic conditions as group 2 environments these plots were grown under rainfed conditions and were subjected to different growing conditions.

The length of the environment vector is proportional to the variance within the environment and hence discriminating ability (Yan et al., 2003). The environments with longer vectors show greater range of variance and larger variability among entries. Figure on page 69 shows that CH2014, DYB2014, and CS2014 had longer vectors than CAS2014 and CS2013; therefore, there was more variability among genotypes in the first three environments than the latter two.

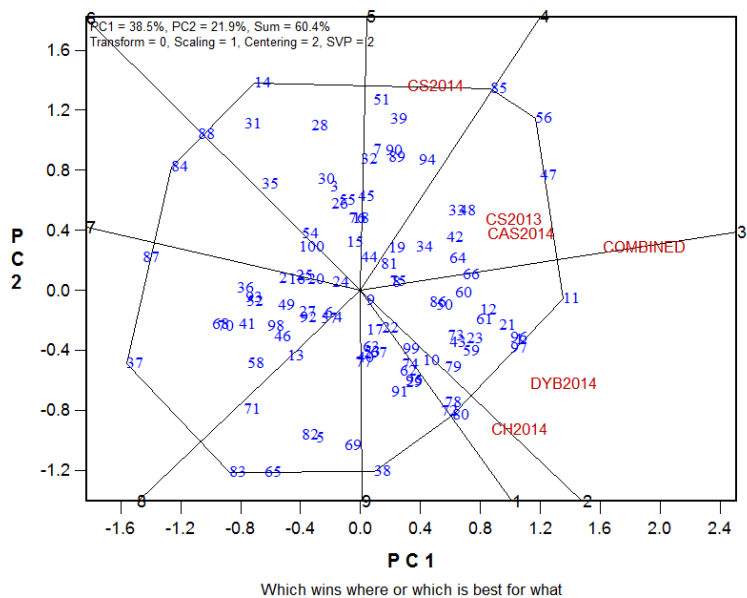


Figure 2.1 GGE biplot showing the best genotypes for grain yield (GY) in each test environment. Codes for environment are: CH2014= Chillicothe, DYB2014= Diyarbakir, CAS2014= Castroville, CS2014= College Station 2014, CS2013= College Station 2013 and Combined = combined mean. ID for lines are: 1=TX11Vsyn0101; 2=TX11Vsyn0103; 3=TX11Vsyn0110; 4=TX11Vsyn0111; 5=TX11Vsyn0112; 6=TX11Vsyn0113; 7=TX11Vsyn0116; 8=TX11Vsyn0118; 9=TX11Vsyn0119; 10=TX11Vsyn0120; 11=TX11Vsyn0122; 12=TX11Vsyn0123; 13=TX11Vsyn0124; 14=TX11Vsyn0127; 15=TX11Vsyn0130; 16=TX11Vsyn0131; 17=TX11Vsyn0133; 18=TX11Vsyn0134; 19=TX11Vsyn0135; 20=TX11Vsyn0136; 21=TX11Vsyn0137; 22=TX11Vsyn0138; 23=TX11Vsyn0140; 24=TX11Vsyn0146; 25=TX11Vsyn0153; 26=TX11Vsyn0154; 27=TX11Vsyn0156; 28=TX11Vsyn0158; 29=TX11Vsyn0159; 30=TX11Vsyn0160; 31=TX11Vsyn0161; 32=TX11Vsyn0164; 33=TX11Vsyn0165; 34=TX11Vsyn0167; 35=TX11Vsyn0168; 36=TX11Vsyn0169; 37=TX11Vsyn0174; 38=TX11Vsyn0175; 39=TX11Vsyn0178; 40=TX11Vsyn0179; 41=TX11Vsyn0180; 42=TX11Vsyn0182; 43=TX11Vsyn0185; 44=TX11Vsyn0188; 45=TX11Vsyn0189; 46=TX11Vsyn0190; 47=TX11Vsyn0191; 48=TX11Vsyn0195; 49=TX11Vsyn0196; 50=TX11Vsyn0197; 51=TX11Vsyn0199; 52=TX11Vsyn0201; 53=TX11Vsyn0208; 54=TX11Vsyn0211; 55=TX11Vsyn0212; 56=TX11Vsyn0213; 57=TX11Vsyn0216; 58=TX11Vsyn0217; 59=TX11Vsyn0219; 60=TX11Vsyn0225; 61=TX11Vsyn0226; 62=TX11Vsyn0228; 63=TX11Vsyn0229; 64=TX11Vsyn0230; 65=TX11Vsyn0232; 66=TX11Vsyn0234; 67=TX11Vsyn0238; 68=TX11Vsyn0240; 69=TX11Vsyn0241; 70=TX11Vsyn0243; 71=TX11Vsyn0253; 72=TX11Vsyn0261; 73=TX11Vsyn0263; 74=TX11Vsyn0264; 75=TX11Vsyn0265; 76=TX11Vsyn0266; 77=TX11Vsyn0267; 78=TX11Vsyn0271; 79=TX11Vsyn0272; 80=TX11Vsyn0275; 81=TX11Vsyn0277; 82=TX11Vsyn0279; 83=TX11Vsyn0280; 84=TX11Vsyn0282; 85=TX11Vsyn0294; 86=TX11Vsyn0300; 87=TX11Vsyn0303; 88=TX11Vsyn0305; 89=TX11Vsyn0306; 90=TX11Vsyn0308; 91=TX11Vsyn0309; 92=TX11Vsyn0312; 93=TX11Vsyn0313; 95=TAM111; 94=TAM112; 96=TAM113; 97=TAM304

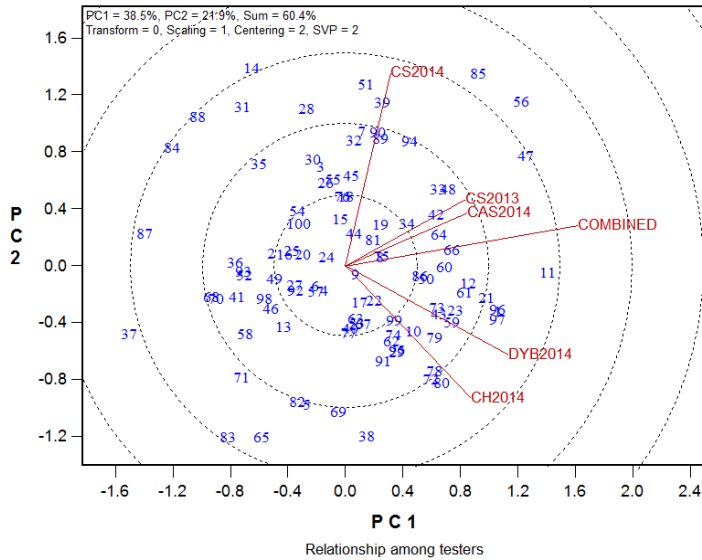


Figure 2.2 Biplot showing the relationship among environments and combined mean (across five environments) for grain yield (GY). Codes for environment are: CH2014= Chillicothe, DYB2014= Diyarbakir, CAS2014= Castroville, CS2014= College Station 2014, CS2013= College Station 2013 and Combined = combined mean. ID for lines are: 1=TX11Vsyn0101; 2=TX11Vsyn0103; 3=TX11Vsyn0110; 4=TX11Vsyn0111; 5=TX11Vsyn0112; 6=TX11Vsyn0113; 7=TX11Vsyn0116; 8=TX11Vsyn0118; 9=TX11Vsyn0119; 10=TX11Vsyn0120; 11=TX11Vsyn0122; 12=TX11Vsyn0123; 13=TX11Vsyn0124; 14=TX11Vsyn0127; 15=TX11Vsyn0130; 16=TX11Vsyn0131; 17=TX11Vsyn0133; 18=TX11Vsyn0134; 19=TX11Vsyn0135; 20=TX11Vsyn0136; 21=TX11Vsyn0137; 22=TX11Vsyn0138; 23=TX11Vsyn0140; 24=TX11Vsyn0146; 25=TX11Vsyn0153; 26=TX11Vsyn0154; 27=TX11Vsyn0156; 28=TX11Vsyn0158; 29=TX11Vsyn0159; 30=TX11Vsyn0160; 31=TX11Vsyn0161; 32=TX11Vsyn0164; 33=TX11Vsyn0165; 34=TX11Vsyn0167; 35=TX11Vsyn0168; 36=TX11Vsyn0169; 37=TX11Vsyn0174; 38=TX11Vsyn0175; 39=TX11Vsyn0178; 40=TX11Vsyn0179; 41=TX11Vsyn0180; 42=TX11Vsyn0182; 43=TX11Vsyn0185; 44=TX11Vsyn0188; 45=TX11Vsyn0189; 46=TX11Vsyn0190; 47=TX11Vsyn0191; 48=TX11Vsyn0195; 49=TX11Vsyn0196; 50=TX11Vsyn0197; 51=TX11Vsyn0199; 52=TX11Vsyn0201; 53=TX11Vsyn0208; 54=TX11Vsyn0211; 55=TX11Vsyn0212; 56=TX11Vsyn0213; 57=TX11Vsyn0216; 58=TX11Vsyn0217; 59=TX11Vsyn0219; 60=TX11Vsyn0225; 61=TX11Vsyn0226; 62=TX11Vsyn0228; 63=TX11Vsyn0229; 64=TX11Vsyn0230; 65=TX11Vsyn0232; 66=TX11Vsyn0234; 67=TX11Vsyn0238; 68=TX11Vsyn0240; 69=TX11Vsyn0241; 70=TX11Vsyn0243; 71=TX11Vsyn0253; 72=TX11Vsyn0261; 73=TX11Vsyn0263; 74=TX11Vsyn0264; 75=TX11Vsyn0265; 76=TX11Vsyn0266; 77=TX11Vsyn0267; 78=TX11Vsyn0271; 79=TX11Vsyn0272; 80=TX11Vsyn0275; 81=TX11Vsyn0277; 82=TX11Vsyn0279; 83=TX11Vsyn0280; 84=TX11Vsyn0282; 85=TX11Vsyn0294; 86=TX11Vsyn0300; 87=TX11Vsyn0303; 88=TX11Vsyn0305; 89=TX11Vsyn0306; 90=TX11Vsyn0308; 91=TX11Vsyn0309; 92=TX11Vsyn0312; 93=TX11Vsyn0313; 95=TAM111; 94=TAM112; 96=TAM113; 97=TAM304

2.3.6.2 GGE biplot for seed weight (SeedWt)

For SeedWt, data is classified into seven sectors and four out of the five environments were grouped into one sector, indicating that this trait was consistent among the environments. Environment CS2013 is classified into a separate sector with genotype 70 (ID) being the best in this sector (Figure 2.3 and Figure 2.4).

2.3.6.3 GGE biplot for seeds per head (SeedsHead⁻¹)

The environments CS2013 and CH2014 were classified into sector 7, CAS2014 into sector 1, and DYB2014 and CS2014 into sector 3 for SeedsHead⁻¹ (Figure 2.5 and Figure 2.6). Genotypes TX11Vsyn0313 (ID = 93), TX11Vsyn0300 (ID = 86), and TX11Vsyn0159 (ID = 29) were the best in sectors 7, 2, and 3, respectively.

2.3.6.4 GGE biplot for heads per square meter (HeadNo)

The variance for HeadNo was lowest in CS2013 followed by CAS2014 and was highest in CS2014 and CH2014 (Figure 2.7 and Figure 2.8). It appears that as the stress level (biotic and abiotic) decreases, the discriminating ability for HeadNo also decreases for the particular environment. This in turn might have affected variability for GY in these environments. Biplot results of HeadNo (Figure 2.8) go hand in hand with the results of GY. As the variance for HeadNo increased variance for GY also increased in each environment. These results are further supported by the correlation coefficients between these two variables as discussed before (refer to table on page 76).

2.3.6.5 GGE biplot for relationship between yield and yield components (5 environments)

Figure 2.9 shows the relationship between GY and its components (SeedWt, SeedsHead⁻¹, and HeadNo). The tester relationship biplot follows similar rules as mentioned for other traits (McDermott and Coe, 2012). Among the three primary yield components of interest, HeadNo and SeedsHead⁻¹ had acute and almost equally sized angles with GY, indicating a positive correlation between each and GY. The increase in any of these factors would increase GY. On the other hand, SeedWt, had an 180 ° angle with GY, pointing to the absence of correlation between the two traits, which is in agreement with Cooper et al., (2012). This confirms results of the Pearson's correlations coefficients (Table 2.14) and path coefficient analysis (Table 2.15).

SeedWt had an obtuse angle with HeadNo and SeedsHead⁻¹. Similarly, HeadNo had an obtuse angle with SeedsHead⁻¹. These results are in accordance with Pearson's correlations coefficients as discussed previously (Table 2.14). These results also agree with Cooper et al., (2012).

Vector length is used to study the variation and discriminating ability of that particular trait (Yan et al., 2003). GY had the shortest vector, followed by SeedWt and equally sized vectors for HeadNo and SeedsHead⁻¹. Therefore, GY had the lowest variation among genotypes, which was followed closely by SeedWt which agrees with Cooper et al., (2012) who also reported that most synthetics have higher SeedWt than check varieties and hence less variation for this trait. The larger variability for

SeedsHead⁻¹ and HeadNo indicates that further improvement can be made by utilizing these two traits.

Figure 2.10 presents the “which won where” view of the biplot that shows which genotypes performed best for a trait or in an environment (McDermott and Coe, 2012). Genotypes 97, 56, 85, and 47 had the highest yield whereas genotypes TX11Vsyn0208 (ID = 53), TX11Vsyn0179 (ID = 40), and TX11Vsyn0228 (ID = 62) had the highest HeadNo across environments. Genotypes TX11Vsyn0174 (ID = 37), TX11Vsyn0219 (ID = 59), and TX11Vsyn0111 (ID = 4) had relatively large SeedWt and genotypes TX11Vsyn0300 (ID = 86), TX11Vsyn0122 (ID = 11), and TX11Vsyn0212 (ID = 55) had the highest SeedsHead⁻¹.

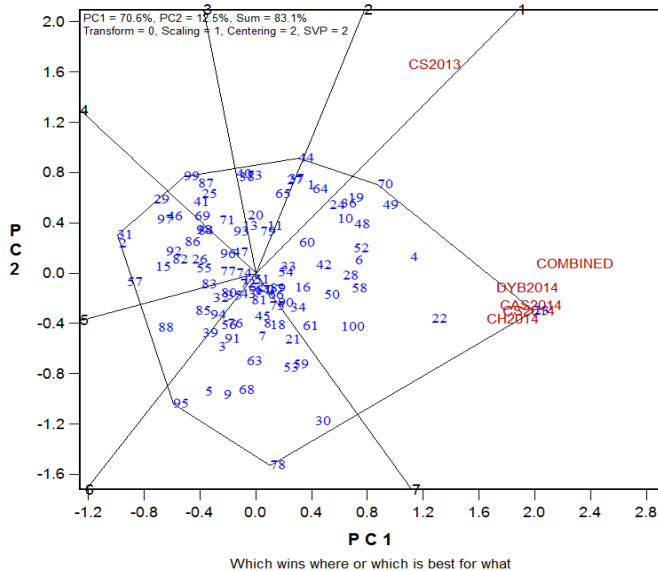


Figure 2.3 GGE biplot showing the best genotypes for single seed weight (SeedWt) in each test environment. Codes for environment are: CH2014= Chillicothe, DYB2014= Diyarbakir, CAS2014= Castroville, CS2014= College Station 2014, CS2013= College Station 2013 and Combined = combined mean. ID for lines are: 1=TX11Vsyn0101; 2=TX11Vsyn0103; 3=TX11Vsyn0110; 4=TX11Vsyn0111; 5=TX11Vsyn0112; 6=TX11Vsyn0113; 7=TX11Vsyn0116; 8=TX11Vsyn0118; 9=TX11Vsyn0119; 10=TX11Vsyn0120; 11=TX11Vsyn0122; 12=TX11Vsyn0123; 13=TX11Vsyn0124; 14=TX11Vsyn0127; 15=TX11Vsyn0130; 16=TX11Vsyn0131; 17=TX11Vsyn0133; 18=TX11Vsyn0134; 19=TX11Vsyn0135; 20=TX11Vsyn0136; 21=TX11Vsyn0137; 22=TX11Vsyn0138; 23=TX11Vsyn0140; 24=TX11Vsyn0146; 25=TX11Vsyn0153; 26=TX11Vsyn0154; 27=TX11Vsyn0156; 28=TX11Vsyn0158; 29=TX11Vsyn0159; 30=TX11Vsyn0160; 31=TX11Vsyn0161; 32=TX11Vsyn0164; 33=TX11Vsyn0165; 34=TX11Vsyn0167; 35=TX11Vsyn0168; 36=TX11Vsyn0169; 37=TX11Vsyn0174; 38=TX11Vsyn0175; 39=TX11Vsyn0178; 40=TX11Vsyn0179; 41=TX11Vsyn0180; 42=TX11Vsyn0182; 43=TX11Vsyn0185; 44=TX11Vsyn0188; 45=TX11Vsyn0189; 46=TX11Vsyn0190; 47=TX11Vsyn0191; 48=TX11Vsyn0195; 49=TX11Vsyn0196; 50=TX11Vsyn0197; 51=TX11Vsyn0199; 52=TX11Vsyn0201; 53=TX11Vsyn0208; 54=TX11Vsyn0211; 55=TX11Vsyn0212; 56=TX11Vsyn0213; 57=TX11Vsyn0216; 58=TX11Vsyn0217; 59=TX11Vsyn0219; 60=TX11Vsyn0225; 61=TX11Vsyn0226; 62=TX11Vsyn0228; 63=TX11Vsyn0229; 64=TX11Vsyn0230; 65=TX11Vsyn0232; 66=TX11Vsyn0234; 67=TX11Vsyn0238; 68=TX11Vsyn0240; 69=TX11Vsyn0241; 70=TX11Vsyn0243; 71=TX11Vsyn0253; 72=TX11Vsyn0261; 73=TX11Vsyn0263; 74=TX11Vsyn0264; 75=TX11Vsyn0265; 76=TX11Vsyn0266; 77=TX11Vsyn0267; 78=TX11Vsyn0271; 79=TX11Vsyn0272; 80=TX11Vsyn0275; 81=TX11Vsyn0277; 82=TX11Vsyn0279; 83=TX11Vsyn0280; 84=TX11Vsyn0282; 85=TX11Vsyn0294; 86=TX11Vsyn0300; 87=TX11Vsyn0303; 88=TX11Vsyn0305; 89=TX11Vsyn0306; 90=TX11Vsyn0308; 91=TX11Vsyn0309; 92=TX11Vsyn0312; 93=TX11Vsyn0313; 95=TAM111; 94=TAM112; 96=TAM113; 97=TAM304; 98=TAM305; 99=TAM401; 100=TAMW101.

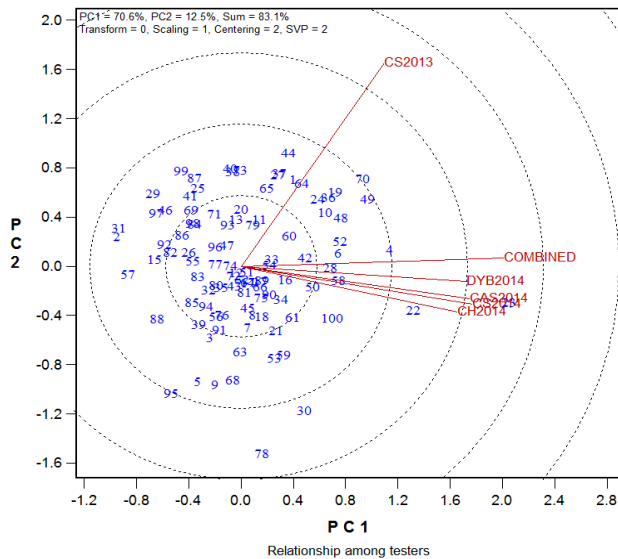


Figure 2.4 Biplot showing the relationship among environments and combined mean (across five environments) for single seed weight (SeedWt). Codes for environment are: CH2014= Chillicothe, DYB2014= Diyarbakir, CAS2014= Castroville, CS2014= College Station 2014, CS2013= College Station 2013 and Combined = combined mean. ID for lines are: 1=TX11Vsyn0101; 2=TX11Vsyn0103; 3=TX11Vsyn0110; 4=TX11Vsyn0111; 5=TX11Vsyn0112; 6=TX11Vsyn0113; 7=TX11Vsyn0116; 8=TX11Vsyn0118; 9=TX11Vsyn0119; 10=TX11Vsyn0120; 11=TX11Vsyn0122; 12=TX11Vsyn0123; 13=TX11Vsyn0124; 14=TX11Vsyn0127; 15=TX11Vsyn0130; 16=TX11Vsyn0131; 17=TX11Vsyn0133; 18=TX11Vsyn0134; 19=TX11Vsyn0135; 20=TX11Vsyn0136; 21=TX11Vsyn0137; 22=TX11Vsyn0138; 23=TX11Vsyn0140; 24=TX11Vsyn0146; 25=TX11Vsyn0153; 26=TX11Vsyn0154; 27=TX11Vsyn0156; 28=TX11Vsyn0158; 29=TX11Vsyn0159; 30=TX11Vsyn0160; 31=TX11Vsyn0161; 32=TX11Vsyn0164; 33=TX11Vsyn0165; 34=TX11Vsyn0167; 35=TX11Vsyn0168; 36=TX11Vsyn0169; 37=TX11Vsyn0174; 38=TX11Vsyn0175; 39=TX11Vsyn0178; 40=TX11Vsyn0179; 41=TX11Vsyn0180; 42=TX11Vsyn0182; 43=TX11Vsyn0185; 44=TX11Vsyn0188; 45=TX11Vsyn0189; 46=TX11Vsyn0190; 47=TX11Vsyn0191; 48=TX11Vsyn0195; 49=TX11Vsyn0196; 50=TX11Vsyn0197; 51=TX11Vsyn0199; 52=TX11Vsyn0201; 53=TX11Vsyn0208; 54=TX11Vsyn0211; 55=TX11Vsyn0212; 56=TX11Vsyn0213; 57=TX11Vsyn0216; 58=TX11Vsyn0217; 59=TX11Vsyn0219; 60=TX11Vsyn0225; 61=TX11Vsyn0226; 62=TX11Vsyn0228; 63=TX11Vsyn0229; 64=TX11Vsyn0230; 65=TX11Vsyn0232; 66=TX11Vsyn0234; 67=TX11Vsyn0238; 68=TX11Vsyn0240; 69=TX11Vsyn0241; 70=TX11Vsyn0243; 71=TX11Vsyn0253; 72=TX11Vsyn0261; 73=TX11Vsyn0263; 74=TX11Vsyn0264; 75=TX11Vsyn0265; 76=TX11Vsyn0266; 77=TX11Vsyn0267; 78=TX11Vsyn0271; 79=TX11Vsyn0272; 80=TX11Vsyn0275; 81=TX11Vsyn0277; 82=TX11Vsyn0279; 83=TX11Vsyn0280; 84=TX11Vsyn0282; 85=TX11Vsyn0294; 86=TX11Vsyn0300; 87=TX11Vsyn0303; 88=TX11Vsyn0305; 89=TX11Vsyn0306; 90=TX11Vsyn0308; 91=TX11Vsyn0309; 92=TX11Vsyn0312; 93=TX11Vsyn0313; 95=TAM111; 94=TAM112; 96=TAM113; 97=TAM304; 98=TAM305; 99=TAM401; 100=TAMW101

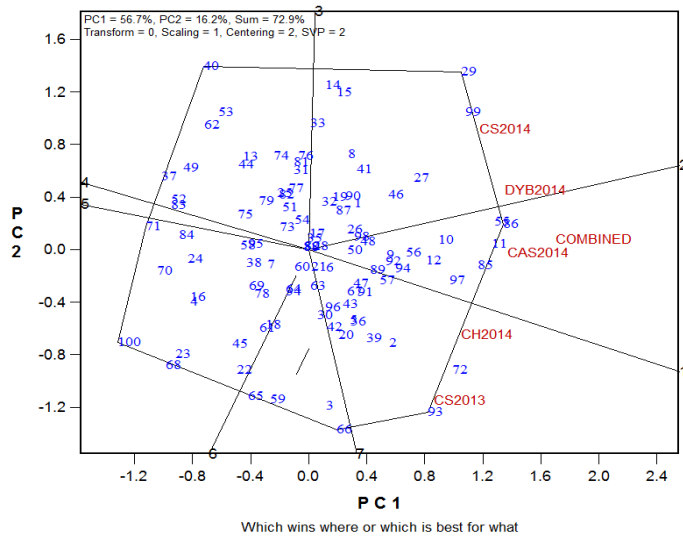


Figure 2.5 GGE biplot showing the best genotypes for seeds per head (SeedsHead⁻¹) in each test environment. Codes for environment are: CH2014= Chillicothe, DYB2014= Diyarbakir, CAS2014= Castroville, CS2014= College Station 2014, CS2013= College Station 2013 and Combined = combined mean. ID for lines are: 1=TX11Vsyn0101; 2=TX11Vsyn0103; 3=TX11Vsyn0110; 4=TX11Vsyn0111; 5=TX11Vsyn0112; 6=TX11Vsyn0113; 7=TX11Vsyn0116; 8=TX11Vsyn0118; 9=TX11Vsyn0119; 10=TX11Vsyn0120; 11=TX11Vsyn0122; 12=TX11Vsyn0123; 13=TX11Vsyn0124; 14=TX11Vsyn0127; 15=TX11Vsyn0130; 16=TX11Vsyn0131; 17=TX11Vsyn0133; 18=TX11Vsyn0134; 19=TX11Vsyn0135; 20=TX11Vsyn0136; 21=TX11Vsyn0137; 22=TX11Vsyn0138; 23=TX11Vsyn0140; 24=TX11Vsyn0146; 25=TX11Vsyn0153; 26=TX11Vsyn0154; 27=TX11Vsyn0156; 28=TX11Vsyn0158; 29=TX11Vsyn0159; 30=TX11Vsyn0160; 31=TX11Vsyn0161; 32=TX11Vsyn0164; 33=TX11Vsyn0165; 34=TX11Vsyn0167; 35=TX11Vsyn0168; 36=TX11Vsyn0169; 37=TX11Vsyn0174; 38=TX11Vsyn0175; 39=TX11Vsyn0178; 40=TX11Vsyn0179; 41=TX11Vsyn0180; 42=TX11Vsyn0182; 43=TX11Vsyn0185; 44=TX11Vsyn0188; 45=TX11Vsyn0189; 46=TX11Vsyn0190; 47=TX11Vsyn0191; 48=TX11Vsyn0195; 49=TX11Vsyn0196; 50=TX11Vsyn0197; 51=TX11Vsyn0199; 52=TX11Vsyn0201; 53=TX11Vsyn0208; 54=TX11Vsyn0211; 55=TX11Vsyn0212; 56=TX11Vsyn0213; 57=TX11Vsyn0216; 58=TX11Vsyn0217; 59=TX11Vsyn0219; 60=TX11Vsyn0225; 61=TX11Vsyn0226; 62=TX11Vsyn0228; 63=TX11Vsyn0229; 64=TX11Vsyn0230; 65=TX11Vsyn0232; 66=TX11Vsyn0234; 67=TX11Vsyn0238; 68=TX11Vsyn0240; 69=TX11Vsyn0241; 70=TX11Vsyn0243; 71=TX11Vsyn0253; 72=TX11Vsyn0261; 73=TX11Vsyn0263; 74=TX11Vsyn0264; 75=TX11Vsyn0265; 76=TX11Vsyn0266; 77=TX11Vsyn0267; 78=TX11Vsyn0271; 79=TX11Vsyn0272; 80=TX11Vsyn0275; 81=TX11Vsyn0277; 82=TX11Vsyn0279; 83=TX11Vsyn0280; 84=TX11Vsyn0282; 85=TX11Vsyn0294; 86=TX11Vsyn0300; 87=TX11Vsyn0303; 88=TX11Vsyn0305; 89=TX11Vsyn0306; 90=TX11Vsyn0308; 91=TX11Vsyn0309; 92=TX11Vsyn0312; 93=TX11Vsyn0313; 95=TAM111; 94=TAM112; 96=TAM113; 97=TAM304; 98=TAM305; 99=TAM401; 100=TAMW101

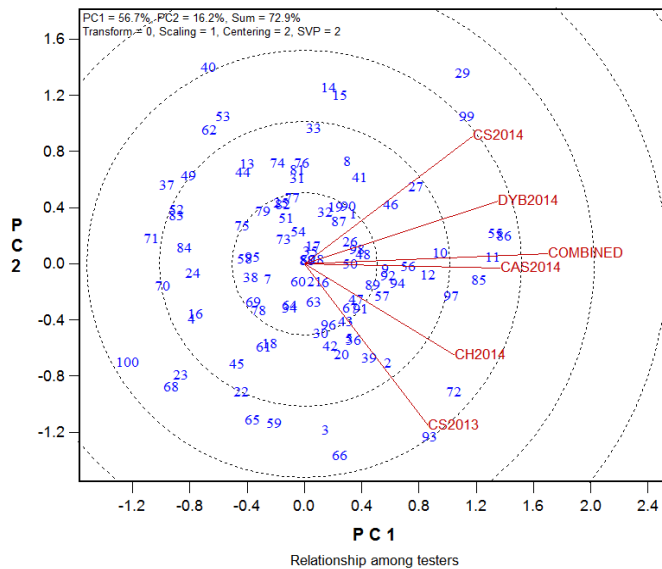


Figure 2.6 Biplot showing the relationship among environments and combined mean (across five environments) for seeds per head (SeedsHead⁻¹). Codes for environment are: CH2014= Chillicothe, DYB2014= Diyarbakir, CAS2014= Castroville, CS2014= College Station 2014, CS2013= College Station 2013 and Combined = combined mean. ID for lines are: 1=TX11Vsyn0101; 2=TX11Vsyn0103; 3=TX11Vsyn0110; 4=TX11Vsyn0111; 5=TX11Vsyn0112; 6=TX11Vsyn0113; 7=TX11Vsyn0116; 8=TX11Vsyn0118; 9=TX11Vsyn0119; 10=TX11Vsyn0120; 11=TX11Vsyn0122; 12=TX11Vsyn0123; 13=TX11Vsyn0124; 14=TX11Vsyn0127; 15=TX11Vsyn0130; 16=TX11Vsyn0131; 17=TX11Vsyn0133; 18=TX11Vsyn0134; 19=TX11Vsyn0135; 20=TX11Vsyn0136; 21=TX11Vsyn0137; 22=TX11Vsyn0138; 23=TX11Vsyn0140; 24=TX11Vsyn0146; 25=TX11Vsyn0153; 26=TX11Vsyn0154; 27=TX11Vsyn0156; 28=TX11Vsyn0158; 29=TX11Vsyn0159; 30=TX11Vsyn0160; 31=TX11Vsyn0161; 32=TX11Vsyn0164; 33=TX11Vsyn0165; 34=TX11Vsyn0167; 35=TX11Vsyn0168; 36=TX11Vsyn0169; 37=TX11Vsyn0174; 38=TX11Vsyn0175; 39=TX11Vsyn0178; 40=TX11Vsyn0179; 41=TX11Vsyn0180; 42=TX11Vsyn0182; 43=TX11Vsyn0185; 44=TX11Vsyn0188; 45=TX11Vsyn0189; 46=TX11Vsyn0190; 47=TX11Vsyn0191; 48=TX11Vsyn0195; 49=TX11Vsyn0196; 50=TX11Vsyn0197; 51=TX11Vsyn0199; 52=TX11Vsyn0201; 53=TX11Vsyn0208; 54=TX11Vsyn0211; 55=TX11Vsyn0212; 56=TX11Vsyn0213; 57=TX11Vsyn0216; 58=TX11Vsyn0217; 59=TX11Vsyn0219; 60=TX11Vsyn0225; 61=TX11Vsyn0226; 62=TX11Vsyn0228; 63=TX11Vsyn0229; 64=TX11Vsyn0230; 65=TX11Vsyn0232; 66=TX11Vsyn0234; 67=TX11Vsyn0238; 68=TX11Vsyn0240; 69=TX11Vsyn0241; 70=TX11Vsyn0243; 71=TX11Vsyn0253; 72=TX11Vsyn0261; 73=TX11Vsyn0263; 74=TX11Vsyn0264; 75=TX11Vsyn0265; 76=TX11Vsyn0266; 77=TX11Vsyn0267; 78=TX11Vsyn0271; 79=TX11Vsyn0272; 80=TX11Vsyn0275; 81=TX11Vsyn0277; 82=TX11Vsyn0279; 83=TX11Vsyn0280; 84=TX11Vsyn0282; 85=TX11Vsyn0294; 86=TX11Vsyn0300; 87=TX11Vsyn0303; 88=TX11Vsyn0305; 89=TX11Vsyn0306; 90=TX11Vsyn0308; 91=TX11Vsyn0309; 92=TX11Vsyn0312; 93=TX11Vsyn0313; 95=TAM111; 94=TAM112; 96=TAM113; 97=TAM304; 98=TAM305; 99=TAM401; 100=TAMW101

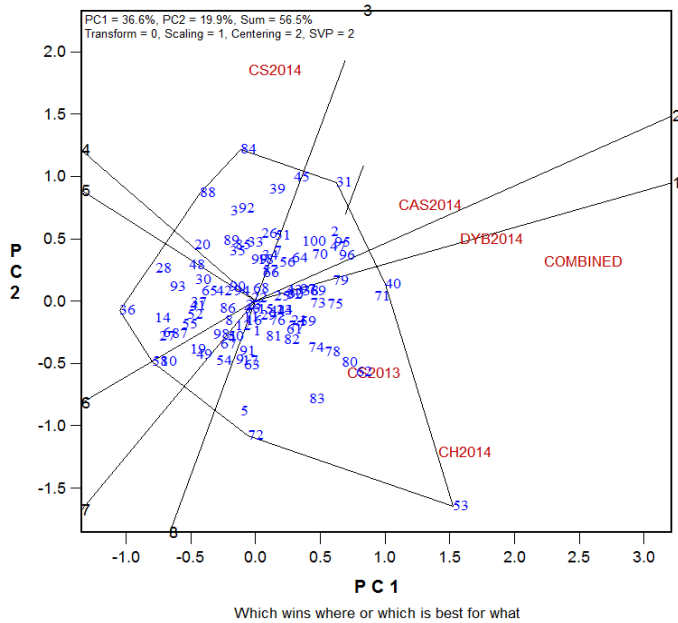


Figure 2.7 GGE biplot showing the best genotypes for heads per square meter (HeadNo) in each test environment. Codes for environment are: CH2014= Chillicothe, DYB2014= Diyarbakir, CAS2014= Castroville, CS2014= College Station 2014, CS2013= College Station 2013 and Combined = combined mean. ID for lines are: 1=TX11Vsyn0101; 2=TX11Vsyn0103; 3=TX11Vsyn0110; 4=TX11Vsyn0111; 5=TX11Vsyn0112; 6=TX11Vsyn0113; 7=TX11Vsyn0116; 8=TX11Vsyn0118; 9=TX11Vsyn0119; 10=TX11Vsyn0120; 11=TX11Vsyn0122; 12=TX11Vsyn0123; 13=TX11Vsyn0124; 14=TX11Vsyn0127; 15=TX11Vsyn0130; 16=TX11Vsyn0131; 17=TX11Vsyn0133; 18=TX11Vsyn0134; 19=TX11Vsyn0135; 20=TX11Vsyn0136; 21=TX11Vsyn0137; 22=TX11Vsyn0138; 23=TX11Vsyn0140; 24=TX11Vsyn0146; 25=TX11Vsyn0153; 26=TX11Vsyn0154; 27=TX11Vsyn0156; 28=TX11Vsyn0158; 29=TX11Vsyn0159; 30=TX11Vsyn0160; 31=TX11Vsyn0161; 32=TX11Vsyn0164; 33=TX11Vsyn0165; 34=TX11Vsyn0167; 35=TX11Vsyn0168; 36=TX11Vsyn0169; 37=TX11Vsyn0174; 38=TX11Vsyn0175; 39=TX11Vsyn0178; 40=TX11Vsyn0179; 41=TX11Vsyn0180; 42=TX11Vsyn0182; 43=TX11Vsyn0185; 44=TX11Vsyn0188; 45=TX11Vsyn0189; 46=TX11Vsyn0190; 47=TX11Vsyn0191; 48=TX11Vsyn0195; 49=TX11Vsyn0196; 50=TX11Vsyn0197; 51=TX11Vsyn0199; 52=TX11Vsyn0201; 53=TX11Vsyn0208; 54=TX11Vsyn0211; 55=TX11Vsyn0212; 56=TX11Vsyn0213; 57=TX11Vsyn0216; 58=TX11Vsyn0217; 59=TX11Vsyn0219; 60=TX11Vsyn0225; 61=TX11Vsyn0226; 62=TX11Vsyn0228; 63=TX11Vsyn0229; 64=TX11Vsyn0230; 65=TX11Vsyn0232; 66=TX11Vsyn0234; 67=TX11Vsyn0238; 68=TX11Vsyn0240; 69=TX11Vsyn0241; 70=TX11Vsyn0243; 71=TX11Vsyn0253; 72=TX11Vsyn0261; 73=TX11Vsyn0263; 74=TX11Vsyn0264; 75=TX11Vsyn0265; 76=TX11Vsyn0266; 77=TX11Vsyn0267; 78=TX11Vsyn0271; 79=TX11Vsyn0272; 80=TX11Vsyn0275; 81=TX11Vsyn0277; 82=TX11Vsyn0279; 83=TX11Vsyn0280; 84=TX11Vsyn0282; 85=TX11Vsyn0294; 86=TX11Vsyn0300; 87=TX11Vsyn0303; 88=TX11Vsyn0305; 89=TX11Vsyn0306; 90=TX11Vsyn0308; 91=TX11Vsyn0309; 92=TX11Vsyn0312; 93=TX11Vsyn0313; 95=TAM111; 94=TAM112; 96=TAM113; 97=TAM304; 98=TAM305; 99=TAM401; 100=TAMW10

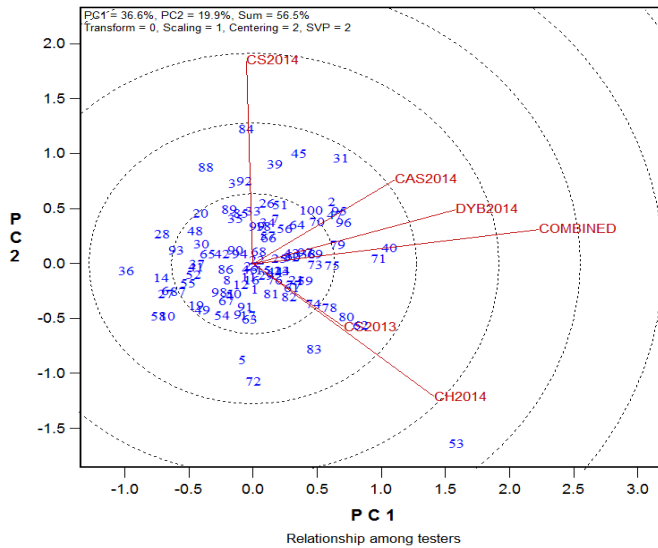


Figure 2.8 Biplot showing the relationship among environments and combined mean (across five environments) for heads per square meter (HeadNo). Codes for environment: CH2014= Chillicothe, DYB2014= Diyarbakir, CAS2014= Castroville, CS2014= College Station 2014, CS2013= College Station 2013 and COMBINED = combined mean. ID for lines are: 1=TX11Vsyn0101; 2=TX11Vsyn0103; 3=TX11Vsyn0110; 4=TX11Vsyn0111; 5=TX11Vsyn0112; 6=TX11Vsyn0113; 7=TX11Vsyn0116; 8=TX11Vsyn0118; 9=TX11Vsyn0119; 10=TX11Vsyn0120; 11=TX11Vsyn0122; 12=TX11Vsyn0123; 13=TX11Vsyn0124; 14=TX11Vsyn0127; 15=TX11Vsyn0130; 16=TX11Vsyn0131; 17=TX11Vsyn0133; 18=TX11Vsyn0134; 19=TX11Vsyn0135; 20=TX11Vsyn0136; 21=TX11Vsyn0137; 22=TX11Vsyn0138; 23=TX11Vsyn0140; 24=TX11Vsyn0146; 25=TX11Vsyn0153; 26=TX11Vsyn0154; 27=TX11Vsyn0156; 28=TX11Vsyn0158; 29=TX11Vsyn0159; 30=TX11Vsyn0160; 31=TX11Vsyn0161; 32=TX11Vsyn0164; 33=TX11Vsyn0165; 34=TX11Vsyn0167; 35=TX11Vsyn0168; 36=TX11Vsyn0169; 37=TX11Vsyn0174; 38=TX11Vsyn0175; 39=TX11Vsyn0178; 40=TX11Vsyn0179; 41=TX11Vsyn0180; 42=TX11Vsyn0182; 43=TX11Vsyn0185; 44=TX11Vsyn0188; 45=TX11Vsyn0189; 46=TX11Vsyn0190; 47=TX11Vsyn0191; 48=TX11Vsyn0195; 49=TX11Vsyn0196; 50=TX11Vsyn0197; 51=TX11Vsyn0199; 52=TX11Vsyn0201; 53=TX11Vsyn0208; 54=TX11Vsyn0211; 55=TX11Vsyn0212; 56=TX11Vsyn0213; 57=TX11Vsyn0216; 58=TX11Vsyn0217; 59=TX11Vsyn0219; 60=TX11Vsyn0225; 61=TX11Vsyn0226; 62=TX11Vsyn0228; 63=TX11Vsyn0229; 64=TX11Vsyn0230; 65=TX11Vsyn0232; 66=TX11Vsyn0234; 67=TX11Vsyn0238; 68=TX11Vsyn0240; 69=TX11Vsyn0241; 70=TX11Vsyn0243; 71=TX11Vsyn0253; 72=TX11Vsyn0261; 73=TX11Vsyn0263; 74=TX11Vsyn0264; 75=TX11Vsyn0265; 76=TX11Vsyn0266; 77=TX11Vsyn0267; 78=TX11Vsyn0271; 79=TX11Vsyn0272; 80=TX11Vsyn0275; 81=TX11Vsyn0277; 82=TX11Vsyn0279; 83=TX11Vsyn0280; 84=TX11Vsyn0282; 85=TX11Vsyn0294; 86=TX11Vsyn0300; 87=TX11Vsyn0303; 88=TX11Vsyn0305; 89=TX11Vsyn0306; 90=TX11Vsyn0308; 91=TX11Vsyn0309; 92=TX11Vsyn0312; 93=TX11Vsyn0313; 95=TAM111; 94=TAM112; 96=TAM113; 97=TAM304; 98=TAM305; 99=TAM401; 100=TAMW101

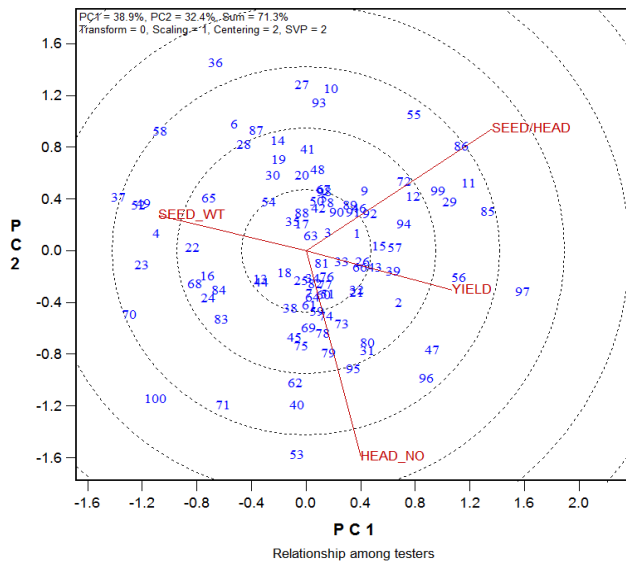


Figure 2.9 Biplot showing relationship among GY (grain yield) and yield components SeedWt (singles seed weight), SeedsHead⁻¹, and HeadNo (heads m⁻²). Codes for traits are: SEED/HEAD = seeds per head, HEAD_NO= Heads m⁻², SEED_WT = single seed weight. ID for lines are: 1=TX11Vsyn0101; 2=TX11Vsyn0103; 3=TX11Vsyn0110; 4=TX11Vsyn0111; 5=TX11Vsyn0112; 6=TX11Vsyn0113; 7=TX11Vsyn0116; 8=TX11Vsyn0118; 9=TX11Vsyn0119; 10=TX11Vsyn0120; 11=TX11Vsyn0122; 12=TX11Vsyn0123; 13=TX11Vsyn0124; 14=TX11Vsyn0127; 15=TX11Vsyn0130; 16=TX11Vsyn0131; 17=TX11Vsyn0133; 18=TX11Vsyn0134; 19=TX11Vsyn0135; 20=TX11Vsyn0136; 21=TX11Vsyn0137; 22=TX11Vsyn0138; 23=TX11Vsyn0140; 24=TX11Vsyn0146; 25=TX11Vsyn0153; 26=TX11Vsyn0154; 27=TX11Vsyn0156; 28=TX11Vsyn0158; 29=TX11Vsyn0159; 30=TX11Vsyn0160; 31=TX11Vsyn0161; 32=TX11Vsyn0164; 33=TX11Vsyn0165; 34=TX11Vsyn0167; 35=TX11Vsyn0168; 36=TX11Vsyn0169; 37=TX11Vsyn0174; 38=TX11Vsyn0175; 39=TX11Vsyn0178; 40=TX11Vsyn0179; 41=TX11Vsyn0180; 42=TX11Vsyn0182; 43=TX11Vsyn0185; 44=TX11Vsyn0188; 45=TX11Vsyn0189; 46=TX11Vsyn0190; 47=TX11Vsyn0191; 48=TX11Vsyn0195; 49=TX11Vsyn0196; 50=TX11Vsyn0197; 51=TX11Vsyn0199; 52=TX11Vsyn0201; 53=TX11Vsyn0208; 54=TX11Vsyn0211; 55=TX11Vsyn0212; 56=TX11Vsyn0213; 57=TX11Vsyn0216; 58=TX11Vsyn0217; 59=TX11Vsyn0219; 60=TX11Vsyn0225; 61=TX11Vsyn0226; 62=TX11Vsyn0228; 63=TX11Vsyn0229; 64=TX11Vsyn0230; 65=TX11Vsyn0232; 66=TX11Vsyn0234; 67=TX11Vsyn0238; 68=TX11Vsyn0240; 69=TX11Vsyn0241; 70=TX11Vsyn0243; 71=TX11Vsyn0253; 72=TX11Vsyn0261; 73=TX11Vsyn0263; 74=TX11Vsyn0264; 75=TX11Vsyn0265; 76=TX11Vsyn0266; 77=TX11Vsyn0267; 78=TX11Vsyn0271; 79=TX11Vsyn0272; 80=TX11Vsyn0275; 81=TX11Vsyn0277; 82=TX11Vsyn0279; 83=TX11Vsyn0280; 84=TX11Vsyn0282; 85=TX11Vsyn0294; 86=TX11Vsyn0300; 87=TX11Vsyn0303; 88=TX11Vsyn0305; 89=TX11Vsyn0306; 90=TX11Vsyn0308; 91=TX11Vsyn0309; 92=TX11Vsyn0312; 93=TX11Vsyn0313; 95=TAM111; 94=TAM112; 96=TAM113; 97=TAM304; 98=TAM305; 99=TAM401; 100=TAMW101

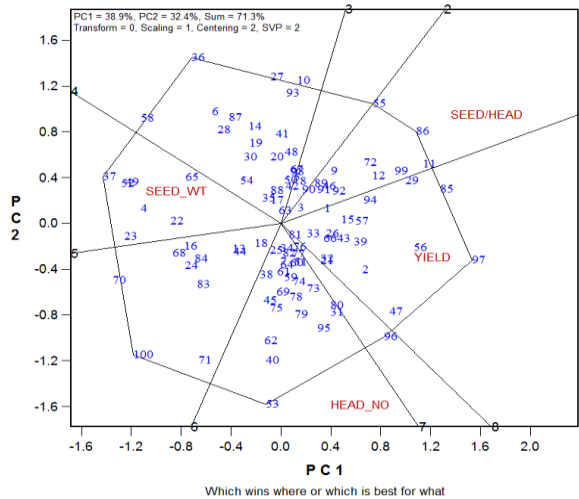


Figure 2.10 Biplot showing best genotypes for GY (grain yield) and yield components SeedWt (single seed weight), SeedsHead⁻¹, and HeadNo (heads m⁻²). Codes for traits are: SEED/HEAD = seeds per head, HEAD_NO= heads m⁻², SEED_WT = single seed weight. ID for lines are:

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 96=TAM113; 97=TAM304; 98=TAM305; 99=TAM401; 100=TAMW101

Table 2.14 Pearson correlation coefficients among GY (grain yield), TW (test weight), SeedWt (Single seed weight), SeedsHead⁻¹, HeadNo (heads m⁻²), HS (heading score), and HT (height) based on combined mean values of five environments

	GY	TW	SeedWt	SeedHead⁻¹	HeadNo	HS	HT
GY		0.22*	0.12 ^{ns}	0.41**	0.31**	-0.26**	-0.15 ^{ns}
TW			0.38**	-0.11 ^{ns}	-0.03 ^{ns}	0.01 ^{ns}	0.05 ^{ns}
SeedWt				-0.42**	-0.30**	-0.15 ^{ns}	0.42**
SeedsHead ⁻¹					-0.32**	0.12 ^{ns}	-0.06 ^{ns}
HeadNo						-0.29**	-0.36**
HS							0.21*
HT							

* Significant at the 0.05 probability level; ** Significant at the 0.01 probability level
ns not significant

Table 2.15 Estimates of direct and indirect effect of yield components on grain yield at five environments and combined mean of five environments

Trait	Seed Wt.	Head No.	Seed Head⁻¹	Yield
<u>Combined (five environments)</u>				
SeedWt	0.76	-0.24	-0.43	<u>0.09</u>
HeadNo	-0.22	0.83	-0.31	<u>0.31</u>
SeedsHead ⁻¹	-0.32	-0.26	1.00	<u>0.41</u>
<u>CAS2014</u>				
SeedWt	0.64	-0.29	-0.14	<u>0.21</u>*
HeadNo	-0.20	0.93	-0.19	<u>0.54</u>**
SeedsHead ⁻¹	-0.13	-0.24	0.71	<u>0.34</u>**
<u>CH2014</u>				
SeedWt	0.30	0.01	-0.06	<u>0.26</u>**
HeadNo	0.01	0.95	-0.14	<u>0.56</u>**
SeedsHead ⁻¹	-0.03	-0.18	0.57	<u>0.36</u>**
<u>DYB2014</u>				
SeedWt	0.77	-0.24	-0.41	<u>0.12</u>^{ns}
HeadNo	-0.18	1.02	-0.56	<u>0.28</u>**
SeedsHead ⁻¹	-0.29	-0.53	1.08	<u>0.26</u>**
<u>CS2014</u>				
SeedWt	0.47	-0.23	-0.20	<u>0.04</u>^{ns}
HeadNo	-0.11	0.95	-0.03	<u>0.81</u>**
SeedsHead ⁻¹	-0.18	-0.06	0.53	<u>0.29</u>**
<u>CS2013</u>				
SeedWt	0.30	-0.43	-0.15	<u>-0.28</u>**
HeadNo	-0.12	1.06	-0.05	<u>0.89</u>**
SeedsHead ⁻¹	-0.11	-0.11	0.44	<u>0.22</u>*

* Significant at the 0.05 probability level; ** Significant at the 0.01 probability level ; ns not significant

Codes for environments are CAS2014 = Castroville, CH2014 = Chillicothe, Combined = across five environments, CS2014 = College Station, and DYB2014 = Diyarbakir

2.3.6.6 Stability analysis using GGE biplot (5 environments)

Presence of G*E interaction in the study affects the usefulness of the genotypes by confounding genotype performance (Kang and Pham, 1991), which also necessitates estimating stability, repeatability, and heritability of the particular trait (Becker and Leon, 1988)

The yield stability of each genotype across five environments was tested using the average environmental coordination (AEC) (Yan et al., 2003). A line known as the average environmental axis is drawn through the center of the biplot and average of principle components (PC1 and PC2) of all environments. The AEC axis with double end arrow separates below average and above average GY means. In our study genotypes 9 to 11 are the above average yielding and genotypes TX11Vsyn0175 (ID = 38) to TX11Vsyn0174 (ID = 37) are the below average yielding ones (Figure 2.11). The AEC axis with one direction arrow in the direction of highest GY mean is used to determine the best genotypes with high mean and good stability. Any genotype with a short vector (stable) and furthest to the right of the biplot in direction of the arrow (high mean) on AEC axis would have high performance and broad adaptation for the given trait (refer to figure in page 80). Genotypes with high GY mean and long vectors are best suited for specific environments. Environments are further classified into two groups; group A consisted of environments CS2013, CAS2014, and CS2014 and group B consisted of environments DYB2014 and CH2014. Genotype TX11Vsyn0191 (ID = 47) was the best in group A due to its high GY mean and good stability. Genotype TX11Vsyn0213 (ID = 56) and TX11Vsyn0294 (ID = 85) are good for specific environments within group B.

Apart from TX11Vsyn0191 (ID = 47), genotypes TX11Vsyn0158 (ID = 28), TX11Vsyn0182 (ID = 42), TX11Vsyn0230 (ID = 64) had good stability but have lower GY mean compared to TX11Vsyn0191 (ID = 47). Genotype TX11Vsyn0122 (ID = 11) was the best in group B due to its high GY mean and good stability. Genotypes TX11Vsyn0234 (ID = 66) and TX11Vsyn0225 (ID = 60) had low GY mean but high stability (Figure 2.11 and Table 2.16). These lines might be good for low yielding environments with highly unpredictable yield levels.

In conclusion, environments that are better discriminating and have closer angles to the average environmental are good for selecting generally adapted germplasm. Discriminating but non-representative test environments are useful for selecting specifically adapted genotypes if the target environment divided into mega environments (Yan et al., 2003; McDermott and Coe, 2012).

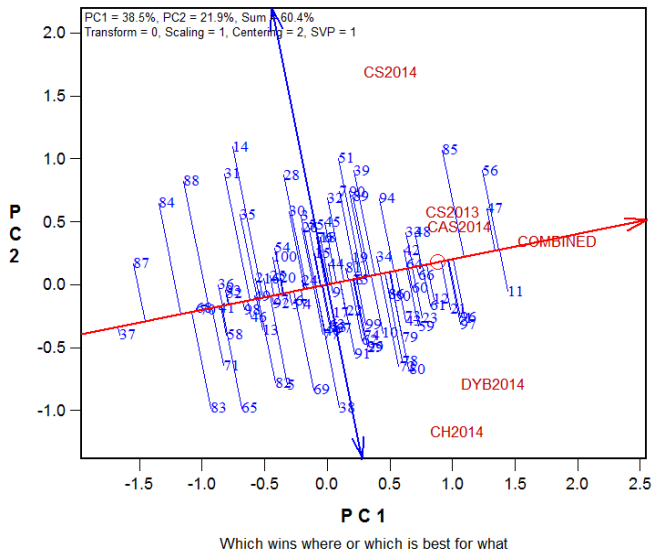


Figure 2.11 GGE biplot showing environment discrimination ability, genotype mean performance and stability. Codes for environment are: CH2014=Chillicothe, DYB2014=Diyarbakir, CAS2014=Castroville, CS2014= College Station-I, CS2013= College Station-II, and COMBINED = combined mean across five environments. ID for lines are: 1=TX11Vsyn0101; 2=TX11Vsyn0103; 3=TX11Vsyn0110; 4=TX11Vsyn0111; 5=TX11Vsyn0112; 6=TX11Vsyn0113; 7=TX11Vsyn0116; 8=TX11Vsyn0118; 9=TX11Vsyn0119; 10=TX11Vsyn0120; 11=TX11Vsyn0122; 12=TX11Vsyn0123; 13=TX11Vsyn0124; 14=TX11Vsyn0127; 15=TX11Vsyn0130; 16=TX11Vsyn0131; 17=TX11Vsyn0133; 18=TX11Vsyn0134; 19=TX11Vsyn0135; 20=TX11Vsyn0136; 21=TX11Vsyn0137; 22=TX11Vsyn0138; 23=TX11Vsyn0140; 24=TX11Vsyn0146; 25=TX11Vsyn0153; 26=TX11Vsyn0154; 27=TX11Vsyn0156; 28=TX11Vsyn0158; 29=TX11Vsyn0159; 30=TX11Vsyn0160; 31=TX11Vsyn0161; 32=TX11Vsyn0164; 33=TX11Vsyn0165; 34=TX11Vsyn0167; 35=TX11Vsyn0168; 36=TX11Vsyn0169; 37=TX11Vsyn0174; 38=TX11Vsyn0175; 39=TX11Vsyn0178; 40=TX11Vsyn0179; 41=TX11Vsyn0180; 42=TX11Vsyn0182; 43=TX11Vsyn0185; 44=TX11Vsyn0188; 45=TX11Vsyn0189; 46=TX11Vsyn0190; 47=TX11Vsyn0191; 48=TX11Vsyn0195; 49=TX11Vsyn0196; 50=TX11Vsyn0197; 51=TX11Vsyn0199; 52=TX11Vsyn0201; 53=TX11Vsyn0208; 54=TX11Vsyn0211; 55=TX11Vsyn0212; 56=TX11Vsyn0213; 57=TX11Vsyn0216; 58=TX11Vsyn0217; 59=TX11Vsyn0219; 60=TX11Vsyn0225; 61=TX11Vsyn0226; 62=TX11Vsyn0228; 63=TX11Vsyn0229; 64=TX11Vsyn0230; 65=TX11Vsyn0232; 66=TX11Vsyn0234; 67=TX11Vsyn0238; 68=TX11Vsyn0240; 69=TX11Vsyn0241; 70=TX11Vsyn0243; 71=TX11Vsyn0253; 72=TX11Vsyn0261; 73=TX11Vsyn0263; 74=TX11Vsyn0264; 75=TX11Vsyn0265; 76=TX11Vsyn0266; 77=TX11Vsyn0267; 78=TX11Vsyn0271; 79=TX11Vsyn0272; 80=TX11Vsyn0275; 81=TX11Vsyn0277; 82=TX11Vsyn0279; 83=TX11Vsyn0280; 84=TX11Vsyn0282; 85=TX11Vsyn0294; 86=TX11Vsyn0300; 87=TX11Vsyn0303; 88=TX11Vsyn0305; 89=TX11Vsyn0306; 90=TX11Vsyn0308; 91=TX11Vsyn0309; 92=TX11Vsyn0312; 93=TX11Vsyn0313; 95=TAM111; 94=TAM112; 96=TAM113; 97=TAM304; 98=TAM305; 99=TAM401; 100=TAMW101

Table 2.16 Mean grain yield (GY), test weight (TW), single seed weight (SeedWt), seeds per head (SeedsHead⁻¹), heads per square meter (HeadNo), plant height (HT), heading score (HS) of top ten yielding synthetic derived wheat lines and check varieties across five environments

ID	Genotype	GY	TW	SeedWt	Seeds Head ⁻¹	HeadNo	HT	HS
		t ha ⁻¹	kg hL ⁻¹	mg	count	Heads m ⁻²	cm	scale 1- 5
56	TX11Vsyn0213	3.46	71.8	30.6	39	272	81.6	2.4
11	TX11Vsyn0122	3.46	72.7	32.3	42	246	88.4	3.2
47	TX11Vsyn0191	3.44	76.8	31.1	37	297	85.4	2.5
85	TX11Vsyn0294	3.36	69.9	29.4	42	254	81.9	2.6
1	TX11Vsyn0101	3.31	73.2	33.9	37	250	88.9	2.8
21	TX11Vsyn0137	3.27	72.9	32.9	35	269	84.3	3.8
12	TX11Vsyn0123	3.22	72.0	31.3	40	243	89.3	2.3
48	TX11Vsyn0195	3.22	71.5	35.8	37	226	94.0	2.7
61	TX11Vsyn0226	3.19	72.7	33.7	33	268	84.1	2.1
33	TX11Vsyn0165	3.19	71.1	32.9	35	257	90.5	2.8
95	TAM111	3.10	73.6	30	38	251	84.4	3.1
94	TAM112	2.98	70.3	28.4	33	291	75.8	2.2
96	TAM113	3.38	73.5	29.6	35	302	78.6	2.9
97	TAM304	3.36	69.7	26.7	41	283	73.4	2.4
98	TAM305	2.58	71.8	28.6	37	235	75.8	2.4
99	TAM401	3.01	71.1	29.3	41	249	85.6	2.3
100	TAMW101	2.71	75.3	34.9	26	290	75.6	3.1
	Mean	2.89	71.9	31.8	35	254	85.6	2.8
	Minimum	2.19	68.7	26.6	26	186	73.4	1.8
	Maximum	3.46	76.8	42.4	43	338	98.8	4.3

3. GENETIC PARAMETERS AND RESPONSE TO INDIRECT SELECTION FOR GRAIN YIELD IN SYNTHETIC DERIVED WHEAT LINES

3.1 Introduction

A number of studies have reported that the rate of genetic gain for grain yield in wheat (*Triticum aestivum* L.) has drastically decreased since the onset of the Green Revolution (Graybosch and Peterson, 2010; Gill et al., 2004; Patrignani et al., 2014). The rate of genetic gain varied from 0.1% in poor to 2-3% in irrigated environments (Pingali and Rajaram, 1999). Globally, grain yield of wheat has been increasing at annual rate of 1%. However, grain yield has to increase at an annual rate of 2% to meet the needs of the growing world's population (Gill et al., 2004).

Crop scientists around the world have been exploring new ways to improve the genetic gain for grain yield, including the use of wheat wild relatives. Synthetic hexaploid wheat (SHW) developed by hybridizing *Triticum turgidum* L. and *Aegilops tauschii* has contributed to improving the genetic diversity in wheat. A number of studies have reported synthetic wheat's have contributed new source of resistance to biotic stresses and tolerance to abiotic tolerance (Mujeeb Kazi et al, 2000a, 2000b, 2001a, 2001b, 2001c) and it is believed that synthetic might have contributed certain set of alleles for yield potential. Although, a number of studies show the utility of synthetic

wheat in breeding programs, few studies have been conducted to understand the genetic parameters associated with SHW and synthetic derived wheat (SDW).

The efficiency of selection depends on the magnitude and nature of the genetic variation present in a population (Farshadfa et al., 2013) and without genetic variation, the progress of a trait is impractical. A detailed study to understand the nature and magnitude of the genetic variation present in a set of SDW populations is of importance for planning ways to improve genetic gain for grain yield in the U.S. Great Plains and other wheat production areas around the world. A number of biometrical methods are available to determine the genetic variation present in any population of a crop (Wolie et al., 2013).

Besides genetic variability, many other factors are important for the success of a breeding program. For instance, knowledge of heritability of a trait plays a significant role by discerning the reliability of the phenotypic value as a guide to the genotypic value. Knowledge of narrow-sense (h^2) and broad-sense heritability (H^2) are very important in determining the selection procedure for the trait, for predicting gain from selection, and determining the relative importance of genetic effects.

Genetic advance (GA), a.k.a. the breeder's equation, indicates the magnitude of expected genetic gain (GG) from one cycle of selection at certain selection intensity (i) (Singh, 2001). A high heritability value along with high genetic gain is the rule of thumb that a breeder follows while making selection. Therefore, it is important to estimate GG, H^2 , and genetic and phenotypic variations for any trait that is intended for improvement.

The ultimate objective of most plant breeding programs is to increase grain yield per se without jeopardizing end-use quality and tolerance to biotic and abiotic stresses. However, direct selection for grain yield is more laborious and time consuming, as large numbers of advanced lines need to be evaluated in multiple years at multiple locations. Moreover, grain yield is poorly heritable and highly influenced by genotype-by-environment (G*E) interaction (Reynolds et al., 1999). This has compelled researchers to identify new ways that can assist in improving grain yield potential in wheat. Many studies have experimented with different ways of conducting indirect selection for grain yield. Babar et al., (2007) have conducted studies to use spectral reflectance as indirect selection criteria to select for higher grain yield. Similarly, Gutierrez et al. (2011) have used canopy temperature as a selection criterion for higher grain yield. However, few studies have investigated the efficiency of indirect selection for grain yield via yield components such as numbers of heads for square meter (HeadNo), seeds per head (SeedsHead⁻¹), and single seed weight (SeedWt).

Cooper et al. (2012, 2013) conducted multiple studies on SDW populations in the BC₁F₄ and BC₁F₅ generations. They reported that grain yield (GY) had a moderate correlation with HeadNo (0.54) and SeedsHead⁻¹ (0.53). Among all yield and yield components, SeedWt (0.32) was the most heritable followed by SeedsHead⁻¹ (0.25), and HeadNo (0.15) but the heritability for grain yield per se was poor in these studies. Therefore, we hypothesized that indirect selection for HeadNo and SeedsHead⁻¹ could increase grain yield in SDW in spite of yield compensatory effects. The objectives of this study were to 1) understand the genetic and phenotypic variability, heritability, and

genetic gain in SDW for yield, its components, and other important agronomic traits and
2) determine the efficiency of indirect selection for grain yield using components such as
HeadNo and SeedsHead⁻¹

3.2 Materials and methods

3.2.1 Plant material

Germplasm used in this study was developed by backcrossing selected CIMMYT (International Maize and Wheat Improvement Center) SHW from Elite I and Elite II set to Texas A&M AgriLife Research hard red winter wheat varieties, TAM 111 and TAM 112. Breeders at Texas A&M AgriLife Research had developed many SDW populations by hybridizing eight SHW from Elite I set and two SHW from Elite II set to TAM 111 and 112. Based on agronomic, morphological, and biotic stress tolerance heads were carefully chosen from selected SDW populations and advanced to head-rows (BC₁F_{5:6}) and later generations. We started with 321 SDW lines as head-rows in 2011. Based on HeadNo and SeedsHead⁻¹ a set of 213 lines were selected from the head-rows generation and advanced to BC₁F_{5:7} and later generations. For this chapter, only BC₁F_{5:8} and BC₁F_{5:9} generations data were used for calculations.

3.2.2 Experimental design and testing locations

In 2013, the set of 213 lines were laid out in an augmented design at Colleges Station, TX (latitude = 30.5°N, longitude = 96.4°W). The following year, a selected set of 93 lines from the 213 lines were laid out in an alpha lattice design with two

replications at Castroville (CAS), TX (latitude = 29.35°N, longitude = 98.88°W), Chillicothe (CH), X (latitude = 34.2°N, longitude = 99.5°W), Diyarbakir (DYB), Turkey (latitude = 38.1422 °N, longitude = 40.2711 °E), and College Station, TX.

3.2.3 Traits recorded

The traits measured or estimated were grain yield (GY) in t ha⁻¹, test weight (TW) in kg hL⁻¹, HeadNo in heads m⁻², SeedsHead⁻¹, SeedWt in mg, plant height (HT) in cm, and heading score (HS) using a scale of 1 to 5; where 1 = very early heading, 2 = early heading, 3 = medium heading, 4 = late heading, 5 = very late heading.

3.2.4 Statistical analysis

Individual analysis of variance for each environment was done using Proc GLM model of SAS 9.4.

3.2.5 Genetic parameter estimates

The genotypic (σ_g^2) and phenotypic (σ_p^2) variances for individual environments (Formula 3.1 to 3.3) were estimated using the methods described by Fehr (1989), where MS_g = mean square for genotype, MS_{g*e} = mean square for genotype-by-environment interaction, MS_{err} = mean square for error, r = replication, and e= environment. Variance components were determined for traits such as GY, TW, SeedWt, SeedsHead⁻¹, HeadNo, HT, and HS.

Broad sense heritability (H^2) for individual environments (Formula 3.4) was estimated as described by Falconer (1989) and Holland et al. (2003). Formula 3.4a was used to estimate heritability values in the alpha lattice experiment. Formula 3.4b was used to estimate heritability values in the augmented design experiment. Heritability estimates were conducted on entry mean basis.

Realized heritability (h_R^2) was determined for GY and HeadNo as described by Guthrie et al. (1984). These calculations were done using the mean of 10% highest and 10% lowest yielding lines from BC₁F_{5;8} and observing their response in the BC₁F_{5;9} generation using formula 3.5.

Genetic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) were estimated using formulae 3.6 and 3.7, respectively (Johnson et al., 1955), where, σ_g^2 = genotypic variance, σ_p^2 = phenotypic variance, and \bar{X} = mean value of a particular trait.

Genetic gain (GG) was calculated using formula 3.8 as suggested by Allard (1960), where, K = constant at 10% selection intensity, σ_p = phenotypic standard deviation, and h^2 = narrow-sense heritability of the trait. Genetic gain mean (GGM) was determined using formula 3.9 as described by Johnson et al. (1955), where, GG = genetic gain and \bar{X} = mean value of a particular trait.

Phenotypic correlation coefficient (r_p) and genotypic correlation coefficient (r_g) were determined using formulae 3.10 and 3.11, respectively (Miller et al., 1958), where, $COV_{p(x,y)}$ = phenotypic covariance between traits x and y, $\sigma_{p(x)}^2$ = phenotypic variance

for x, $\sigma_{g(y)}^2$ = phenotypic variance for y, $COV_{g(x,y)}$ = genetic covariance between x and y, $\sigma_{g(x)}^2$ = genotypic variance for trait x, and $\sigma_{g(y)}^2$ = genotypic variance for trait y.

Expected response (R) to direct selection for a primary trait such as GY was calculated using formula 3.12 (Falconer, 1996), where, h_x^2 = narrow sense heritability of the primary trait (GY), $\sqrt{\sigma_{p(x)}^2}$ = phenotypic standard deviation for primary trait, $i = 10\%$ selection intensity expressed as standardized units.

Formula 3.13 was used to calculate the correlated response (CR) in the primary trait (GY) resulted from the selection for a secondary trait (HeadNo or SeedsHead⁻¹ or SeedWt) as described by Falconer (1996). The components of this formula are described as follow: h_x = square root of narrow sense heritability of the primary trait, h_y = square root of narrow sense heritability of secondary trait, r_g = genetic correlation between primary and secondary traits, $\sqrt{\sigma_{p(x)}^2}$ = phenotypic standard deviation of primary trait, and $i = 10\%$ selection intensity expressed as standardized units.

The relative efficiency ($\frac{CR}{R}$) of indirect selection (using HeadNo or SeedWt or SeedsHead⁻¹) for GY versus direct selection for GY was calculated using formula 3.14 (Falconer, 1996), where, h_y = square root of heritability of secondary trait (HeadNo or SeedsHead⁻¹ or SeedWt), h_x = square root of heritability of primary trait, r_g = genetic correlation between primary and secondary traits.

The rationale for doing indirect selection was that in BC₁F₅ generation HeadNo had a highest correlation (0.54) with the GY and higher heritability (0.15) than the grain

yield (-0.21). From this study, we also came to know that SeedWt (0.30) had higher heritability than SeedsHead⁻¹ and HeadNo. Therefore, we proposed optimizing either HeadNo or SeedsHead⁻¹ would increase GY in spite of yield compensatory effects. Hence, this study was conducted to understand the efficiency of indirect selection for GY via the use of yield components (HeadNo or SeedsHead⁻¹ or SeedWt). As yield is a complex quantitative trait, any method for predicting it early on in the pipeline would help to speed the breeding process.

Individual variances:

Genetic variance(σ_g^2):
$$\frac{MS_g - MS_{err}}{r}$$
 ...Formula 3.1

Error variance(σ_{err}^2):
$$MS_{err}$$
 ... Formula 3.2

Phenotypic variance(σ_p^2):
$$\sigma_g^2 + \sigma_{err}^2$$
Formula 3.3

Heritability (H^2) alpha lattice:
$$\left(\frac{\sigma_g^2}{\sigma_g^2 + \frac{\sigma_{err}^2}{r}} \right)$$
 ...Formula 3.4a

Heritability (H^2) augmented design:
$$\frac{\sigma_g^2}{\sigma_p^2}$$
 ...Formula 3.4b

Realized heritability(h_R^2):
$$\frac{BC_1F_{5:9} \text{ high} - BC_1F_{5:9} \text{ low}}{BC_1F_{5:8} \text{ high} - BC_1F_{5:8} \text{ low}}$$
 ...Formula 3.5

Genotypic coefficient of variation (GCV):
$$\frac{\sqrt{\sigma_g^2}}{\bar{X}} \times 100$$
Formula 3.6

Phenotypic coefficient of variation (PCV):
$$\frac{\sqrt{\sigma_p^2}}{\bar{X}} \times 100$$
Formula 3.7

Genetic gain (GG):	$(K) \sigma_p (h^2)$Formula 3.8
Genetic gain mean (GGM):	$\frac{GG}{\bar{X}} \times 100$Formula 3.9
Phenotypic correlation coefficient (r_p):	$\frac{COV_{p(x,y)}}{\sqrt{\sigma_{p(x)}^2 \sigma_{p(y)}^2}}$Formula 3.10
Genotypic correlation coefficient (r_g):	$\frac{COV_{g(x,y)}}{\sqrt{\sigma_{g(x)}^2 \sigma_{g(y)}^2}}$ Formula 3.11
Response to selection (R):	$i h_x^2 \sqrt{\sigma_{p(x)}^2}$Formula 3.12
Correlated response to selection (CR):	$i h_y h_x r_g \sqrt{\sigma_{p(x)}^2}$Formula 3.13
Efficiency of selection:	$\frac{CR}{R} \quad \frac{h_y}{h_x} r_g$ Formula 3.14

3.3 Results and discussion

3.3.1 Analysis of variance

Analysis of variance for individual environments showed significant differences among genotypes for most of the traits in this study, except for GY in CAS2014 and CS2014 (refer to tables on page 36 to 40). This suggested that there was a considerable amount of genetic variability among the SDW lines. These 93 SDW lines were selected from 12 populations with different pedigrees.

3.3.2 Estimation of heritability

Heritability values of more than 0.8 are considered high, values 0.79 - 0.4 medium, and less than 0.39 low (Singh et al., 2001). Wolie et al. (2013) indicated that it is easy to select for and advance traits possessing high as opposed to poor heritability values due to the high environmental influence on the latter.

Values of H^2 for individual environments and across five environments are presented on page 97 and 98. Mean values across five environments indicated that HS (0.88), SeedWt (0.82), HT (0.82), and TW (0.84) were highly heritable, SeedsHead⁻¹ (0.55) was moderately heritable, and GY (0.38) and HeadNo (0.14) were poorly heritable (Table 3.1). Most of these results are in accordance with the studies conducted by Meena et al. (2013). Moderate to high heritability for HT, HS, and SeedWt were reported by Hokrani et al. (2013). Considering the range of environments where trials were conducted and with additional stress factors (drought, leaf and stripe rust, root rot, and lodging) in different environments, it was expected to see the trends of this nature for these traits. In addition, traits that are associated with reproductive fitness are likely to have low heritability (Falconer, 1961). In most of the individual environments, HeadNo had the lowest, SeedsHead⁻¹ moderate, and SeedWt had the highest heritability values.

Realized heritability (h_R^2) accounts for the additive genetic variation that is transmitted to the subsequent generation and is the true heritability that is of most practical value to breeders (Kharkwal and Jain, 2004). Falconer (1989) indicated that low heritability values are associated with low additive genetic variation and high

phenotypic variation. In contrary to this statement, Visscher et al. (2008) pointed out that low heritability doesn't necessarily mean low additive variance. Low heritability might be the result of small proportion of variation caused by genotype out of all the observed variation. High phenotypic variation can arise for multiple reasons such as gene mutation, selection pressure, and environmental factors. Realized heritability calculated in this study indicated that GY was poorly to moderately heritable with a range of 0.29 to 0.55 values. HeadNo was classified as low to moderately heritable with a range of 0.37 to 0.76 values and SeedsHead⁻¹ was classified as moderately to highly heritable with a range of 0.44 to 0.82 values. SeedWt was also classified as moderately to highly heritable with a range of 0.47 to 0.83 values (Table 3.2). These realized heritability values were determined at 10% selection intensity. Based on the moderately heritability values for most of these traits, it seems there is good scope for improving each of these traits.

3.3.3 Estimation of variance components

Yield and its components in wheat are quantitative in nature and are influenced by environmental conditions (Wu et al., 2012). Therefore, understanding the phenotypic and genotypic variations present in the population by studying the PCV and GCV can be useful in estimating the scope for improvement by selection (Wolie et al., 2013). This would in turn help in improving the efficiency of breeding programs by redesigning the selection processes. A breeder's main objective is to have a high percentage of GCV that provides more choice for selection. However, a certain percent of variation could be

attributed to environmental factors, which cannot be inherited. Therefore, determining the percentage of environmental influence on visible variation is very critical. The ultimate objective of most breeding programs is to make incremental increases in yield potential while maintaining end-use quality and tolerance to biotic and abiotic stresses. Therefore, most breeding programs desire to have high GCV values with minimal difference between PCV and GCV. The results that are presented in tables on page 97 to 102 show \bar{X} , σ_g^2 , σ_p^2 , H^2 , GCV, PCV, GG, and GGM for each trait in each individual environment (Table 3.3, Table 3.4, Table 3.5, Table 3.6, and Table 3.7) and also across the five environments (Table 3.8). Since there weren't any standardized statistical methods to combine the alpha lattice design studies with an augmented design, we determined the mean values across five environments by weighed mean analysis. Having more environments and replications increased the accuracy of estimation for these traits.

Combined analysis of the five environments showed that GCV/PCV ratio for SeedWt (96%) was highest, followed by TW (95%), HS (91%), HT (85%), SeedsHead⁻¹ (63%), GY (47%), and HeadNo (31%) (Table 3.8). The highest values indicated that environment had least influence on the expression of these traits and prospects of gain from selection were very high for these traits and vice versa for lower value traits. High influence of the environment fades one's ability to see and select for the variation that is heritable and hence results in low selection gains. Similar results were reported by Meena et al. (2014) and Abinasa et al. (2011). Based on studies conducted in groundnut by Deshmukh et al. (1986), PCV and GCV values more than 20% were considered to be high, values between 10-20% to be moderate, and values less than 10% to be low.

Table 3.1 Mean, genotypic variance (σ_g^2), phenotypic variance (σ_p^2), error variance (σ_{err}^2) and heritability (H^2) for GY (grain yield), TW (test weight), SeedWt (single seed weight), SeedsHead⁻¹, HeadNo (heads m⁻²), HT (plant height), and HS (heading score) under different environments

Trait	GY t ha⁻¹	TW kg hL⁻¹	SeedWt Mg	SeedsHead⁻¹ count	HeadNo Heads m⁻²	HT Cm	HS scale 1-5
<u>CAS2014</u>							
Mean	3.45	78.6	34.3	36	289	90.6	3
σ_g^2	0.01	4.0	1.49x10 ⁵	16	223	42.1	0.98
σ_{err}^2	0.67	0.9	0.34X10 ⁵	14	6346	1.7	0.21
σ_p^2	0.69	4.9	0.00001.824	30	6569	43.8	1.19
H^2	0.04	0.9	0.90	0.70	0.07	0.98	0.90
<u>CH2014</u>							
Mean	1.01	71.1	26.1	22	181	58.8	2
σ_g^2	0.06	3.3	0.39 X10 ⁵	6	1062	13.3	0.20
σ_{err}^2	0.03	1.8	0.42 X10 ⁵	11	5422	13.1	0.07
σ_p^2	0.09	5.1	0.81 X10 ⁵	16	6484	26.4	0.27
H^2	0.81	0.79	0.65	0.51	0.28	0.67	0.85
<u>DYB2014</u>							
Mean	5.33	83.3	34.0	38	431	93.8	3
σ_g^2	0.13	1.2	1.16 X10 ⁵	15	350	53.8	0.53
σ_{err}^2	0.49	0.7	0.66 X10 ⁵	38	9109	50.2	0.42
σ_p^2	0.62	1.9	1.83 X10 ⁵	53	9460	104	0.95
H^2	0.34	0.63	0.78	0.44	0.07	0.68	0.72
<u>CS2014</u>							
Mean	1.88	70.6	28.8	43	158	87.2	3
σ_g^2	0.04	4.46	0.79 X10 ⁵	13	16	32.5	0.78
σ_{err}^2	0.23	0.92	0.43 X10 ⁵	28	2018	1.6	0.10
σ_p^2	0.27	5.38	1.22 X10 ⁵	41	2034	34.1	0.87
H^2	0.26	0.91	0.79	0.47	0.02	0.98	0.94

Table 3.1 Continued

Trait	GY	TW	SeedWt	SeedsHead ⁻¹	HeadNo	HT	HS
<u>CS2013</u>							
Mean	2.65	76.1	36.1	37	203	99	3
σ_g^2	0.2699	36.6684	21.60 X10 ⁵	20.6	1135	80.14	1.413
σ_{err}^2	0.3429	0.6616	0.21 X10 ⁵	12.55	3110	20.68	0
σ_p^2	0.6128	37.33	21.82 X10 ⁵	33.15	4245	100.82	1.413
H^2	0.44	0.98	0.99	0.62	0.27	0.79	1
<u>Combined</u>							
Mean	2.86	75.94	31.9	35.2	252	85.9	2.80
σ_g^2	0.10	9.93	5.1X10 ⁵	14.0	557	44.4	0.78
σ_{err}^2	0.35	1.00	0.4 X10 ⁵	20.8	5201	17.4	0.16
σ_p^2	0.45	10.93	5.5 X10 ⁵	34.8	5758	61.8	0.94
H^2	0.38	0.84	0.82	0.55	0.14	0.82	0.88

Codes for environments are CAS2014 = Castroville, CH2014 = Chillicothe, Combined = across four environments, CS2014= college station, and DYB2014 = Diyarbakir

Table 3.2 Realized heritability (h_R^2) values for grain yield (GY), heads per square meter (HeadNo), seeds per head (SeedHead⁻¹), and single seed weight (SeedWt) for CS2014 (College Station), CAS2014 (Castroville), CH2014 (Chillicothe), DYB2014 (Diyarbakir)

	GY	HeadNo	SeedsHead ⁻¹	SeedWt
CS2013-CS2014	0.32	0.37	0.82	0.64
CS2013-CAS2014	0.49	0.61	0.78	0.83
CS2013-CH2014	0.29	0.58	0.44	0.47
CS2013-DYB2014	0.55	0.76	0.84	0.63

However, based on studies conducted in wheat, Abinasa et al. (2011) have reported values more than 10% were considered to be high and values less than 5% to be low. For this study GCV values more than 20% were recorded for HS (31%) and SeedWt (22%), 10 to 20% for GY (11%) and SeedsHead⁻¹ (11%), 5 to 10% for HeadNo (9%), HT (8%), and less than 5% were documented for TW (4%) (Table 3.8). Selections for HeadNo and SeedsHead⁻¹ were done at the BC₁F₅ and in BC₁F_{5:8} generations, and this might have resulted in moderate to low genetic variation for these traits. Similarly, multiple selections for medium plant height were done from early to advanced generations. This might have reduced the available amount of genetic variation for HT. The PCV and GCV values for TW were less than 5%, suggesting that it is very difficult to improve this trait through plant breeding. Similar results were reported by Abinasa et al. (2011). The PCV values for HeadNo (30%), GY (24%) and SeedWt (23%) were considered very high. These results indicated that there was a broad range of diversity for these traits. A greater proportion of the variation for HeadNo and GY was caused by the environmental coefficient of variation (ECV). In conclusion, there seems to be ample amount of heritable variation in the SDW lines that can be utilized to improve yield and its components via both direct and indirect selection.

Table 3.3 Estimates of genotypic (σ_g^2) and phenotypic (σ_p^2) variance, genotypic (GCV) and phenotypic coefficient of variation (PCV), heritability (H^2), genetic gain (GG) and genetic gain as percent mean (GGM) for Castroville, TX in 2014 at 10% selection intensity (K=1.76). Where, GY = grain yield, TW = test weight, SeedWt = Single seed weight, SeedsHead⁻¹ = Seeds per head, HeadNo = heads per square meter, HT = plant height, HS = heading score

Trait	Mean	σ_g^2	σ_p^2	H^2	GCV (%)	PCV (%)	GG	GGM (%)
GY	3.45	0.014	0.68	0.04	3.4	24.0	0.06	1.7
TW	78.6	4	5.0	0.9	2.6	2.8	3.5	4.5
SeedWt	34.3	1.5 X10 ⁵	1.8 X10 ⁵	0.9	11.2	12.5	0.68	19.7
SeedHead ⁻¹	36	16	30	0.7	11	15	7	18.9
HeadNo	289	223	6569	0.07	5	28	9	3.2
HT	90.6	42.1	43.8	0.98	7.2	7.3	11.4	12.6
HS	3	1	1.2	0.9	33.9	37.4	1.7	59.4

Table 3.4 Estimates of genotypic (σ_g^2) and phenotypic (σ_p^2) variance, genotypic (GCV) and phenotypic coefficient of variation (PCV), heritability (H^2), genetic gain (GG) and genetic gain as percent mean (GGM) for Chillicothe, TX in 2014 at 10% selection intensity (K=1.76). Where, GY = grain yield, TW = test weight, SeedWt = Single seed weight, SeedsHead-1 = Seeds per head, HeadNo = Heads per square meter, HT = Plant height, HS = Heading score

Trait	Mean	σ_g^2	σ_p^2	H^2	GCV (%)	PCV (%)	GG	GGM (%)
GY	1.01	0.058	0.072	0.81	23.9	26.6	0.38	37.7
TW	71.1	3.3	4.2	0.79	2.5	2.9	2.8	4.0
SeedWt	26.1	0.4 X10 ⁵	0.6 X10 ⁵	0.65	7.5	9.3	0.28	10.6
SeedHead ⁻¹	22	6	11	0.51	10.8	15	3	13.6
HeadNo	181	1062	3773	0.28	18	34	30	16.8
HT	58.8	13.3	19.8	0.67	6.2	7.6	5.2	8.9
HS	2	0.2	0.2	0.85	21.0	22.8	0.7	34.2

Table3.5 Estimates of genotypic (σ_g^2) and phenotypic (σ_p^2) variance, genotypic (GCV) and phenotypic coefficient of variation (PCV), heritability (H^2), genetic gain (GG) and genetic gain as percent mean (GGM) for Diyarbakir, Turkey in 2014 at 10% selection intensity (K=1.76). Where, GY = grain yield, TW = test weight, SeedWt = Single seed weight, SeedsHead⁻¹ = Seeds per head, HeadNo = heads per square meter, HT = plant height, HS = heading score

Trait	Mean	σ_g^2	σ_p^2	H^2	GCV (%)	PCV (%)	GG	GGM (%)
GY	5.33	0.13	0.37	0.34	6.7	11.4	0.37	6.9
TW	83.3	1.2	1.6	0.63	1.7	2.0	1.4	2.2
SeedWt	34.0	1.2 X10 ⁵	1.5 X10 ⁵	0.78	10.0	11.4	0.53	15.6
SeedHead ⁻¹	38	15	34	0.44	10.2	15.4	4	11.8
HeadNo	431	350	4905	0.07	4.3	16.3	9	2.0
HT	93.8	53.8	78.9	0.68	7.8	9.5	10.7	11.4
HS	3	0.5	0.7	0.72	24.8	29.3	1.1	37.0

Table 3.6 Estimates of genotypic (σ_g^2) and phenotypic (σ_p^2) variance, genotypic (GCV) and phenotypic coefficient of variation (PCV), heritability (H^2), genetic gain (GG) and genetic gain as percent mean (GGM) for College Station, TX in 2014 at 10% selection intensity (K=1.76). Where, GY = grain yield, TW = test weight, SeedWt = Single seed weight, SeedsHead⁻¹ = Seeds per head, HeadNo = heads per square meter, HT = plant height, HS = heading score

Trait	Mean	σ_g^2	σ_p^2	H^2	GCV (%)	PCV (%)	GG	GGM (%)
GY	1.88	0.04	0.15	0.26	10.6	20.9	0.18	9.53
TW	70.6	4.5	4.9	0.91	2.9	3.1	3.5	5.01
SeedWt	28.8	0.8 X10 ⁵	1.0 X10 ⁵	0.79	9.7	10.9	0.44	15.18
SeedHead ⁻¹	43	13	27	0.47	8.4	12.3	4	10.21
HeadNo	158	16	1025	0.02	2.5	20.3	1	0.55
HT	87.2	32.5	33.3	0.98	6.5	6.6	9.9	11.36
HS	3	0.8	0.8	0.94	30.5	31.4	1.5	52.13

Table 3.7 Estimates of genotypic (σ_g^2) and phenotypic (σ_p^2) variance, genotypic (GCV) and phenotypic coefficient of variation (PCV), heritability (H^2), genetic gain (GG) and genetic gain as percent mean (GGM) for College Station, TX in 2013 at 10% selection intensity (K=1.76). Where, GY = grain yield, TW = test weight, SeedWt = Single seed weight, SeedsHead⁻¹ = Seeds per head, HeadNo = heads per square meter, HT = plant height, HS = heading score

Trait	Mean	σ_g^2	σ_p^2	H^2	GCV (%)	PCV (%)	GG	GGM (%)
GY	2.86	0.27	0.61	0.44	19.6	29.6	0.61	22.9
TW	71.8	36.7	37.3	0.98	8.0	8.0	10.6	13.9
SeedWt	32.0	21.6 X10 ⁵	21.8X10 ⁵	0.99	40.7	40.9	25.7	71.3
SeedHead ⁻¹	35	21	33	0.62	12.2	15.5	6	16.9
HeadNo	252	1135	4245	0.27	16.6	32.2	31	15.1
HT	85.9	80.1	100.8	0.79	9.0	10.1	14.0	14.2
HS	2.8	1.4	1.4	1.00	35.8	35.8	2.1	63.0

Table 3.8 Mean genotypic variance (σ_g^2), phenotypic variance (σ_p^2), genotypic (GCV) and phenotypic coefficient of variation (PCV), heritability (H^2), genetic gain (GG) and genetic gain as percent mean (GGM) across five environments at 10% selection intensity (K=1.76). Where, GY = grain yield, TW = test weight, SeedWt = Single seed weight, SeedsHead⁻¹ = Seeds per head, HeadNo = heads per square meter, HT = plant height, HS = heading score

Trait	Mean	σ_g^2	σ_p^2	H^2	GCV (%)	PCV (%)	GG	GGM (%)
GY	2.86	0.10	0.45	0.38	11.14	23.52	0.45	15.64
TW	75.9	9.9	10.9	0.84	4.15	4.35	4.9	6.45
SeedWt	31.9	5.09 X10 ⁵	5.50 X10 ⁵	0.82	22.38	23.28	11.0	33.67
Seed Head ⁻¹	35	14	35	0.55	10.63	16.76	6	16.16
HeadNo	252	557	5758	0.14	9.35	30.06	19	7.51
HT	85.9	44.4	61.8	0.82	7.76	9.15	11.3	13.21
HS	2.8	0.8	0.9	0.88	31.53	34.59	1.5	53.69

3.3.4 Estimation of genetic gain

Burton et al. (1952) reported that determining heritability along with GCV would help with the reliable estimation of the amount of genetic gain that is possible through phenotypic selection. Heritability values are of less practical importance without the knowledge of the amount of genetic gain, especially while making a selection based on phenotypic appearance. Johnson et al. (1955) indicated that high heritability values are not necessarily associated with high genetic gain, and, therefore, advised to consider genetic gain values along with GCV and heritability values simultaneously in a logical breeding program.

Mean heritability values across five environments had a range of 0.14 - 0.88 and genetic gain mean (GGM) had a range of 5.8% - 50.8% at 10% selection intensity (refer to the table on page 104). Bello et al (2012) stated that high heritability along with high genetic gain indicates additive genetic variance might be governing the trait under consideration and that high heritability with low genetic gain indicates non-additive genetic effect might be governing the trait. They also indicated that low heritability along with low genetic gain indicates non-additive genetic effect might be governing the trait. Among all traits, HS and SeedWt had the highest heritability values along with high GGM. This suggests the presence of additive genetic effects for these traits, which makes it relatively easy to fix these traits in the early generations. Similar results were observed in the individual environments (Table 3.3, Table 3.4, Table 3.5, Table 3.6, and Table 3.7). These results are in accordance with some of the results reported by Shah (1998). On the other hand, HeadNo exhibited low heritability (0.14) and low GGM

(5.8%) in the combined environments analysis. Almost similar results were observed in individual environments. For those traits that had low heritability and GGM, indirect selection based on the secondary traits might be more efficient than direct selection, as long as the assumptions of indirect selection are satisfied. Indirect selection is appropriate when progress through direct selection is slow, hard and less efficient such as in the case of HeadNo and GY. Grain yield had low heritability with moderate genetic gain. SeedsHead⁻¹ had moderate heritability and moderate GGM values. Similar results for SeedsHead⁻¹ were reported by Rana et al. (1999) and Meena et al. (2014).

The main objective of most plant breeding programs is to create plant types with improved traits to increase the overall productivity of economical products with the minimal use of resources and time. In order to do so, a good understanding of the genetic material at hand is very critical. This study gave us an idea of available genetic variation, heritability, and percentage of genetic gain that can be achieved for each of these traits.

Based on the multi-environment analysis, GY can be increased by 15.6% at 10% selection intensity ($K = 1.76$) in these SDW lines. Estimated genetic gain for grain yield at 10% selection was 0.45 t ha⁻¹. This indicated that whenever we selected best 10% of lines in one generation the resulting progenies would have their mean yield increased by 0.45 t ha⁻¹. In other words, GY mean was increased from 2.86 t ha⁻¹ to 3.31 t ha⁻¹ across generations due to the culling of poorer types that do not have merit for advance. Same rules apply for other traits as well. Mean SeedWt value increased from 0.032 to 0.043 g, seedHead⁻¹ from 35 to 41, TW from 75.9 to 80.8 kg ha⁻¹, HeadNo from 252 to 271

heads m⁻², HT from 85.9 to 97.2 cm, and HS increased from 3 to 4 days (Table 3.8).

Mean HT and HS can be increased or decreased depending on the target environment.

3.3.5 Estimation of response to selection, correlated response to selection and efficiency of indirect selection

The efficiency of direct selection (R) for a trait is equivalent to 1.00, as selection is based on grain yield per se. When the $\frac{CR}{R}$ or $\left(\frac{\text{Response to indirect selection}}{\text{Response to direct selection}}\right)$ is less than 1.00, indirect selection is considered to be less efficient than direct selection for the primary trait per se. In contrast, when the $\frac{CR}{R}$ is more than 1.00, indirect selection is considered to be more efficient than direct selection. Often times, indirect selection for GY is never as efficient as direct selection per se (Gallais, 1984).

Conner and Hartl (2003) have reported indirect selection and CR are not the same but they are closely related. They have also stated “Indirect selection occurs within a generation and is caused by phenotypic correlation (r_p), while correlated response occurs across generations and is caused by genotypic correlations (r_g)”. Genotypic correlation values are derived from phenotypic correlation values after eliminating environmental correlation values. In most of the studies, r_g are very close to r_p values. Waitt and Levin (1998) conducted a study to determine the relationship between r_g and r_p correlation using 4000 data points collected from 27 plant species for 40 years. Based on this study, Waitt and Levin have reported 94% of matrix correlations between r_g and r_p were similar at significance level 0.05. Therefore, use of r_p to calculate correlated response and efficiency of indirect selection does gives very close estimates as

calculating these values using r_g . Many other studies utilized r_p for the calculation of indirect selection. In wheat, Cooper et al. (1997) have also shown that r_p can be used to determine the efficiency of indirect selection. In this study, r_p was used instead of r_g to calculate CR and $(\frac{CR}{R})$. As r_g and r_p are closely linked the CR and $(\frac{CR}{R})$ values should not have changed very much.

In this study, indirect selection for HeadNo was not as efficient as direct selection for GY per se. Similar results were reported for SeedWt. Except for one instance, almost similar results were reported for SeedsHead⁻¹. The Mean $(\frac{CR}{R})$ across five environment for HeadNo, SeedWt, and SeedHead⁻¹ were at 0.41, 0.46 and 0.21, respectively (Table 3.9). These results are in accordance with Gallais's (1984) statement "indirect selection is rarely as efficient as direct selection for the primary trait".

There might be multiple reasons for low efficiency for indirect selection for HeadNo. Among them, the relatively low heritability might have played a significant role. Although the correlation between HeadNo and GY seems to be between low to moderate (chapter II results) heritability of the secondary was no better than the primary trait. Mean H^2 for GY across five environments was at 0.38; however; for HeadNo it was only 0.14 (Table 3.8). Gallais (1984) stated that indirect selection is more efficient than direct selection only when the secondary trait had higher heritability than the primary one and there is high genetic correlation between the two traits. This does not rule out the option of utilizing HeadNo as indirect selection component because indirect selection can still be practiced as it is easy to score for HeadNo and it is more time

efficient than selecting for GY per se. From this study, we can say that HeadNo is 41% as efficient as directly selecting for GY.

Results showed the efficiency of indirect selection for SeedHead⁻¹ was higher than HeadNo possibly because of a balance between heritability and correlation. Although correlation was low (chapter II results), heritability (0.55) was moderate for SeedHead⁻¹ (Table 3.9).

The efficiency of indirect selection for SeedWt remains to be lowest among the three components. Although the heritability of this trait is high (0.82), its correlation with grain yield was very low (chapter II results).

Based on results presented in Table 3.9, indirect selection for grain yield components (HeadNo, SeedsHead⁻¹, and SeedWt) is not as efficient as direct selection for GY per se. However, chances for increasing the efficiency of indirect selection using yield components is not ruled out. Based on simulation models developed in ryegrass, Conaghan et al. (2008) reported efficiency of indirect selection can be increased by increasing the number of replications and increasing selection intensity (*i*) by including more number of genotypes but essentially testing the same number of genotypes at similar selection intensities.

Table 3.9 Correlated response (CR) in grain yield for indirect selection based on HeadNo (heads m⁻²), SeedsHead⁻¹, and SeedWt (single seed weight) at 10% selection intensity (K=1.76), and relative efficiency (CR/R) of indirect selection to direct selection for grain yield

Environment	Mean Yield (t ha ⁻¹)	Yield	HeadNo (heads m ⁻²)		SeedsHead ⁻¹		SeedWt (g)	
		R (%)	CR (%)	CR/R	CR (%)	CR/R	CR (%)	CR/R
CAS2014	3.45	1.23	0.84	0.69	1.73	1.41	1.21	0.99
CH2014	1.01	37.69	12.48	0.33	10.79	0.29	8.77	0.23
DYB2014	5.33	6.85	0.88	0.13	2.01	0.29	1.24	0.18
CS2014	1.88	9.47	1.86	0.20	0.67	0.07	0.66	0.07
CS2013	2.65	22.87	15.88	0.69	5.98	0.26	-9.62	-0.42
Mean	2.86	15.62	6.39	0.41	4.24	0.46	0.45	0.21
Mean <i>H</i> ²	0.38	0.38	0.14		0.55		0.82	

4. RESPONSE OF SYNTHETIC DERIVED HARD RED WINTER WHEAT LINES TO BIOTIC STRESSES IN THE GREAT PLAINS OF USA

4.1 Introduction

The U.S. is the third largest wheat (*Triticum aestivum* L.) producing (46 thousand MT) and top wheat exporting country (32 thousand MT) (FAO STAT, 2013; USDA-NASS, 2013) with largest share of total production coming from winter wheat. Winter wheat production (33 thousand MT) accounts for 72% and hard red winter wheat (16 thousand MT) accounts for 36% of the total wheat production in the USA (USDA-NASS, 2013).

The Great Plains, where wheat is constantly exposed to a wide range of biotic and abiotic stresses (Liu et al., 2014), is an important wheat-growing region in the U.S. Rust is the most important and economical fungal disease in the U.S. Great Plains and worldwide (Kolmer et al., 2009, Wegulo and Byamukama, n.d.). Leaf rust (LR; *Puccinia triticina* Erikss. & Henn.), stripe rust (YR; *Puccinia striiformis* Westend. f. sp. *Triticici*), and stem rust (SR; *Puccinia graminis* Pers. f. sp. *Triticici*) are the three types of rust diseases that attack wheat (Kolmer et al., 2009, 2013). In the U.S. Great Plains, LR is the most common of the three rust fungal pathogens followed by YR and SR, respectively (Kolmer et al., 2009). Wegulo and Byamukama reported that YR used to be a common

disease in the Great Plains, but its incidence in mid and southern Great Plains has been on the rise since 2000.

Depending on stage and severity of the infection, LR can cause yield loss anywhere from trace level to over 40% under most favorable conditions (Bowden, n.d.). Yield losses caused by YR can be much more severe than LR at a similar level of infection (Basnet, 2012). Wellings (2011) recorded up to 60% yield loss due to YR on susceptible spring wheat varieties. Some other studies reported losses of up to 40% and 76% under farmers and experimental field conditions (Wegulo and Byamukama, n.d.).

Texas is in the forefront of *Puccinia* Pathway in the U.S. (Stakman, 1934). Rust urediniospores that are carried to northern and eastern states usually overwinter in the southern parts of Texas (Kolmer et al., 2009). Therefore, planting wheat fields in Texas with rust resistant varieties can eventually decrease the distribution and severity of rust diseases in northern parts of the U.S. ‘TAM 111’ and ‘TAM 112’ are the two most widely cultivated hard red winter wheat varieties in the U.S. Great Plains and Texas (Reddy et al., 2014). TAM 112 scored susceptible to naturally occurring races of LR and YR in the region. TAM 111 was also susceptible to LR but scored resistant or moderately resistant to YR until a new race of YR caused a major epidemic in 2012 (Basnet, 2012). Many breeding programs in the U.S. are utilizing race-specific major genes to alleviate the epidemic of LR and YR. However, race specific resistant genes are likely to be defeated within a short period with the rise of new virulent races (Kolmer et al., 2009).

Singh et al., (2010) reported that pyramiding four to five of the slow-rusting race non-specific minor genes can result in a genotype that has near the immune level of resistance to rust diseases. Therefore, many wheat-breeding programs have been looking for race non-specific resistant genes to build durable rust resistance. Some of the studies have shown few accessions of synthetic hexaploid wheat (SHW) are resistant to LR and YR (Mujeeb-Kazi et al., 2000; Mujeeb-Kazi and Delgado, 2001a) and seem to have both race-specific and race non-specific resistant genes (Zegeye, et al., 2014). Therefore, hybridizing the Great Plains wheat germplasm with SHW should improve rust resistance in the region.

Greenbug (*Schizaphis graminum* Rondani) is a major economical pest in the U.S. Great Plains. To date, eleven biotypes (biotype A to biotype K) have been identified (Weng et al., 2004). Among these, biotypes E and I are the most predominant in the southern Great Plains and cause significant yield losses in wheat and grain sorghum (*sorghum bicolor* L.) (Porter et al., 1997). Host plant resistance is the most economical and eco-friendly approach to control greenbug infection (Punnuri et al., 2012). A number of greenbug resistant genes originated from different sources *Gb₁* (durum wheat), *Gb₂* and *Gb₆* (rye), *Gb₅* (*Aegilops speltoides*), *Gb_{x1}*, *Gb_a*, *Gb_b*, *Gb_c*, *Gb_d*, *Gb_z*, *Gb₃*, and *Gb_{x2}* (*Aegilops tauschii*) (Azhaguvel et al., 2012) have been identified. Among all these genes, *Gb₃* has shown high level of resistance to biotype C, E, H, I, and K (Weng and Rudd, 2015). Scientists at Texas A&M AgriLife Research have identified SNP markers that are closely linked to *Gb₃* gene (Liu et al., 2014).

Mujeeb-Kazi et al. (2000a, 2000b, 2001a) documented that some accessions of SHW are resistant to greenbug, LR, and YR. Some of these SHW from CIMMYT have been hybridized with elite wheat varieties of Texas A&M AgriLife Research wheat breeding program, with an objective to introduce new useful genetic diversity into the breeding program. The objectives of this study are to 1) characterize SDW lines for common biotic stresses in the U.S. Great Plains and 2) validate the greenbug resistance present in SDW lines with known markers for *Gb3* gene.

4.2 Materials and methods

4.2.1 Diseases

4.2.1.1 Germplasm

The plant materials used in this study were developed by hybridizing and backcrossing a selected set of SHW from Elite-I and Elite-II sets to Texas A&M AgriLife Research elite varieties, TAM 111 and TAM 112. Germplasm was advanced using bulk and modified bulk methods until BC₁F₅. Selections for best plant type were done in the BC₁F₂ and BC₁F₃ generations. In BC₁F₅ generation, heads were selected from agronomically superior plants with low levels of disease pressure. A total of 321 synthetic derived wheat (SDW) lines from 12 populations were planted as head-rows (BC₁F_{5:6}) at Chillicothe (CH), TX (latitude = 34.2°N, longitude = 99.5°W) in 2011. A set of 213 lines were selected based on heads per square meter (HeadNo) and seeds per head (SeedsHead⁻¹). These 213 lines were planted in one meter (BC₁F_{5:7}) rows in a randomized complete block design at Bushland (BD), TX (latitude = 35.2°N, longitude =

102.1°W) and Castroville (CAS), TX. (latitude = 29.35°N, longitude = 98.88°W). In 2013, same 213 lines (BC₁F_{5.8}) were planted as yield plots in College Station (CS), TX (latitude = 30.5°N, longitude = 96.4°W). In 2014, a selected set of 93 lines were planted as yield plots in multiple environments. These 93 lines were selected from 12 populations with diverse *Aegilops tauschii* parentage in the pedigree (Table 4.1).

4.2.1.2 Field trials, scoring, and statistical analysis

In 2013, one replication of 213 SDW lines was planted as yield plots in an augmented design. In 2014, a selected set of 93 SDW lines from these 213 lines was planted as yield plots. Two replications of these 93 lines were laid out in an alpha-lattice design. More details on experimental designs and experimental setup can be found in chapter II materials and methods. Although, each generation was planted at multiple locations, only one location in each generation had a damageable level of disease pressure. Rust scoring was done at CS in 2013 and CAS in 2014. Scoring for LR was done around mid-late April in CS trial (CS2013). The incidence of YR in the CS2013 trial was not high enough to be documented. In the CAS2014 trial, scoring for LR was done around mid-April and for YR around late March.

Field evaluations were done with naturally occurring races of rust pathogen that are prevalent in TX. The LR races that show virulence to *Lr24*, *Lr17*, *Lr21*, and *Lr39/Lr41* are the most common races in the southern Great Plains (USDA-CDL). The YR races virulent to *Yr9* and *Yr8* are the most common in the U.S. (Kolmer et al., 2009). Scoring for LR and YR was done with different scoring methods. A visual score of 0 to

9 was recorded based on the cleanliness of the leaves. Simultaneously, Modified Cobb's scale based on disease severity and infection type was also used (Peterson et al., 1948).

The host response to infection was scored as resistant (R), moderately resistant (MR), moderately susceptible (MS), and susceptible (S). Coefficient of infection (CI) was calculated by multiplying infection type (R=0.2, MR=0.4, M=0.6, MS=0.8, S=1.0) with disease severity as described by Roelfs et al. (1992). All statistical analysis, including mean values and Pearson's correlation coefficients, were performed using SAS 9.4 PROC GLM procedure.

Screening for powdery mildew (*Erysiphe graminis* f. sp. *Tritici*) resistance was done with naturally occurring races under field conditions. Disease severity scoring for powdery mildew was done using a scale of 0-9 where 0 = no disease and 9 = more than 90% of plant surface covered with powdery mildew (Bennett and Westcott, 1982; Cufner, 2015).

4.2.2 Insect pest (Greenbug)

4.2.2.1 Plant material

Phenotypic screening for greenbug resistance was done on the same set of 213 SDW lines as described in the rust studies.

Table 4.1 Primary Synthetic wheat and their associated *Aegilops tauschii* accession, synthetic derived wheat populations, pedigrees and number of lines that were evaluated in each of these families under field trials in 2013 and 2014

Pop.ID	<i>Aegilops tauschii</i> Accession	Name (Synthetic hexaploid wheat)	Name (Synthetic Derived Wheat)	Pedigree (Synthetic Derived Wheat)	Number of Lines
1	WX198	CIMMYT E95Syn4152-5	X05VSBC01	TAM 111*2/CIMMYT E95Syn4152-5	11
2	WX198	CIMMYT E95Syn4152-5	X05VSBC49	TAM 112*2/CIMMYT E95Syn4152-5	8
3	WX219	CIMMYT E95Syn4152-16	X05VSBC07	TAM 111*2/CIMMYT E95Syn4152-16	8
4	WX219	CIMMYT E95Syn4152-16	X05VSBC51	TAM 112*2/CIMMYT E95Syn4152-16	4
5	WX629	CIMMYT E95Syn4152-37	X05VSBC17	TAM 111*2/CIMMYT E95Syn4152-37	5
6	WX629	CIMMYT E95Syn4152-37	X05VSBC57	TAM 112*2/CIMMYT E95Syn4152-37	13
7	WX408	CIMMYT E95Syn4152-61	X05VSBC31	TAM 111*2/CIMMYT E95Syn4152-61	11
8	WX408	CIMMYT E95Syn4152-61	X05VSBC60	TAM 112*2/CIMMYT E95Syn4152-61	1
9	WX314	CIMMYT E95Syn4152-78	X05VSBC35	TAM 111*2/CIMMYT E95Syn4152-78	5
10	WX314	CIMMYT E95Syn4152-78	X05VSBC65	TAM 112*2/CIMMYT E95Syn4152-78	9
11	WX417	CIMMYT E2Syn4153-31	X05VSBC46	TAM 111*2/CIMMYT E2Syn4153-31	6
12	.	CIMMYT E95Syn4152-51	X05VSBC24	TAM 111*2/CIMMYT E95Syn4152-51	12

4.2.2.2 Greenbug colony

Greenbug biotype E is the major prevailing biotype in the U.S. Great Plains. Initial colonies of biotype E were received from USDA-ARS, Stillwater, OK. These colonies were reared for several generations for a period of 1 month on susceptible check varieties ‘TAM 105’ and ‘TAM 107’ under controlled growth chamber conditions at Texas A&M AgriLife Research facility at Bushland, TX.

4.2.2.3 Experimental setup and phenotyping

A seedbed of 32 linear rows was prepared using potting soil (Miracle Grow). Ten seeds from each genotype were planted in every linear row (Figure 4.1). Each seedbed comprised one row of susceptible check cultivar (TAM 111) and one row of resistant check cultivar (TAM 112) and 30 rows of SDW lines. Seedlings were infested at the two-leaf stage at the rate of 5-6 nymphs of greenbugs per seedling. Following infestation, seedlings were kept in climate-controlled growth chambers with 22 ± 2 °C temperature, 21% relative humidity, and 12 hours photoperiod. Infested seedlings started showing stress symptoms approximately around ten days after infestation. Seedlings were scored around 13 – 16 days from the date of infestation when susceptible check (TAM 111) showed more than 80% yellow leaf area. Each seedling was scored as either R or S based on total percentage of green and yellow leaf area. A standardized scale of 1-9 was developed to determine percent green area and yellow leaf area (refer to figure on page 119). Seedlings with a score of 1-4 were classified R; where, a score of 1 = 0% yellow leaf area, 2 = 1-10% yellow leaf area, 3 = 11-20% yellow leaf area, and 4 = 21-

30% yellow leaf area. Seedlings with a score of 5 and above were classified as S; where, 5 = 31 to 50% yellow leaf area, 6 = 51 to 70% yellow leaf area, 7 = 71 to 90% yellow leaf area, 8 = more than 90% yellow leaf area with some dead tissue, and 9 = 100% dead tissue. Screening was repeated three times to have a more accurate estimate of the resistance. Data analysis was done across all three replications using MS Excel. Lines were classified as R or S depending on percentage of seedlings scored as R or S. A line with more than 75% of the seedlings scored as R were classified as R type. Similarly, a line with 75 to 50% seedlings scored as R were classified as MR type, a line with 50 to 25% seedlings scored as R were classified as MS type, and a line with less than 25% seedlings scored as R were classified as S type.

4.2.2.4 Molecular analysis

Only a set of 93 lines from these 213 lines were selected for preliminary yield trials. Lines that had the best performance across the broad spectrum of traits and environments were selected for these preliminary yield trials. Therefore, molecular studies were restricted to these 93 lines. Ten randomly selected seeds from each of these 93 lines (BC₁F_{5;9} generation) and two check varieties (TAM 111 and TAM 112) were grown on cotton swabs. DNA was extracted from 10-day old seedlings using a modified CTAB protocol standardized at the AgriGenomics laboratory, Texas A&M University, College Station, TX (Doyle and Doyle, 1987). Extracted DNA samples were tested for quality using the gel electrophoresis and quantified using NanoDrop 1000

spectrophotometer. NanoDrop concentration values were used to dilute the DNA samples for required concentration ($10 \text{ ng } \mu\text{L}^{-1}$) for marker analysis.

Texas A&M AgriLife Research has identified two closely linked SNP markers (*Gb₃*-SNP15318 and *Gb₃*-SNP18260) for *Gb₃* gene. For each SNP, an allele-specific forward primers and reverse primer were designed. KASP (KBioscience) genotyping assay was performed for each sample on 384 well plate using these primers. A total volume of 4 μL of KASP 1X master mixture was prepared using 1.912 μL of sterile deionized water, 2 μL KASP 2X reaction mixture, 0.032 μL of 50 mM MgCl_2 , 2 μL of template DNA (dry) at $10 \text{ ng } \mu\text{L}^{-1}$ concentration (10% extra volume was prepared in order to avoid pipetting losses). Robotic pipetting was used to dispense master mix into 384 well. Plates were sealed using flexi-seal heat based plate sealer at $170 \text{ }^\circ\text{C}$ temperature for 4 seconds. PCR was done using ABI 2720 thermal cycler. Touchdown cycling program as mentioned below was used for PCR cycling: 15 min at $94 \text{ }^\circ\text{C}$; 10 touchdown cycles of 20 seconds at $94 \text{ }^\circ\text{C}$, and 60 seconds at $61\text{-}55 \text{ }^\circ\text{C}$ (the annealing temperature for each cycle being reduced by $0.6 \text{ }^\circ\text{C}$ per cycle); and 26-35 cycles of 20 s at $94 \text{ }^\circ\text{C}$ and 60 s at $55 \text{ }^\circ\text{C}$.

The plates were read on a fluorescent plate reader Pherastar Plus. High temperatures of the plate would result in poor or no data read. Therefore, it has been recommended to let the plates cool down to ambient temperature or less than $40 \text{ }^\circ\text{C}$ before reading on fluorescent plate reader. KASP uses the fluorophores FAM and CAL Fluor Orange 560 for distinguishing genotypes. The FAM and VIC data are plotted on

the x and y axes, respectively (Dreisigacker et al., 2013). Allelic combinations in each genotype were determined according to sample clusters (Figure 4.2).



Figure 4.1 Step by step procedure to screen germplasm for greenbug resistance under control growth chamber conditions

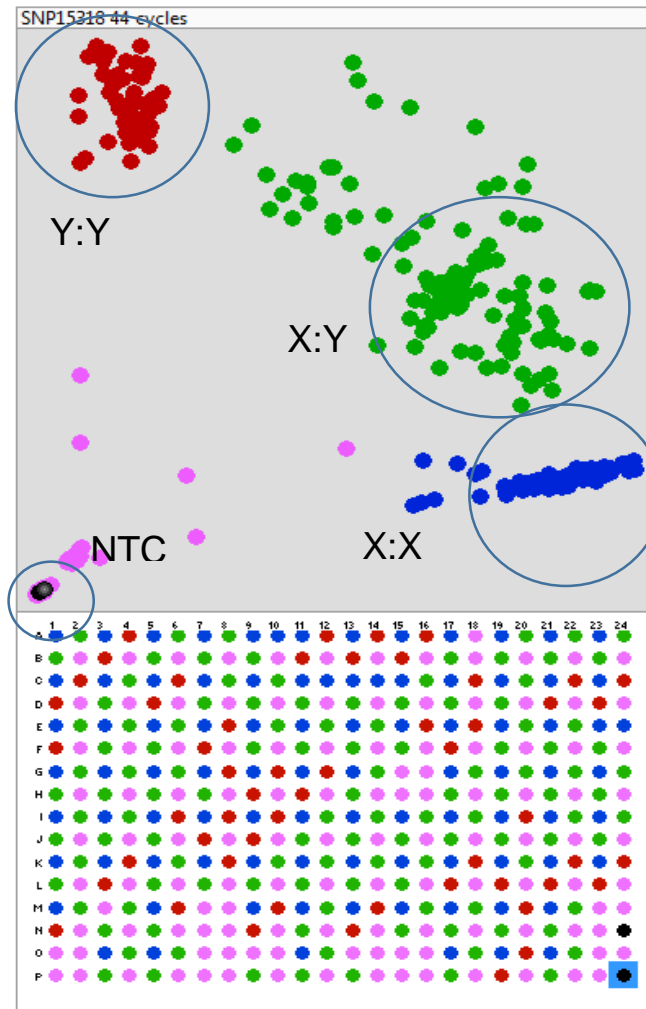


Figure 4.2. KASP graphs of SNPs validation and cluster data presentation for distinguishing 93 genotypes

4.3 Results and discussion

4.3.1 Analysis of variance (ANOVA)

Analysis of variance (ANOVA) for LR and YR indicated there was a significant difference among the genotypes in CS2013 and CAS2014 trials (Table 4.2). In addition, significant differences were observed among the replications in CAS2014. For YR, no significant differences were observed between the replications at 0.05 significance level. However, significant differences could be seen at 0.07 significance level. This advocates that there was a trend to show the variability between the replications for YR distribution.

Table 4.2 Analysis of variance (ANOVA) for leaf rust and stripe rust coefficient of infection (CI)

Leaf rust 2013		
	df	Mean squares
Genotype	99	1199**
Rep	1	1660**
Block (Rep)	18	163
Error	80	87

Leaf rust - 2014		
	df	Mean squares
Genotype	205	699**
Error	26	45.5
Corrected Total		

Stripe Rust - 2014		
	df	Mean squares
Genotype	99	8.51**
Rep	1	0.72
Block(Rep)	18	0.16
Error	81	0.22

*, ** Significant at 0.05 and 0.01, respectively

4.3.2 Response to biotic stresses (by individual line)

The D-genome contributing wild relative of wheat (*Aegilops tauschii* Coss.) has shown broad range of diversity for biotic and abiotic stresses (Assefa and Fehrman, 2000). Synthetic wheats, both SHW and SDW, have inherited part of this diversity because of hybridization with *Aegilops tauschii* Coss. This study has demonstrated that genetic diversity introduced from SHW, has improved the biotic stress tolerance in SDW lines (Table 4.3).

These SDW lines showed broad range of diversity for LR, YR, powdery mildew and greenbug resistance (Table 4.3; Figure 4.3). Differences in infection types and disease severity were observed between CS2013 and CS2014. These differences might have occurred due to changes in the distribution of pathogen races between the two environments. In 2013, race MFPSB that showed virulence to *Lr17* and *Lr24* was most prevalent. In 2014, LR race MBDSB that showed virulence to *Lr39*, *Lr17* was most prevalent. Ironically, the race that was absent (MBDSB) in 2013 was most prevalent in 2014 (USDA-CDL). Similar trends were observed for a number of other races as well. Therefore, values have been converted to CI to facilitate better comparison across infection types and disease severities. These CI values across two environments were transformed to average coefficient of infection (ACI) by weighed mean analysis.

Overall, SDW lines had better YR than LR resistance (Table 4.3; Figure 4.3). The mean ACI value for YR was 6.9, the minimum was 0.2, and the maximum was 50. The mean ACI value for LR was 32, the minimum was 0.2, and the maximum was 71 (Table 4.3). This high YR resistance may be due to the transfer of race-specific major

resistant QTL (*QYr.tam-2BL*) from the TAM 111 parent (Basnet et al., 2014). Around 62% of the SDW lines had TAM 111 as their primary and backcross parent. Therefore, there was a high probability to find this major QTL in the progenies. Around 25% of the lines in CAS2014 were classified as R or MR for LR resistance (Table 4.3). Both the backcross parents, TAM 111 and TAM 112, were susceptible to prevailing races of LR pathogen in the southern Great Plains of Texas. Therefore, resistance present in SDW are believed to have been inherited from the SHW. Reasons for this assumption are that some of the SHW lines (CIMMYT E95Syn4152-78, CIMMYT E95Syn4152-5, CIMMYT E95Syn4152-61 etc.) used in this study were scored MR/MS or MS/MR during preliminary studies (Genetic resources of the WGRC). As 1/3rd of the genome in a backcross is from the donor parent (primary synthetic) there is probability to find some MR/MS genotypes in the progenies (Robbins, 2012).

Resistance to a wide range of insect pests has been reported in SHW (Mujeeb-Kazi 2000b). For greenbug, 30% of lines were classified as R, 10% of lines were classified as MR/MS, and 60% of lines were classified as S (refer to figure in page 130). Most of the SDW with TAM 112 in the pedigree were resistant to greenbug and lines with TAM 111 in the pedigree were susceptible (Table 4.3; Figure 4.3). Hence, *Gb₃* gene is believed to be the major source of resistance in the SDW.

Table 4.3 Response of synthetic derived wheat (SDW) lines for naturally occurring races of leaf and stripe rust, powdery mildew and greenbug biotype E

ID	Genotype	Leaf rust Infection Type-2014	Leaf rust CI-2014	Leaf rust Infection Type-2013	Leaf rust CI-2013	Leaf rust ACI	Stripe rust Infection Type-2014	Stripe Rust CI-2014	Green bug	Powdery Mildew
1	TX11Vsyn0101	MS	31.2	S	30.3	30.9	R	0.3	S	0
2	TX11Vsyn0103	S	40.5	S	54.7	45.2	S	20.0	R	0
3	TX11Vsyn0110	MS	27.5	MS	16.3	23.8	R	0.2	S	5
4	TX11Vsyn0111	tMR	4.2	S	34.5	14.3	R	0.2	S	0
5	TX11Vsyn0112	MS	10.6	MS	19.9	13.7	R	0.2	S	0
6	TX11Vsyn0113	S	29.9	S	16.9	25.6	R	0.2	S	0
7	TX11Vsyn0116	MS	22.8	S	44.7	30.1	R	0.2	S	5
8	TX11Vsyn0118	S	10.4	S	46.0	22.2	R	0.2	S	0
9	TX11Vsyn0119	MS	14.9	S	40.1	23.3	MS	10.0	S	0
10	TX11Vsyn0120	MS	0.6	MS	35.6	12.3	R	0.2	S	0
11	TX11Vsyn0122	R	1.8	MS	31.2	11.6	S	20.0	S	0
12	TX11Vsyn0123	tMR	0.2	MS	38.0	12.8	TS	19.9	MS	0
13	TX11Vsyn0124	R	0.2	S	18.7	6.4	R	0.2	S	0
14	TX11Vsyn0127	R	0.4	S	49.5	16.8	R	0.2	S	6
15	TX11Vsyn0130	S	53.0	S	40.1	48.7	S	20.0	R	2
16	TX11Vsyn0131	S	56.5	S	45.4	52.8	S	20.0	S	0
17	TX11Vsyn0133	S	72.9	S	58.1	68.0	R	0.2	MR	3
18	TX11Vsyn0134	tMR	4.6	S	71.8	27.0	MR	2.2	MS	0
19	TX11Vsyn0135	S	55.5	S	36.5	49.2	R	0.2	MS	0
20	TX11Vsyn0136	MS	20.0	S	17.8	19.3	R	0.2	S	0
21	TX11Vsyn0137	R	0.2	MS	34.6	11.7	MR	0.4	S	5
22	TX11Vsyn0138	S	30.2	S	41.8	34.0	R	0.2	S	0
23	TX11Vsyn0140	S	29.4	S	52.9	37.3	R	0.2	S	2
24	TX11Vsyn0146	MS	14.5	MS	32.4	20.4	R	0.2	S	0
25	TX11Vsyn0153	S	73.0	S	46.9	64.3	S	15.0	S	1

Table 4.3 Continued

ID	Genotype	Leaf rust Infection Type-2014	Leaf rust CI-2014	Leaf rust Infection Type-2013	Leaf rust CI-2013	Leaf rust ACI	Stripe rust Infection Type-2014	Stripe Rust CI-2014	Green bug	Powdery Mildew
26	TX11Vsyn0154	S	58.6	S	28.9	48.7	R	0.2	R	6
27	TX11Vsyn0156	MS	12.9	S	4.9	10.2	R	0.2	S	0
28	TX11Vsyn0158	S	81.1	S	26.1	62.8	R	0.2	S	0
29	TX11Vsyn0159	tMR	8.1	S	27.2	14.5	R	0.2	S	4
30	TX11Vsyn0160	R/R	2.2	S	40.9	15.1	R	0.2	S	0
31	TX11Vsyn0161	S	18.5	S	33.2	23.4	R	0.2	S	0
32	TX11Vsyn0164	R	4.1	S	36.0	14.7	S	15.0	S	2
33	TX11Vsyn0165	R	0.3	R	13.6	4.7	R	0.2	S	2
34	TX11Vsyn0167	TR	0.2	MS	28.3	9.6	R	0.2	S	5
35	TX11Vsyn0168	S	54.1	S	44.4	50.9	R	0.2	MS	2
36	TX11Vsyn0169	tMR	1.0	S	22.1	8.0	R	0.2	S	2
37	TX11Vsyn0174	S	74.6	S	65.5	71.5	R	0.2	S	5
38	TX11Vsyn0175	S	68.3	MS/MR	17.6	51.4	S	15.0	S	0
39	TX11Vsyn0178	S	51.4	S	44.4	49.0	R	0.2	S	1
40	TX11Vsyn0179	S	25.1	S	15.2	21.8	S	50.0	S	1
41	TX11Vsyn0180	S	28.9	S	55.2	37.7	R	0.2	S	0
42	TX11Vsyn0182	S	48.0	MS	5.1	33.7	TMS	4.4	S	3
43	TX11Vsyn0185	S	40.5	S	37.1	39.4	R	0.2	S	0
44	TX11Vsyn0188	R	0.2	S	48.8	16.4	R	0.2	S	0
45	TX11Vsyn0189	S	66.8	S	36.1	56.6	S	15.0	S	0
46	TX11Vsyn0190	S	71.1	S	35.8	59.4	S	30.0	S	0
47	TX11Vsyn0191	MS	5.0	S	51.6	20.5	R	0.2	S	0
48	TX11Vsyn0195	S	22.8	S	37.4	27.6	R	0.2	R	2
49	TX11Vsyn0196	MS	21.8	S	32.8	25.4	S	12.5	S	0
50	TX11Vsyn0197	R	0.2	MS	32.7	11.0	R	0.2	R	0

Table 4.3 Continued

ID	Genotype	Leaf rust Infection Type-2014	Leaf rust CI-2014	Leaf rust Infection Type-2013	Leaf rust CI-2013	Leaf rust ACI	Stripe rust Infection Type-2014	Stripe Rust CI-2014	Green bug	Powdery Mildew
51	TX11Vsyn0199	R	4.0	MS	23.1	10.4	R	0.2	S	0
52	TX11Vsyn0201	R	0.6	S/MS	36.7	12.6	R	0.2	S	3
53	TX11Vsyn0208	S	55.0	S	42.7	50.9	S	40.0	S	0
54	TX11Vsyn0211	MR	0.4	S	42.7	14.5	MR	0.4	R	3
55	TX11Vsyn0212	S	41.1	S	22.5	34.9	R	0.2	R	0
56	TX11Vsyn0213	S	58.8	S	38.4	52.0	R	0.2	MS	5
57	TX11Vsyn0216	.	14.2	S	10.0	12.8	S	25.0	S	0
58	TX11Vsyn0217	S	13.4	S	45.2	24.0	R	0.2	S	1
59	TX11Vsyn0219	S	23.8	tS	29.5	25.7	R	0.2	S	0
60	TX11Vsyn0225	tMR	6.1	S	41.8	18.0	R	0.2	R	0
61	TX11Vsyn0226	S	35.2	S	35.8	35.4	MR	0.4	R	0
62	TX11Vsyn0228	S	65.1	S	52.5	60.9	MR	0.3	S	0
63	TX11Vsyn0229	R	5.2	S	33.2	14.6	R	0.3	MR	0
64	TX11Vsyn0230	MS	9.8	S	5.8	8.5	R	0.2	R	1
65	TX11Vsyn0232	S	42.8	S	37.5	41.0	S	25.0	R	2
66	TX11Vsyn0234	S	49.1	S	36.1	44.8	R	0.2	R	0
67	TX11Vsyn0238	S	61.0	S	33.2	51.8	R	0.2	R	0
68	TX11Vsyn0240	S	62.3	S	48.3	57.6	R	0.2	MR	2
69	TX11Vsyn0241	S	72.4	S	50.5	65.1	MR	0.4	R	0
70	TX11Vsyn0243	S	56.2	S	23.8	45.4	R	0.2	R	3
71	TX11Vsyn0253	S	17.3	S	50.4	28.3	S	15.0	R	0
72	TX11Vsyn0261	S	74.1	S	54.3	67.5	S	15.0	R	0
73	TX11Vsyn0263	S	46.6	S	23.8	39.0	TMS	4.4	R	0
74	TX11Vsyn0264	S	79.1	S	52.9	70.4	MR	0.4	R	0
75	TX11Vsyn0265	S	61.8	S	52.9	58.8	S	45.0	R	3
76	TX11Vsyn0266	S	64.0	S	38.7	55.5	S	35.0	R	0

Table 4.3 Continued

ID	Genotype	Leaf rust Infection Type-2014	Leaf rust CI-2014	Leaf rust Infection Type-2013	Leaf rust CI-2013	Leaf rust ACI	Stripe rust Infection Type-2014	Stripe Rust CI-2014	Green bug	Powdery Mildew
77	TX11Vsyn0267	S	55.5	.	.	55.5	MS	23.0	R	0
78	TX11Vsyn0271	S	45.6	S	80.6	57.2	MS	8.0	R	4
79	TX11Vsyn0272	S	28.4	S	43.0	33.3	S	35.0	R	3
80	TX11Vsyn0275	S	18.3	S	45.7	27.5	R	0.2	MR	6
81	TX11Vsyn0277	tMR	0.4	S	51.9	17.6	R	0.2	R	0
82	TX11Vsyn0279	S	29.6	S	22.1	27.1	S	35.0	R	0
83	TX11Vsyn0280	S	60.1	S	25.5	48.6	MS	12.0	R	0
84	TX11Vsyn0282	S	16.0	S	39.6	23.8	R	0.3	R	0
85	TX11Vsyn0294	S	20.7	S	44.7	28.7	R	0.2	S	0
86	TX11Vsyn0300	S	57.8	S	28.1	47.9	TMS	2.4	S	3
87	TX11Vsyn0303	MR	0.9	MS	8.5	3.4	R	0.2	S	4
88	TX11Vsyn0305	S	20.4	S	53.8	31.5	S	40.0	S	7
89	TX11Vsyn0306	MS	11.9	MS	25.0	16.3	R	0.2	S	5
90	TX11Vsyn0308	MS	15.0	S	37.5	22.5	R	0.2	S	0
91	TX11Vsyn0309	S	27.8	S	22.2	25.9	R	0.2	S	2
92	TX11Vsyn0312	S	60.2	S	51.4	57.2	R	0.2	S	0
93	TX11Vsyn0313	MS	11.0	S	33.2	18.4	R	0.2	S	0
95	TAM111	S	41.4	S	58.5	47.1	tMR/MS	0.6	S	3
94	TAM112	S	71.0	S	51.8	64.6	S/MS	19.0	R	0
96	TAM113	R	0.2	.	.	0.2	MR	0.3	.	.
97	TAM304	tR	3.2	.	.	3.2	MS	4.2	.	.
98	TAM305	tR	0.2	.	.	0.2	R	0.2	.	.
99	TAM401	R	5.7	S/MS	18.7	10.0	R	2.1	.	0
100	TAMW101	R	0.4	.	.	0.4	S	20.0	.	.
	Mean		29.9		36.2	31.6		6.9		
	Minimum		0.2		0.2	0.2		0.2		
	Maximum		81.1		80.6	71.5		50.0		

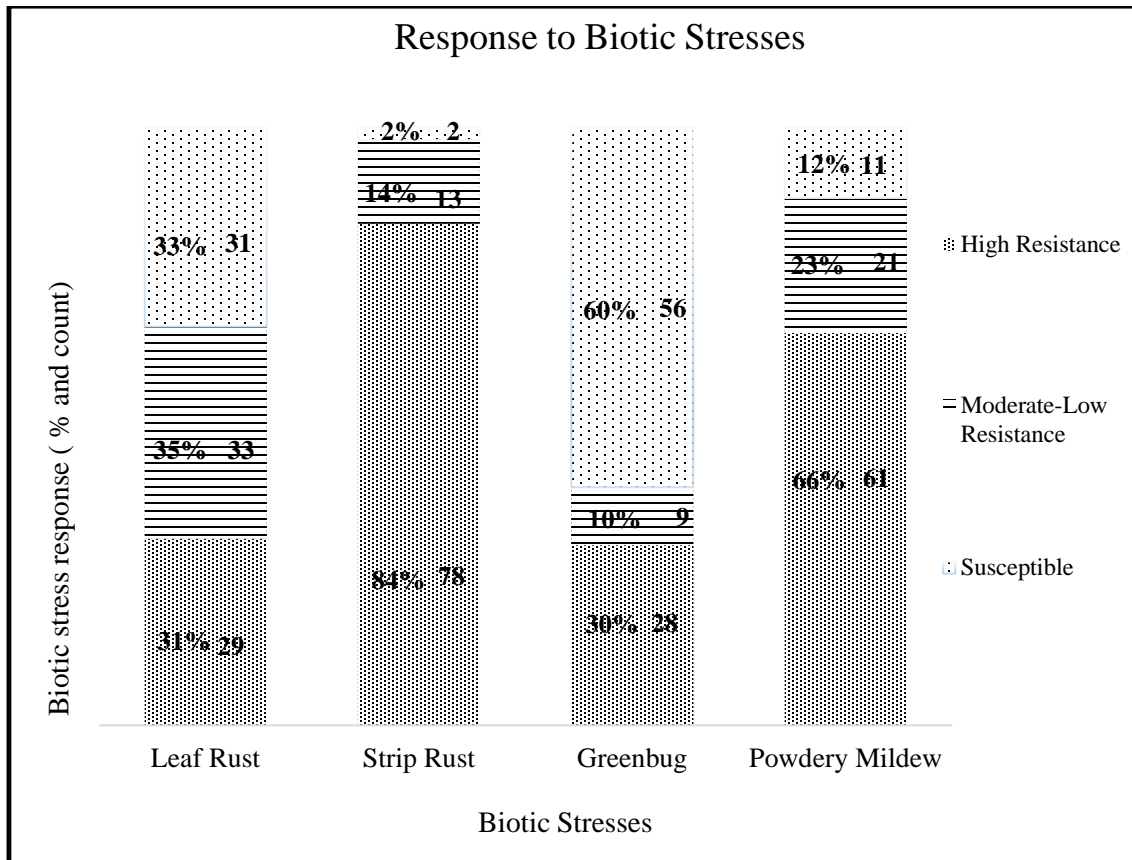


Figure 4.3 Response of synthetic derived wheat lines to common biotic stresses in the USA Great Plain.

Table 4.4 Pearson correlation of rust diseases with yield (GY), test weight (TW), single seed weight (SeedWt), seeds per head (SeedsHead⁻¹), heads per square meter (HeadNo), plant height (HT), heading date (HS)

Trait	Leaf Rust CI-2014	Leaf Rust CI-2013	Combined leaf rust CI	Stripe Rust CI-2014
GY	-0.24**	0.11 ^{ns}	-0.12	-0.17**
TW	-0.24**	0.031*	-0.15	-0.14*
SeedWt	-0.22**	-0.35**	-0.26	-0.27**
SeedsHead ⁻¹	-0.06 ^{ns}	0.07 ^{ns}	-0.02	-0.21**
HeadNo	-0.07 ^{ns}	0.21**	0.02	0.07 ^{ns}
HT	-0.24**	-0.23 ^{ns}	-0.24	-0.15*
HS	-0.27**	-0.27 ^{ns}	-0.27	-0.20**

Disease pressure for powdery mildew was not very high. A maximum score of 7 was recorded during the cropping season. For this study, genotypes with powdery mildew score of 0-1 were considered as highly resistant, 2-4 as moderately resistant and 5-7 as susceptible. TAM 112 was reported to have a high level of resistance to a score of 0. The other backcross parent TAM 111 was reported to have powdery mildew score of 3. Around 66% of genotypes were highly resistant, 23% moderately resistant, and 12% were susceptible (Figure 4.3).

4.3.3 Scope for durable rust resistance

Most of the wheat breeding programs in the U.S. Great Plains rely heavily on race-specific major genes for LR and YR resistance (Bockus et al., 2009). However, often times resistance based on major genes has a short life span because of the evolution of new virulent pathogen races to the resistant gene (Bux et al., 2012). This had compelled scientists to explore new ways to build durable rust resistance in wheat. Caldwell (1968) had proposed the concept of slow-rusting resistance in wheat, which was further supported by Dr. Sanjaya Rajaram of CIMMYT for developing durable rust-resistant wheat (Singh et al., 2010). It took over 30 years to realize the concept of durable rust resistance using slow-rusting APR genes (Singh et al., 2010). CIMMYT has produced a large number of lines that are near immune to rust with APR genes (Singh et al. 2010). To date, *Lr34/Yr18*, *Lr46/Yr29*, and *Lr67/Yr46* are the most studied and most effective APR genes (Singh et al., 2010). Lack of efficient genotyping tools to identify slow-rusting APR genes has raised the need for thorough field-based studies. Many

studies have shown CI is one of the most efficient ways to identify the level of APR (Pathan and Park, 2006; Ali et al., 2009; Gashaw and Bazie, 2014). Along with CI traits such as final rust severity (FRS) and area under disease progress curve (AUDPC) can act as supplemental information in determining slow-rusting genotypes. A number of studies have reported high correlation ($R^2 > 90$) between CI and FRS and also CI and AUDPC (Nzuve et al., 2012; Safavi and Afshari, 2012). For this study, the level of APR was determined on the basis of the average coefficient of infection (ACI) by combining two environments CI values (Pathan and Park, 2006).

Few studies have reported ACI values of 0-20, 20-40, and 40-60 can be classified as high, moderate, and low levels of resistance, respectively. The ACI values over 60 can be considered as susceptible (Pathan and Park, 2006; Ali et al., 2009; Gashaw and Bazie, 2014). However, experimental conditions for these studies were different from those conducted by previous authors. The high ACI value for this study was 20-30 points lower than what was reported by Ali et al. 2009. Therefore, we propose considering ACI values over 40 as susceptible, values 0-19 as high resistance, 20-39 as moderate-low resistance. Infection types and CI values for this study gave us a hint that there are possibly some slow-rusting APR genes in the SDW lines (Table 4.3). Data would have been more reliable if multiple CI values were recorded from each location. However, one time score does not rule out the utility of CI in determining slow rusting APR (Pathan and Park 2006; Ali et al., 2009). A summary of ACI values across all SDW lines showed around 31% of lines for LR and 84% of lines for YR are considered to have a high level of resistance. Similarly, 35% and 14% of lines were considered to have

moderate-low level of resistance, and 33% and 2% were susceptible for LR and YR, respectively (Figure 4.3). These values suggest that there was a partial level of resistance in the SDW lines. Many studies have credited partial resistance to APR genes (Ellis et al., 2014). Therefore, there seems to be high chances for having one or few adult plant resistance (APR) genes in the SDW lines. A detailed classification of resistance within each population and among populations are presented in the figure on page 137 and 138.

4.3.4 Pearson's correlation coefficients

Pearson correlation coefficients were obtained to determine the relationship between rust resistance, yield, yield components and morphological traits in the SDW lines (Table 4.4). Among the yield components, SeedWt was most affected by diseases. Both LR and YR had a highly significant negative correlation with SeedWt. YR had significant negative effect on SeedsHead⁻¹. These results are justifiable as YR in CAS2014 occurs during jointing to booting stage. Herberk and Lee have reported jointing is a critical stage for determining seed number in wheat. Therefore, any stress (including YR) during this stage reduces seeds per head and later stresses reduce SeedWt. Similar results were reported by Murray et al. (1995). Pearson correlations showed there was highly significant negative correlation between each of LR (-0.24;) and YR (-0.17) on one hand and GY (refer to the table on page 130). Many other studies such as Basnet (2012) have shown YR causes higher yield loss than LR at similar infection levels.

4.3.5 Response to biotic stresses (by population and primary synthetic)

A detailed comparison for biotic stress response within and among 12 populations is presented in table 4.5 and in figures on page 137 to 138. Figure 4.4 presents LR resistance among 12 SDW populations. Populations X05VBC35 (ID = 9), X05VBC60 (ID = 8) and X05VBC01 (ID = 1) had high percentage of partial resistance (ACI values 1-39%) and populations X05VBC07 (ID = 3) and X05VBC51 (ID = 4) had lowest level of usable and partial resistance. A detailed study of pedigrees of population 9, 8 and 1 revealed primary synthetic with MR/MS response to LR were part of their pedigrees. However, population 3 and 4 had primary synthetic with 40S response to LR in their pedigrees (Genetic resources of the WGRC).

Similarly, for YR, populations X05VBC51 (ID = 4), X05VBC17 (ID = 5), X05VBC24 (ID = 12), and X05VBC35 (ID = 9) had highest level of partial resistance and populations X05VBC46 (ID = 11), X05VBC57 (ID = 6), and X05VBC31 (ID = 7) had lowest level of partial resistance (Figure 4.5). Most genotypes were recorded as having R or MR infection type with few immune (I) types. The key finding here is that populations (ID = 2, 4, 6, 8, and 10), which have susceptible TAM 112 in their pedigree, were classified as having high level of resistance and most of these reactions were either R or MR or close to I type reaction (Figure 4.5). These results confirm that synthetics might have contributed one or few major genes or contributed multiple slow rusting genes as blocks.

The response of SDW to greenbug biotype E infestation is presented in figure 4.6. Results showed populations X05VBC49 (ID = 2), X05VBC51 (ID = 4), X05VBC57

(ID = 6), and X05VBC60 (ID = 8) had high percentage of resistant lines. All these resistant populations had TAM 112 in their pedigrees. These populations were developed by hybridizing and backcrossing SHW to TAM 112. Therefore, probabilities of finding *Gb3* gene as the resistant resource in these SDW lines was also high. However, population 10 had contradictory results. Although it had TAM 112 in the pedigree all the lines in this population were classified as susceptible. Chances of inheriting a single dominant gene, such as *Gb3*, in a backcross is supposed to be high but our results showed there was no inheritance (Figure 4.6). Therefore, we speculate that TAM 112 might not be the actual parent of this pedigree. There might have been some problem either during hybridization or backcrossing or labelling the genotype.

In case of powdery mildew, populations that had TAM 111 in their pedigrees seem to have around 20% lines with low level of resistance (Figure 4.7). However, lines with TAM 112 in their pedigrees had greater level of resistance. These results are justifiable as TAM 111 is classified as moderately susceptible and TAM 112 as resistant to powdery mildew. Overall, there seems to be a good level of resistance in the SDW lines for this disease agent.

Lines used in this study were selected from 12 populations with different pedigrees. Every population had at least one of seven SHW as a parent in the pedigree. Therefore, a comparative study among populations will be useful for breeding programs. In addition, it gives an indirect estimate of best SHW for the trait of interest. Possibilities for calculating combining ability for every population was limited as not every SHW was

hybridized with both TAM 111 and TAM 112. Therefore, weighed mean analysis was done to determine the value of each SHW.

For LR ACI values were used to determine the worth of SHW line. The weighed mean analysis showed SHW line CIMMYTE95Syn4152-78 (*Aegilops tauschii* = WX314) had the highest percentage of lines (86%) with ACI values 0-39. In contrast, primary synthetic CIMMYTE95Syn4152-16 (*Aegilops tauschii* = WX198) had the lowest percentage of lines (50%) with ACI values 0-39. This indicates CIMMYTE95Syn4152-78 might be one of the best parents that can be studied more for APR genes. Similar studies for YR showed 100% of lines in SHW CIMMYTE95Syn4152-5 (*Aegilops tauschii* = WX198) and CIMMYTE95Syn4152-16 (*Aegilops tauschii* = WX219) had ACI value 0-39. SHW CIMMYTE95Syn4152-31 (*Aegilops tauschii* = WX417) had lowest percentage (83%) of lines with ACI values 0-39. However, most of the YR resistance was either I or R types. Therefore, there is less scope for utilizing them in building durable rust resistance. Overall, SHW CIMMYTE95Syn4152-78 (*Aegilops tauschii* = WX314) seems to be an ideal parent of choice for building resistance.

Table 4.5 Leaf rust and stripe rust coefficient of infection (CI) by population

Pop ID	Pedigree		Leaf rust CI CAS2014	Leaf rust CI CS2013	Leaf rust ACI	Yellow rust CI CAS2014
1	TAM 111*2/CIMMYT E95Syn4152-5	Mean	17.7	33.7	23	4.7
		Range	(0.6-40.5)	(16.3- 54.7)	(11.6-45.2)	(0.2-20.0)
2	TAM 112*2/CIMMYT E95Syn4152-5	Mean	29.6	34.0	31.1	3.4
		Range	(5.2-65.1)	(5.8-52.5)	(8.5-60.9)	(0.2-25)
3	TAM 111*2/CIMMYT E95Syn4152-16	Mean	30.4	44.8	35.2	7.9
		Range	(0.2-72.9)	(18.7-71.8)	(6.4-68)	(0.2-20)
4	TAM 112*2/CIMMYT E95Syn4152-16	Mean	63	39.0	55	0.3
		Range	(56.2-72.4)	(23.8-50.5)	(45.4-65.1)	(0.2-0.4)
5	TAM 111*2/CIMMYT E95Syn4152-37	Mean	18.8	35.9	24.5	0.2
		Range	(0.2-30.2)	(17.8-52.9)	(11.7-37.3)	(0.2-0.4)
6	TAM 112*2/CIMMYT E95Syn4152-37	Mean	44.7	41.7	43.7	17.6
		Range	(0.4-79.1)	(0.0-80.6)	(17.6-70.4)	(0.2-45.0)
7	TAM 111*2/CIMMYT E95Syn4152-61	Mean	43.6	37.5	41.6	10.5
		Range	(0.2-74.6)	(5.1-65.5)	(16.4-71.5)	(0.2-50.0)
8	TAM 112*2/CIMMYT E95Syn4152-61	Mean	16	39.6	23	0.30
9	TAM 111*2/CIMMYT E95Syn4152-78	Mean	9.9	32.5	17.4	2.7
		Range	(0.2-22.8)	(23.1-37.4)	(10.4-27.6)	(0.2-12.5)
10	TAM 112*2/CIMMYT E95Syn4152-78	Mean	25	33.8	28	4.9
		Range	(0.9-60.2)	(8.5-53.8)	(3.4-57.2)	(0.2-40.0)
11	TAM 111*2/CIMMYT E2Syn4153-31	Mean	30.5	33.6	31.5	11.0
		Range	(0.4-58.8)	(10.0-45.2)	(12.8-52)	(0.2-40.0)
12	TAM 111*2/CIMMYT E95Syn4152-51	Mean	26.2	29.4	27.3	2.7
		Range	(0.2-81.1)	(4.9-46.9)	(4.7-64.3)	(0.2-15.0)
Check	TAM111		41.4	58.5	47.1	0.60
Check	TAM112		71.	51.8	64.6	19.00
Check	TAM113		0.2	.	0.2	0.30
Check	TAM304		3.2	.	3.2	4.20
Check	TAM305		0.2	.	0.2	0.20
Check	TAM401		5.7	18.7	10	2.10
Check	TAMW101		0.4	.	0.4	20.00

Table 4.6 Number of synthetic derived wheat (SDW) lines with high level of resistance (High R), moderate to low level of resistance (M-L R), and susceptible (S) within each population

Pop. ID	Pedigree	Total	Leaf Rust			Stripe Rust			Powdery Mildew		
			High R	M-L R	S	High R	M-L R	S	High R	M-L R	S
1	TAM 111*2/CIMMYT E95Syn4152-5	11	4	4	2	9	2	0	9	0	2
2	TAM 112*2/CIMMYT E95Syn4152-5	8	2	4	2	7	1	0	7	1	0
3	TAM 111*2/CIMMYT E95Syn4152-16	8	1	3	4	6	2	0	4	2	1
4	TAM 112*2/CIMMYT E95Syn4152-16	4	0	2	2	4	0	0	2	2	0
5	TAM 111*2/CIMMYT E95Syn4152-37	5	2	2	1	5	0	0	3	1	1
6	TAM 112*2/CIMMYT E95Syn4152-37	13	0	7	6	8	4	1	9	3	1
7	TAM 111*2/CIMMYT E95Syn4152-61	11	1	5	5	9	1	1	9	1	1
8	TAM 112*2/CIMMYT E95Syn4152-61	1	0	1	0	1	0	0	1	0	0
9	TAM 111*2/CIMMYT E95Syn4152-78	5	1	4	0	5	0	0	3	2	0
10	TAM 112*2/CIMMYT E95Syn4152-78	9	0	7	2	8	0	1	4	3	2
11	TAM 111*2/CIMMYT E2Syn4153-31	6	0	5	1	4	1	1	4	1	1
12	TAM 111*2/CIMMYT E2Syn4153-51	12	3	6	3	12	0	0	5	5	2

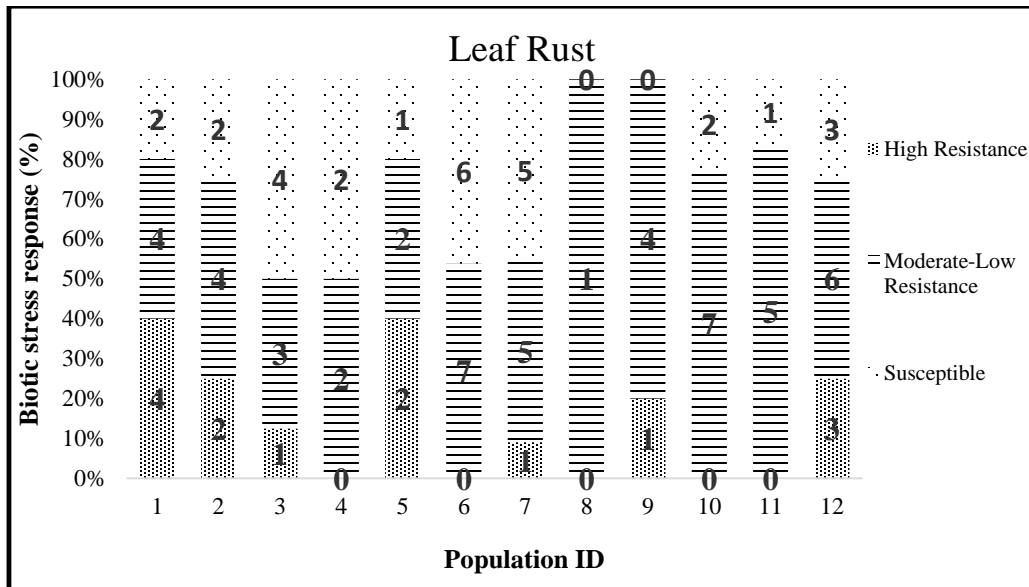


Figure 4.4 Proportion of leaf rust resistance within and among synthetic derived (SDW) wheat populations Pop. ID of synthetic derived wheat 1 = X05VSBC01, 2 = X05VSBC42, 3 = X05VSBC67, 4 = X05VSBC51, 5 = X05VSBC17, 6 = X05VSBC57, 7 = X05VSBC37, 8 = X05VSBC60, 9 = X05VSBC35, 10 = X05VSBC65, 11 = X05VSBC46, 12 = X05VSBC24

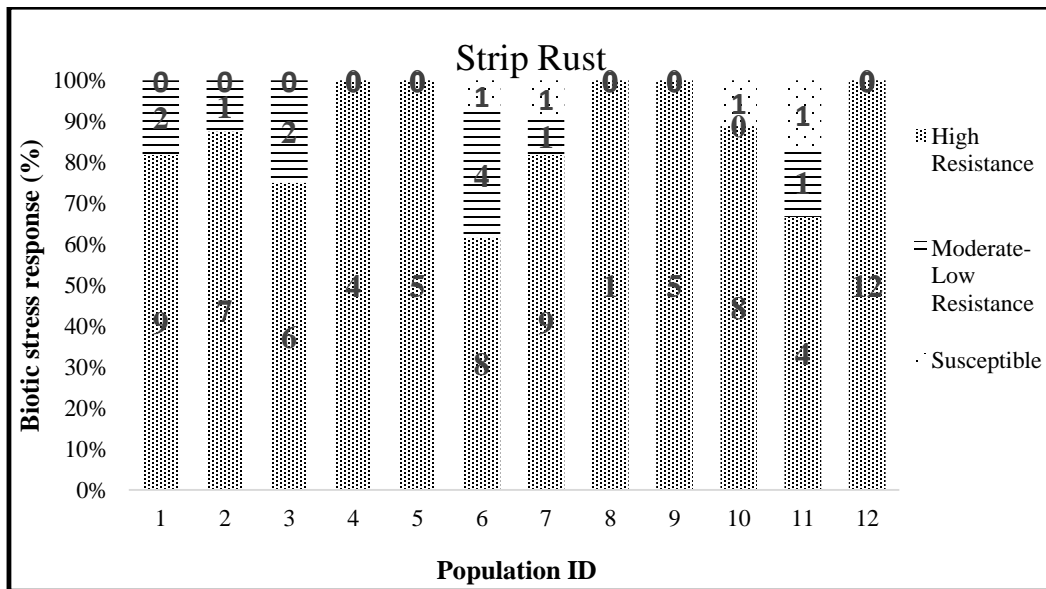


Figure 4.5 Proportion of stripe rust resistance within and among synthetic derived (SDW) wheat populations. Pop. ID of synthetic derived wheat 1 = X05VSBC01, 2 = X05VSBC42, 3 = X05VSBC67, 4 = X05VSBC51, 5 = X05VSBC17, 6 = X05VSBC57, 7 = X05VSBC37, 8 = X05VSBC60, 9 = X05VSBC35, 10 = X05VSBC65, 11 = X05VSBC46, 12 = X05VSBC24

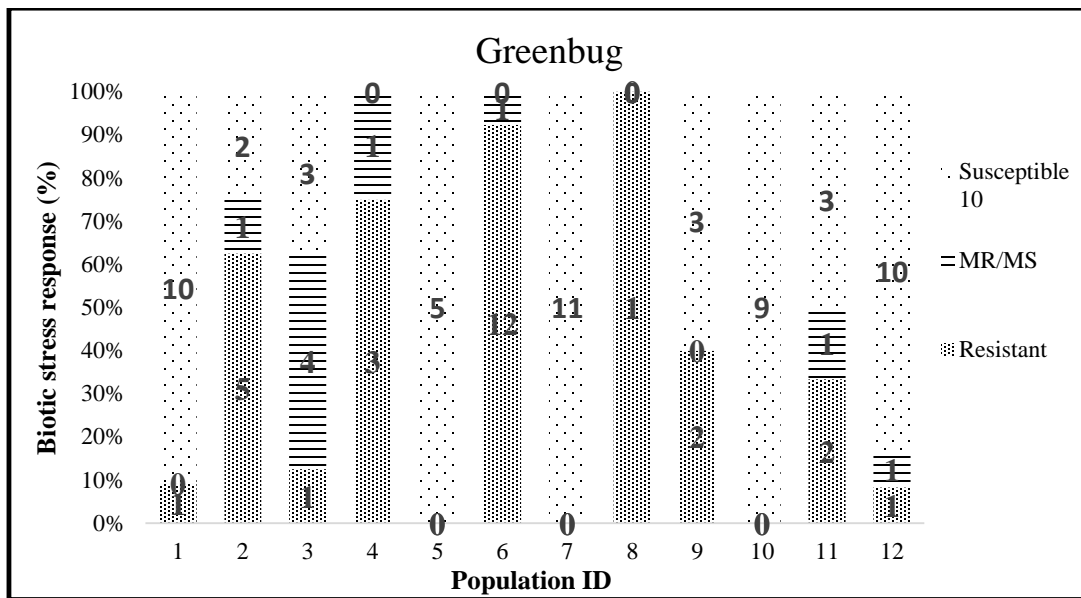


Figure 4.6 Proportion of greenbug resistance within and among synthetic derived (SDW) wheat populations. Pop. ID of synthetic derived wheat 1 = X05VSBC01, 2 = X05VSBC42, 3 = X05VSBC67, 4 = X05VSBC51, 5 = X05VSBC17, 6 = X05VSBC57, 7 = X05VSBC37, 8 = X05VSBC60, 9 = X05VSBC35, 10 = X05VSBC65, 11 = X05VSBC46, 12 = X05VSBC24

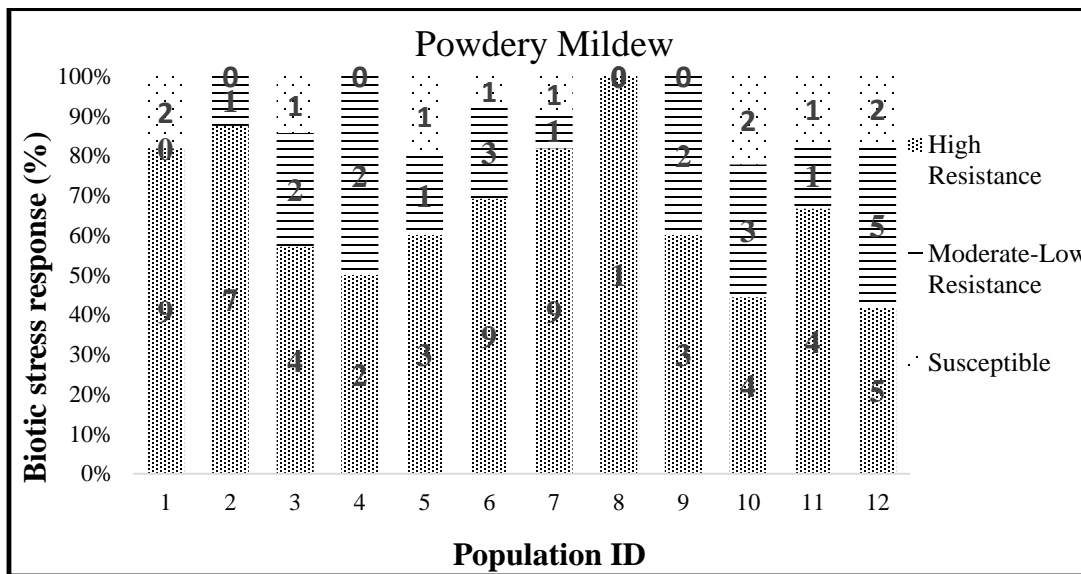


Figure 4.7 Proportion of powdery mildew resistance within and among synthetic derived (SDW) wheat populations. Pop. ID of synthetic derived wheat 1 = X05VSBC01, 2 = X05VSBC42, 3 = X05VSBC67, 4 = X05VSBC51, 5 = X05VSBC17, 6 = X05VSBC57, 7 = X05VSBC37, 8 = X05VSBC60, 9 = X05VSBC35, 10 = X05VSBC65, 11 = X05VSBC46, 12 = X05VSBC24

4.3.6 Validation of markers for greenbug resistance

Phenotypic classification of lines as R or S was done based on a mean value obtained across three replications. There was great level of variability among SDW lines for greenbug resistance (Table 4.6). Around 30% (28 lines) of lines were classified as R, 60% lines (56 lines) as S, and 10% (9 lines) lines as MR/MS. Most of the resistant lines had greenbug-resistant cultivar TAM 112 as a parent in their pedigree. TAM 112 carries single dominant greenbug resistant gene *Gb₃*. Therefore, chances of finding *Gb₃* gene in this set of SDW lines was very high. However, we also found few resistant genotypes that had susceptible TAM 111 as a parent in their pedigrees. This indicates that SHW might also be contributing some source of greenbug resistance. Therefore, molecular studies were done to validate the source of resistance present in SDW.

Texas A&M AgriLife research has developed flanking SNP markers that are closely linked to *Gb₃* gene (*Gb₃*-SNP15318 and *Gb₃*-SNP18260) on chromosome 7DL. We validated these SNP markers on SDW lines using KASP assay genotyping technology. Plates were read using a fluorescent plate reader Pherastar Plus and data was plotted on X and Y-axes. Based on the type of alleles present, we partitioned data into different clusters

These results showed a great level of harmony between SNP markers and phenotypic scores (Table 4.7). For SNP15318 marker, there was 96% harmony between the genotyping and phenotyping score for susceptible lines and 80% harmony between the genotyping and phenotyping score for resistant genotypes. Similarly, for SNP18260 marker, there was 98% harmony for susceptible genotypes and 90% harmony for

resistant genotypes. There were very few false positives and false negatives in this study. There were some missing values in the data. One possible reason for missing values might be attributed to the limited knowledge of temperature requirements for the KASP protocol. Plates were read right after removing from PCR cyclers. Temperature of the plates during that time might have been higher than 40 °C. Genotyping and phenotyping scores show that greenbug resistant gene(s) is/are very closely linked to *Gb3* loci.

Future course of work includes doing genetic analyses to identify candidate genes and to do functional analysis for these two SNP markers. The functional analysis will eventually help to determine the source of resistance in these SDW lines.

Table 4.7. Phenotypic score and marker validation for greenbug resistance

Pop. ID	Pedigree	Phenotype (No. of lines)					<i>Gb</i> ₃ -SNP15318 (No. of lines)			<i>Gb</i> ₃ -SNP18260 (No. of lines)		
		R	MR	MS	S	Total	X:X	X:Y	Y:Y	X:X	X:Y	Y:Y
1	TAM 111*2/CIMMYT E95Syn4152-5	1	0	0	10	11	4	5	1	6	1	1
2	TAM 112*2/CIMMYT E95Syn4152-5	5	1	0	2	8	0	4	4	0	0	6
3	TAM 111*2/CIMMYT E95Syn4152-16	1	1	3	3	8	5	1	2	4	1	2
4	TAM 112*2/CIMMYT E95Syn4152-16	3	1	0	0	4	0	1	3	0	0	4
5	TAM 111*2/CIMMYT E95Syn4152-37	0	0	0	5	5	5	0	0	5	0	0
6	TAM 112*2/CIMMYT E95Syn4152-37	12	1	0	0	13	1	2	9	1	0	11
7	TAM 111*2/CIMMYT E95Syn4152-61	0	0	0	11	11	10	0	1	8	2	1
8	TAM 112*2/CIMMYT E95Syn4152-61	1	0	0	0	1	0	0	1	0	0	0
9	TAM 111*2/CIMMYT E95Syn4152-78	2	0	0	3	5	3	2	0	3	0	2
10	TAM 112*2/CIMMYT E95Syn4152-78	0	0	0	9	9	9	0	0	8	1	0
11	TAM 111*2/CIMMYT E2Syn4153-31	2	0	1	3	6	4	0	2	4	0	2
12	TAM 111*2/CIMMYT E95Syn4152-51	1	0	1	10	12	10	0	2	10	1	1
	TAM 112	1	0	0	0		0	0	1	0	0	1
	TAM 111	0	0	0	1		1	0	0	1	0	0
	Sum	28	4	5	56	93	51	15	25	49	6	30
	Consistent of G to P						49		20	48		27
	False positive						0		5	0		3

5. SUMMARY

Wheat (*Triticum aestivum* L.) is one of the major staple food crops that contributes 19% calories and 21% protein of the global population diet. With the increase in global population, the demand for wheat could reach 900 million tons by 2050. However, narrow genetic base and continued pressure from abiotic and biotic stresses pose a tough challenge to achieve the expected increase in wheat grain yield. Research leading to the evolution of synthetic hexaploid wheat (*Triticum durum* x *Aegilops tauschii*) and synthetic derived wheat (SDW) (elite bread wheat X synthetic hexaploid wheat) provided a tremendous opportunity to improve wheat production.

Multi-location yield trials indicated certain SDW produced higher grain yield than their recurrent parents and common check varieties. The proportion of yield advantage varied from 12% to 39%, depending on the type of the environment. Combined analysis across locations demonstrated that mean of top ten yielding SDW lines was 12% higher than mean of recurrent parents, TAM 111 and TAM 112. A thorough investigation of factors contributing to higher yield showed that SDW had similar seeds per head, head m^{-2} as recurrent parents, however, had 10% higher single seed weight than recurrent parents. Overall, these SDW lines proved advantageous under biotic and abiotic stress environments, high and low yielding, rainfed, and irrigated environments. The highest percentage of yield advantage was observed in low yielding abiotic stress environments.

In addition, quantitative genetic parameters such as variance components, genotypic coefficient of variation, heritability, and genetic gain were also estimated for

various traits. These estimates indicated that there is tremendous scope for grain yield improvement by utilizing SDW. Overall, genotypic coefficient of variation for grain yield and yield components indicated that there is high level of genetic variation among SDW lines for single seed weight (22%), seeds per head (11%), and grain yield (11%). Genetic gain results indicated that grain yield can be improved by 15.6% per cycle at 10% selection intensity ($i = 1.76$). Indirect selection for yield using yield components indicated that the efficiency of indirect selection for yield is never as efficient as direct selection. Efficiency of indirect selection using heads per meter square, seeds per head, and single seed weight is 0.41, 0.46, and 0.21, respectively.

In addition, SDW showed better resistance to leaf and stripe rust, greenbug, and powdery mildew resistance compared to their recurrent parents. Overall, 31%, 84%, 30%, and 66% of lines in this population showed high level of resistance or low incidence of leaf rust, stripe rust, greenbug, and powdery mildew, respectively. There were certain indications to show that some resistance was transmitted from primary synthetics.

These results indicate that SDW contributed some favorable alleles for yield, biotic stress resistance, and abiotic stress tolerance. These results also showed that SDW contributions were advantageous under both rainfed and irrigated conditions, which makes them an invaluable source for increasing genetic diversity and improving performance of Texas A&M AgriLife wheat germplasm.

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APPENDIXES

Appendix 1

Mean, range, LSD (least significant difference), CV (coefficient of variation) for grain yield, test weight, seed wt. (single seed weight), seeds head⁻¹, head number, height, heading score at Chillicothe, TX in 2014

ID	Genotype	Grain Yield t ha ⁻¹	Test Weight kg hL ⁻¹	Seed Wt g	Seeds Head-1 count	Head No Heads m ⁻²	HT cm	HS scale 1-5
1	TX11Vsyn0101	1.23	71.2	0.0278	24	178	64.8	2
2	TX11Vsyn0103	1.14	68.7	0.0222	27	165	53.7	2
3	TX11Vsyn0110	0.7	65.7	0.0235	21	151	60	2
4	TX11Vsyn0111	1.03	73.1	0.0309	20	184	57.4	2
5	TX11Vsyn0112	1.06	70.5	0.0263	25	174	55.4	2
6	TX11Vsyn0113	1	73.4	0.03	20	152	59.8	2
7	TX11Vsyn0116	0.62	71.7	0.0271	19	111	51.5	2
8	TX11Vsyn0118	1.19	69.4	0.0257	23	189	68.9	1
9	TX11Vsyn0119	0.73	70.5	0.0248	22	132	55.7	2
10	TX11Vsyn0120	1.31	70.7	0.0282	28	176	60.2	2
11	TX11Vsyn0122	1.39	69.6	0.0275	26	185	58.6	1
12	TX11Vsyn0123	1.24	69.8	0.025	27	178	62.1	2
13	TX11Vsyn0124	1.12	68.6	0.0251	22	201	59.8	2
14	TX11Vsyn0127	0.35	69.1	0.0225	18	110	59.3	3
15	TX11Vsyn0130	0.58	72.1	0.022	16	137	44.2	3
16	TX11Vsyn0131	0.98	72.2	0.0264	22	213	67.1	2
17	TX11Vsyn0133	0.94	69.7	0.025	19	204	57.4	2
18	TX11Vsyn0134	0.85	70.2	0.0262	24	125	55.5	2
19	TX11Vsyn0135	0.8	72.7	0.0283	19	157	57.8	1
20	TX11Vsyn0136	0.81	68	0.0254	26	112	62.9	3
21	TX11Vsyn0137	1.23	72.8	0.0269	23	191	57.5	2
22	TX11Vsyn0138	1.45	69.5	0.0336	24	188	67.4	2
23	TX11Vsyn0140	1.59	71.6	0.0361	26	166	67.7	2
24	TX11Vsyn0146	0.88	73	0.0281	21	130	63.2	2
25	TX11Vsyn0153	1.05	71.9	0.024	24	169	56.2	2
26	TX11Vsyn0154	1	71.7	0.0245	22	196	60.6	2
27	TX11Vsyn0156	0.92	69.7	0.0235	24	177	63.6	2
28	TX11Vsyn0158	0.58	73	0.027	22	101	49.8	3
29	TX11Vsyn0159	1.31	71.3	0.023	24	250	60.7	2
30	TX11Vsyn0160	0.51	72.7	0.0294	21	109	54.6	4
31	TX11Vsyn0161	0.29	60.7	0.0185	17	109	59.4	2
32	TX11Vsyn0164	1.26	72.6	0.0258	21	231	59.7	2

Table Continued

ID	Genotype	Grain Yield	Test Weight	Seed Wt	Seeds Head ⁻¹	Head No	HT	HS
33	TX11Vsyn0165	0.98	69.2	0.0264	23	149	65.4	2
34	TX11Vsyn0167	1.06	72.3	0.0297	21	185	60.7	2
35	TX11Vsyn0168	0.74	70.8	0.0257	18	146	52.3	2
36	TX11Vsyn0169	0.66	72.7	0.0285	24	98	56.1	3
37	TX11Vsyn0174	0.79	69.3	0.0277	18	132	51.6	2
38	TX11Vsyn0175	1.4	70.6	0.0264	22	227	61.2	2
39	TX11Vsyn0178	0.77	73.3	0.0255	29	106	56.9	2
40	TX11Vsyn0179	0.8	69.8	0.0242	16	224	54.5	2
41	TX11Vsyn0180	0.57	69.9	0.0244	20	147	55.4	3
42	TX11Vsyn0182	0.78	72.9	0.0266	20	140	66.3	2
43	TX11Vsyn0185	0.79	70.6	0.0249	24	124	60	3
44	TX11Vsyn0188	0.95	70.2	0.0292	20	184	57	3
45	TX11Vsyn0189	0.92	69.9	0.0273	20	167	57.6	2
46	TX11Vsyn0190	1.13	68.6	0.0255	26	185	57.6	2
47	TX11Vsyn0191	1.24	73.8	0.0239	29	175	58.8	2
48	TX11Vsyn0195	0.95	72.5	0.0295	23	135	63.7	2
49	TX11Vsyn0196	1	69.2	0.0283	15	204	60.5	2
50	TX11Vsyn0197	1.14	71.9	0.0277	23	169	60.2	2
51	TX11Vsyn0199	0.62	73.2	0.0269	20	116	56.6	3
52	TX11Vsyn0201	0.75	70.6	0.0288	20	126	57	2
53	TX11Vsyn0208	1.03	70.6	0.0271	14	600	61.6	2
54	TX11Vsyn0211	0.81	72.7	0.0278	19	154	53.1	2
55	TX11Vsyn0212	0.97	67.1	0.0253	28	139	55	2
56	TX11Vsyn0213	1.07	68.6	0.0232	25	158	56.6	2
57	TX11Vsyn0216	1.11	71.1	0.0211	27	200	61	2
58	TX11Vsyn0217	0.76	72.4	0.0263	22	95	59.4	2
59	TX11Vsyn0219	1.46	73.6	0.0279	21	244	60.4	2
60	TX11Vsyn0225	1.23	71.6	0.0277	24	193	55	2
61	TX11Vsyn0226	1.43	74.4	0.0273	22	221	62.6	2
62	TX11Vsyn0228	1.23	73.4	0.0248	18	295	58.8	2
63	TX11Vsyn0229	1.34	72.3	0.0278	20	214	60.2	2
64	TX11Vsyn0230	1.34	71.4	0.0275	21	232	59.7	2
65	TX11Vsyn0232	1.34	73.3	0.0269	24	196	64.7	2
66	TX11Vsyn0234	1.11	72.9	0.0287	22	188	61	2
67	TX11Vsyn0238	1.21	69.7	0.0262	22	211	55.7	2
68	TX11Vsyn0240	0.82	72.6	0.0264	18	153	58.7	2
69	TX11Vsyn0241	1.27	72.4	0.0244	22	251	56.5	2
70	TX11Vsyn0243	1.04	71	0.0263	21	196	71	1
71	TX11Vsyn0253	0.94	72.6	0.0259	19	214	57.2	2
72	TX11Vsyn0261	1.55	72.7	0.0247	27	255	61.5	2

Table Continued

ID	Genotype	Grain Yield	Test Weight	Seed Wt	Seeds Head-1	Head No	HT	HS
73	TX11Vsyn0263	1.15	70.1	0.024	21	229	57.2	2
74	TX11Vsyn0264	1.4	70.2	0.0253	17	297	60.7	2
75	TX11Vsyn0265	1.09	73.5	0.0266	21	219	60.2	2
76	TX11Vsyn0266	0.98	73.6	0.0254	19	211	54.7	2
77	TX11Vsyn0267	1.1	73.1	0.0278	17	251	54.8	2
78	TX11Vsyn0271	1.19	71.8	0.0301	18	249	60.6	2
79	TX11Vsyn0272	1.34	68.3	0.0274	24	214	63.4	2
80	TX11Vsyn0275	1.59	73.8	0.025	23	266	64.2	2
81	TX11Vsyn0277	0.8	70.2	0.0235	17	190	59.3	2
82	TX11Vsyn0279	1.36	72.7	0.0255	20	280	58	3
83	TX11Vsyn0280	0.95	69.3	0.0268	17	213	58.8	2
84	TX11Vsyn0282	0.41	72.4	0.0256	14	134	48.6	4
85	TX11Vsyn0294	0.81	69.9	0.0261	22	131	54.8	2
86	TX11Vsyn0300	0.98	68.2	0.024	24	172	65.2	2
87	TX11Vsyn0303	0.76	71.7	0.0239	24	132	60.2	3
88	TX11Vsyn0305	0.74	69.1	0.0237	20	122	63.3	2
89	TX11Vsyn0306	1.08	68.8	0.0277	25	154	60.1	2
90	TX11Vsyn0308	0.75	70.1	0.0277	21	128	56.5	2
91	TX11Vsyn0309	1.09	71.1	0.0259	22	177	65.7	2
92	TX11Vsyn0312	0.68	71.4	0.0263	22	98	51.7	2
93	TX11Vsyn0313	0.85	71.1	0.0245	28	136	59.2	3
94	TAM111	1	72.9	0.0255	27	135	59.9	2
95	TAM112	1.12	73	0.0254	24	174	57.6	2
96	TAM113	1.06	74.2	0.0249	27	156	55.9	2
97	TAM304	1.28	67.4	0.0223	30	196	54.3	1
98	TAM305	0.93	72.5	0.0222	24	176	49.5	2
99	TAM401	1.06	69.7	0.0218	23	216	60.2	2
100	TAMW101	1.13	70.8	0.0288	18	206	55.9	2
	Mean	1.01	71.1	0.0261	22	181	58.8	2
	Minimum	0.29	60.7	0.0185	14	95	44.2	1
	Maximum	1.59	74.4	0.0361	30	600	71	4
	LSD	0.35	2.7	0.0043	7	149	7.5	1
	CV (%)	17.6	1.9	8.2	16.1	41.5	6.4	12.9

Mean, range, LSD (least significant difference), CV (coefficient of variation) for grain yield, test weight, seed wt. (single seed weight), seeds head⁻¹, head number, height, heading score at Chillicothe, TX in 2014

ID	Genotype	Grain	Test	Seed	Seeds	Head	HT	HS
		Yield t ha ⁻¹	Weight kg hL ⁻¹	Wt g	Head ⁻¹ count	No Heads m ⁻²	cm	scale 1-5
1	TX11Vsyn0101	1.23	71.2	0.0278	24	178	64.8	2
2	TX11Vsyn0103	1.14	68.7	0.0222	27	165	53.7	2
3	TX11Vsyn0110	0.70	65.7	0.0235	21	151	60.0	2
4	TX11Vsyn0111	1.03	73.1	0.0309	20	184	57.4	2
5	TX11Vsyn0112	1.06	70.5	0.0263	25	174	55.4	2
6	TX11Vsyn0113	1.00	73.4	0.0300	20	152	59.8	2
7	TX11Vsyn0116	0.62	71.7	0.0271	19	111	51.5	2
8	TX11Vsyn0118	1.19	69.4	0.0257	23	189	68.9	1
9	TX11Vsyn0119	0.73	70.5	0.0248	22	132	55.7	2
10	TX11Vsyn0120	1.31	70.7	0.0282	28	176	60.2	2
11	TX11Vsyn0122	1.39	69.6	0.0275	26	185	58.6	1
12	TX11Vsyn0123	1.24	69.8	0.0250	27	178	62.1	2
13	TX11Vsyn0124	1.12	68.6	0.0251	22	201	59.8	2
14	TX11Vsyn0127	0.35	69.1	0.0225	18	110	59.3	3
15	TX11Vsyn0130	0.58	72.1	0.0220	16	137	44.2	3
16	TX11Vsyn0131	0.98	72.2	0.0264	22	213	67.1	2
17	TX11Vsyn0133	0.94	69.7	0.0250	19	204	57.4	2
18	TX11Vsyn0134	0.85	70.2	0.0262	24	125	55.5	2
19	TX11Vsyn0135	0.80	72.7	0.0283	19	157	57.8	1
20	TX11Vsyn0136	0.81	68.0	0.0254	26	112	62.9	3
21	TX11Vsyn0137	1.23	72.8	0.0269	23	191	57.5	2
22	TX11Vsyn0138	1.45	69.5	0.0336	24	188	67.4	2
23	TX11Vsyn0140	1.59	71.6	0.0361	26	166	67.7	2
24	TX11Vsyn0146	0.88	73.0	0.0281	21	130	63.2	2
25	TX11Vsyn0153	1.05	71.9	0.0240	24	169	56.2	2
26	TX11Vsyn0154	1.00	71.7	0.0245	22	196	60.6	2
27	TX11Vsyn0156	0.92	69.7	0.0235	24	177	63.6	2
28	TX11Vsyn0158	0.58	73.0	0.0270	22	101	49.8	3
29	TX11Vsyn0159	1.31	71.3	0.0230	24	250	60.7	2
30	TX11Vsyn0160	0.51	72.7	0.0294	21	109	54.6	4
31	TX11Vsyn0161	0.29	60.7	0.0185	17	109	59.4	2
32	TX11Vsyn0164	1.26	72.6	0.0258	21	231	59.7	2

Table Continued

ID	Genotype	Grain Yield	Test Weight	Seed Wt	Seeds Head ⁻¹	Head No	HT	HS
33	TX11Vsyn0165	0.98	69.2	0.0264	23	149	65.4	2
34	TX11Vsyn0167	1.06	72.3	0.0297	21	185	60.7	2
35	TX11Vsyn0168	0.74	70.8	0.0257	18	146	52.3	2
36	TX11Vsyn0169	0.66	72.7	0.0285	24	98	56.1	3
37	TX11Vsyn0174	0.79	69.3	0.0277	18	132	51.6	2
38	TX11Vsyn0175	1.40	70.6	0.0264	22	227	61.2	2
39	TX11Vsyn0178	0.77	73.3	0.0255	29	106	56.9	2
40	TX11Vsyn0179	0.80	69.8	0.0242	16	224	54.5	2
41	TX11Vsyn0180	0.57	69.9	0.0244	20	147	55.4	3
42	TX11Vsyn0182	0.78	72.9	0.0266	20	140	66.3	2
43	TX11Vsyn0185	0.79	70.6	0.0249	24	124	60.0	3
44	TX11Vsyn0188	0.95	70.2	0.0292	20	184	57.0	3
45	TX11Vsyn0189	0.92	69.9	0.0273	20	167	57.6	2
46	TX11Vsyn0190	1.13	68.6	0.0255	26	185	57.6	2
47	TX11Vsyn0191	1.24	73.8	0.0239	29	175	58.8	2
48	TX11Vsyn0195	0.95	72.5	0.0295	23	135	63.7	2
49	TX11Vsyn0196	1.00	69.2	0.0283	15	204	60.5	2
50	TX11Vsyn0197	1.14	71.9	0.0277	23	169	60.2	2
51	TX11Vsyn0199	0.62	73.2	0.0269	20	116	56.6	3
52	TX11Vsyn0201	0.75	70.6	0.0288	20	126	57.0	2
53	TX11Vsyn0208	1.03	70.6	0.0271	14	600	61.6	2
54	TX11Vsyn0211	0.81	72.7	0.0278	19	154	53.1	2
55	TX11Vsyn0212	0.97	67.1	0.0253	28	139	55.0	2
56	TX11Vsyn0213	1.07	68.6	0.0232	25	158	56.6	2
57	TX11Vsyn0216	1.11	71.1	0.0211	27	200	61.0	2
58	TX11Vsyn0217	0.76	72.4	0.0263	22	95	59.4	2
59	TX11Vsyn0219	1.46	73.6	0.0279	21	244	60.4	2
60	TX11Vsyn0225	1.23	71.6	0.0277	24	193	55.0	2
61	TX11Vsyn0226	1.43	74.4	0.0273	22	221	62.6	2
62	TX11Vsyn0228	1.23	73.4	0.0248	18	295	58.8	2
63	TX11Vsyn0229	1.34	72.3	0.0278	20	214	60.2	2
64	TX11Vsyn0230	1.34	71.4	0.0275	21	232	59.7	2
65	TX11Vsyn0232	1.34	73.3	0.0269	24	196	64.7	2
66	TX11Vsyn0234	1.11	72.9	0.0287	22	188	61.0	2
67	TX11Vsyn0238	1.21	69.7	0.0262	22	211	55.7	2
68	TX11Vsyn0240	0.82	72.6	0.0264	18	153	58.7	2
69	TX11Vsyn0241	1.27	72.4	0.0244	22	251	56.5	2
70	TX11Vsyn0243	1.04	71.0	0.0263	21	196	71.0	1
71	TX11Vsyn0253	0.94	72.6	0.0259	19	214	57.2	2
72	TX11Vsyn0261	1.55	72.7	0.0247	27	255	61.5	2

Table Continued

ID	Genotype	Grain Yield	Test Weight	Seed Wt	Seeds Head ⁻¹	Head No	HT	HS
73	TX11Vsyn0263	1.15	70.1	0.0240	21	229	57.2	2
74	TX11Vsyn0264	1.40	70.2	0.0253	17	297	60.7	2
75	TX11Vsyn0265	1.09	73.5	0.0266	21	219	60.2	2
76	TX11Vsyn0266	0.98	73.6	0.0254	19	211	54.7	2
77	TX11Vsyn0267	1.10	73.1	0.0278	17	251	54.8	2
78	TX11Vsyn0271	1.19	71.8	0.0301	18	249	60.6	2
79	TX11Vsyn0272	1.34	68.3	0.0274	24	214	63.4	2
80	TX11Vsyn0275	1.59	73.8	0.0250	23	266	64.2	2
81	TX11Vsyn0277	0.80	70.2	0.0235	17	190	59.3	2
82	TX11Vsyn0279	1.36	72.7	0.0255	20	280	58.0	3
83	TX11Vsyn0280	0.95	69.3	0.0268	17	213	58.8	2
84	TX11Vsyn0282	0.41	72.4	0.0256	14	134	48.6	4
85	TX11Vsyn0294	0.81	69.9	0.0261	22	131	54.8	2
86	TX11Vsyn0300	0.98	68.2	0.0240	24	172	65.2	2
87	TX11Vsyn0303	0.76	71.7	0.0239	24	132	60.2	3
88	TX11Vsyn0305	0.74	69.1	0.0237	20	122	63.3	2
89	TX11Vsyn0306	1.08	68.8	0.0277	25	154	60.1	2
90	TX11Vsyn0308	0.75	70.1	0.0277	21	128	56.5	2
91	TX11Vsyn0309	1.09	71.1	0.0259	22	177	65.7	2
92	TX11Vsyn0312	0.68	71.4	0.0263	22	98	51.7	2
93	TX11Vsyn0313	0.85	71.1	0.0245	28	136	59.2	3
94	TAM111	1.00	72.9	0.0255	27	135	59.9	2
95	TAM112	1.12	73.0	0.0254	24	174	57.6	2
96	TAM113	1.06	74.2	0.0249	27	156	55.9	2
97	TAM304	1.28	67.4	0.0223	30	196	54.3	1
98	TAM305	0.93	72.5	0.0222	24	176	49.5	2
99	TAM401	1.06	69.7	0.0218	23	216	60.2	2
100	TAMW101	1.13	70.8	0.0288	18	206	55.9	2
	Mean	1.01	71.1	0.0261	22	181	58.8	2
	Minimum	0.29	60.7	0.0185	14	95	44.2	1
	Maximum	1.59	74.4	0.0361	30	600	71.0	4
	LSD	0.35	2.7	0.0043	7	149	7.5	1
	CV (%)	17.6	1.9	8.2	16.1	41.5	6.4	12.9

Mean, range, LSD (least significant difference), CV (coefficient of variation) for grain yield, test weight, seed wt. (single seed weight), seeds head⁻¹, head number, height, heading score at Diyarbakir, Turkey in 2014

ID	Genotype	Grain Yield	Test Weight	Seed Wt.	Seeds Head ⁻¹	Head No	HT	HS
		t ha ⁻¹	kg hL ⁻¹	g	count	Heads m ⁻²	cm	1-5
1	TX11Vsyn0101	6.76	85.7	0.0352	41	467	98.1	3
2	TX11Vsyn0103	4.13	82.6	0.0268	35	603	82.7	3
3	TX11Vsyn0110	5.41	83.4	0.0351	33	444	94.7	3
4	TX11Vsyn0111	5.12	84.0	0.0376	31	412	92.6	2
5	TX11Vsyn0112	5.24	83.4	0.0311	40	421	93.1	5
6	TX11Vsyn0113	5.65	85.0	0.0389	39	393	102.1	3
7	TX11Vsyn0116	4.96	82.2	0.0344	30	476	82.0	4
8	TX11Vsyn0118	5.40	81.4	0.0328	42	377	94.4	3
9	TX11Vsyn0119	5.73	81.4	0.0334	40	437	92.8	4
10	TX11Vsyn0120	6.15	82.8	0.0388	47	343	102.1	3
11	TX11Vsyn0122	6.28	84.5	0.0333	49	401	105.5	3
12	TX11Vsyn0123	6.03	82.1	0.0313	47	403	98.3	3
13	TX11Vsyn0124	4.99	79.3	0.0331	36	412	94.6	5
14	TX11Vsyn0127	4.72	81.2	0.0392	41	281	99.4	5
15	TX11Vsyn0130	5.40	83.0	0.0275	44	416	80.7	4
16	TX11Vsyn0131	5.02	85.6	0.0370	36	379	101.3	2
17	TX11Vsyn0133	5.79	85.7	0.0349	37	445	93.6	3
18	TX11Vsyn0134	4.89	83.0	0.0342	30	480	89.7	2
19	TX11Vsyn0135	5.37	82.9	0.0365	41	339	85.9	3
20	TX11Vsyn0136	5.83	81.0	0.0359	44	376	116.5	5
21	TX11Vsyn0137	5.88	82.4	0.0379	37	422	93.5	5
22	TX11Vsyn0138	5.27	85.4	0.0444	25	500	104.7	1
23	TX11Vsyn0140	5.41	84.9	0.0489	28	391	107.1	2
24	TX11Vsyn0146	4.60	85.0	0.0367	27	475	102.8	4
25	TX11Vsyn0153	4.43	80.6	0.0288	35	457	72.9	3
26	TX11Vsyn0154	4.57	85.2	0.0339	43	341	83.6	3
27	TX11Vsyn0156	5.54	85.0	0.0372	42	351	103.9	3
28	TX11Vsyn0158	5.15	85.3	0.0391	38	356	94.0	4
29	TX11Vsyn0159	5.70	81.2	0.0276	51	385	85.7	2
30	TX11Vsyn0160	4.89	86.1	0.0362	37	364	88.8	3
31	TX11Vsyn0161	4.43	82.1	0.0317	42	368	87.2	4
32	TX11Vsyn0164	3.93	81.4	0.0312	36	359	78.3	3
33	TX11Vsyn0165	6.02	82.5	0.0364	42	378	106.2	3
34	TX11Vsyn0167	5.81	84.9	0.0376	35	445	91.0	3
35	TX11Vsyn0168	4.66	82.5	0.0323	33	467	93.6	3
36	TX11Vsyn0169	4.62	83.7	0.0342	40	331	94.6	3

Table Continued

ID	Genotype	Grain Yield	Test Weight	Seed Wt.	Seeds Head ⁻¹	Head No	HT	HS
37	TX11Vsyn0174	4.67	83.2	0.0402	30	430	97.8	4
38	TX11Vsyn0175	5.74	84.2	0.0350	32	464	95.3	3
39	TX11Vsyn0178	4.81	83.8	0.0307	32	477	85.7	3
40	TX11Vsyn0179	6.23	83.4	0.0334	29	608	91.6	2
41	TX11Vsyn0180	5.23	82.9	0.0300	46	389	93.6	4
42	TX11Vsyn0182	6.18	87.1	0.0400	39	415	101.3	2
43	TX11Vsyn0185	6.31	86.0	0.0344	41	490	89.1	3
44	TX11Vsyn0188	4.73	80.8	0.0334	33	425	89.8	5
45	TX11Vsyn0189	5.58	82.0	0.0335	35	514	88.8	3
46	TX11Vsyn0190	4.51	78.8	0.0272	49	386	90.1	4
47	TX11Vsyn0191	6.00	85.4	0.0326	34	529	95.9	3
48	TX11Vsyn0195	6.14	83.8	0.0388	43	371	109.0	2
49	TX11Vsyn0196	4.43	80.8	0.0407	37	287	97.6	2
50	TX11Vsyn0197	5.75	82.8	0.0381	38	415	103.1	2
51	TX11Vsyn0199	4.90	81.8	0.0320	35	446	103.6	5
52	TX11Vsyn0201	4.95	81.4	0.0393	32	379	97.0	3
53	TX11Vsyn0208	5.42	86.1	0.0399	35	413	92.7	3
54	TX11Vsyn0211	4.48	82.2	0.0325	36	386	84.0	3
55	TX11Vsyn0212	4.44	79.8	0.0288	45	326	86.6	3
56	TX11Vsyn0213	5.27	81.6	0.0337	41	382	88.4	3
57	TX11Vsyn0216	5.32	83.4	0.0302	45	410	83.3	4
58	TX11Vsyn0217	5.36	83.2	0.0433	33	373	101.3	3
59	TX11Vsyn0219	5.72	81.2	0.0315	36	498	92.6	4
60	TX11Vsyn0225	5.62	85.7	0.0356	36	476	85.5	2
61	TX11Vsyn0226	5.40	84.8	0.0357	35	426	96.7	2
62	TX11Vsyn0228	5.48	86.5	0.0310	41	468	86.1	1
63	TX11Vsyn0229	5.03	82.4	0.0342	36	405	97.4	3
64	TX11Vsyn0230	5.38	86.3	0.0372	33	483	85.5	3
65	TX11Vsyn0232	5.19	85.8	0.0356	38	418	108.1	2
66	TX11Vsyn0234	6.04	85.7	0.0326	37	486	87.8	2
67	TX11Vsyn0238	5.27	80.0	0.0348	42	356	89.9	2
68	TX11Vsyn0240	4.55	87.1	0.0359	29	475	111.1	2
69	TX11Vsyn0241	5.75	81.6	0.0285	42	475	82.0	2
70	TX11Vsyn0243	4.55	85.0	0.0368	27	625	110.5	2
71	TX11Vsyn0253	5.21	84.6	0.0293	26	673	92.1	2
72	TX11Vsyn0261	5.16	84.0	0.0331	43	362	79.0	3
73	TX11Vsyn0263	6.36	83.2	0.0334	37	498	97.9	3
74	TX11Vsyn0264	5.81	81.7	0.0351	37	452	94.2	3
75	TX11Vsyn0265	5.73	82.1	0.0338	33	504	99.2	3
76	TX11Vsyn0266	5.33	82.5	0.0324	38	444	87.4	2
77	TX11Vsyn0267	5.80	85.4	0.0326	39	452	88.4	3

Table Continued

ID	Genotype	Grain Yield	Test Weight	Seed Wt.	Seeds Head ⁻¹	Head No	HT	HS
78	TX11Vsyn0271	5.95	85.0	0.0327	38	486	103.0	2
79	TX11Vsyn0272	6.17	82.0	0.0332	37	523	87.5	2
80	TX11Vsyn0275	5.74	84.8	0.0311	37	514	91.4	2
81	TX11Vsyn0277	5.67	82.9	0.0338	42	426	89.8	3
82	TX11Vsyn0279	5.49	84.9	0.0313	41	418	87.3	3
83	TX11Vsyn0280	4.95	79.7	0.0284	33	509	95.6	2
84	TX11Vsyn0282	4.42	81.7	0.0298	32	446	96.3	4
85	TX11Vsyn0294	5.27	79.8	0.0313	48	363	89.7	2
86	TX11Vsyn0300	6.01	82.6	0.0326	46	396	103.1	3
87	TX11Vsyn0303	3.86	83.2	0.0271	40	376	118.2	5
88	TX11Vsyn0305	3.88	78.1	0.0285	36	372	82.0	4
89	TX11Vsyn0306	4.85	82.8	0.0337	38	369	90.5	4
90	TX11Vsyn0308	5.02	82.5	0.0352	38	389	93.7	4
91	TX11Vsyn0309	5.96	83.4	0.0342	40	423	102.8	3
92	TX11Vsyn0312	5.83	84.5	0.0292	39	529	93.6	3
93	TX11Vsyn0313	4.69	83.7	0.0337	45	348	126.9	4
94	TAM111	4.77	82.8	0.0319	44	343	90.5	3
95	TAM112	6.21	85.0	0.0298	36	566	81.4	2
96	TAM113	6.59	87.3	0.0335	33	558	87.9	3
97	TAM304	5.83	82.2	0.0286	46	414	77.6	3
98	TAM305	5.48	85.0	0.0360	34	447	88.9	2
99	TAM401	6.39	83.3	0.0287	56	441	92.1	2
100	TAMW101	4.33	84.9	0.0369	29	432	82.9	3
	Mean	5.33	83.3	0.0340	38	431	93.8	3
	Minimum	3.86	78.1	0.0268	25	281	72.9	1
	Maximum	6.76	87.3	0.0489	56	673	126.9	5
	LSD	1.47	.	0.0055	12	195	15.1	1
	CV (%)	13.9	.	8.2	16.4	22.7	8.1	22.6

Mean, range, LSD (least significant difference), CV (coefficient of variation) for grain yield, test weight, seed wt. (single seed weight), seeds head⁻¹, head number, height, heading score at College Station, TX in 2014

ID	Genotype	Grain	Test	Seed	Seeds	Head	HT	HS
		Yield	Weight	Wt	Head ⁻¹	No		
		t ha ⁻¹	kg hL ⁻¹	g	count	Heads m ⁻²	cm	1-5
1	TX11Vsyn0101	1.94	71.9	0.0307	45	139	90.5	3
2	TX11Vsyn0103	1.79	67.4	0.0218	47	180	84.4	3
3	TX11Vsyn0110	2.42	72.9	0.0285	40	216	99.0	3
4	TX11Vsyn0111	1.68	72.8	0.0367	35	129	79.6	2
5	TX11Vsyn0112	0.88	70.2	0.0282	42	75	91.1	3
6	TX11Vsyn0113	2.02	73.5	0.0344	41	147	85.5	2
7	TX11Vsyn0116	2.09	69.4	0.0285	44	170	90.5	4
8	TX11Vsyn0118	2.15	70.1	0.0306	47	151	87.9	2
9	TX11Vsyn0119	1.51	71.0	0.0286	52	101	80.1	3
10	TX11Vsyn0120	2.00	69.6	0.0324	48	125	97.1	3
11	TX11Vsyn0122	2.05	70.3	0.0274	52	144	87.0	5
12	TX11Vsyn0123	1.94	72.1	0.0307	44	145	93.0	2
13	TX11Vsyn0124	1.66	72.3	0.0285	46	134	89.0	3
14	TX11Vsyn0127	2.56	73.2	0.0303	50	174	99.0	3
15	TX11Vsyn0130	1.67	72.1	0.0266	49	127	88.0	3
16	TX11Vsyn0131	2.00	72.6	0.0319	33	178	84.9	3
17	TX11Vsyn0133	1.66	72.8	0.0301	42	130	90.0	3
18	TX11Vsyn0134	1.95	70.4	0.0288	39	175	93.0	2
19	TX11Vsyn0135	1.75	72.6	0.0320	45	130	81.5	3
20	TX11Vsyn0136	2.16	70.3	0.0292	42	177	99.0	5
21	TX11Vsyn0137	1.64	72.4	0.0304	43	134	89.0	5
22	TX11Vsyn0138	2.11	67.3	0.0354	41	165	96.0	3
23	TX11Vsyn0140	1.96	72.4	0.0407	34	147	88.5	2
24	TX11Vsyn0146	1.39	72.6	0.0311	39	122	91.0	4
25	TX11Vsyn0153	1.80	67.5	0.0259	43	160	82.0	3
26	TX11Vsyn0154	2.24	70.8	0.0262	42	210	88.0	3
27	TX11Vsyn0156	2.00	72.1	0.0283	55	131	85.9	2
28	TX11Vsyn0158	2.61	70.5	0.0316	42	199	82.0	3
29	TX11Vsyn0159	1.70	70.6	0.0209	55	143	86.0	4
30	TX11Vsyn0160	1.92	71.8	0.0311	42	150	90.0	5
31	TX11Vsyn0161	2.01	71.7	0.0259	48	165	91.0	5
32	TX11Vsyn0164	2.46	70.6	0.0279	48	182	85.9	3
33	TX11Vsyn0165	2.49	69.3	0.0282	47	199	90.0	3
34	TX11Vsyn0167	2.31	72.3	0.0277	41	206	89.5	3
35	TX11Vsyn0168	2.36	66.3	0.0277	44	188	89.1	3
36	TX11Vsyn0169	1.50	72.7	0.0319	39	131	86.1	3

Table Continued

ID	Genotype	Grain Yield	Test Weight	Seed Wt	Seeds Head ⁻¹	Head No	HT	H S
37	TX11Vsyn0174	1.60	68.5	0.0274	40	143	91.0	4
38	TX11Vsyn0175	1.27	68.8	0.0259	39	127	89.0	3
39	TX11Vsyn0178	2.44	70.8	0.0254	45	210	88.4	3
40	TX11Vsyn0179	1.57	71.4	0.0275	53	131	90.4	3
41	TX11Vsyn0180	1.42	70.5	0.0251	43	136	75.0	3
42	TX11Vsyn0182	2.02	70.1	0.0298	40	169	88.0	2
43	TX11Vsyn0185	1.31	71.3	0.0282	43	108	81.6	4
44	TX11Vsyn0188	1.68	66.0	0.0282	41	142	86.0	5
45	TX11Vsyn0189	2.63	69.8	0.0301	36	245	83.5	2
46	TX11Vsyn0190	1.57	64.6	0.0242	45	158	91.0	3
47	TX11Vsyn0191	2.67	73.5	0.0277	46	215	90.0	2
48	TX11Vsyn0195	2.46	71.7	0.0303	43	189	88.5	3
49	TX11Vsyn0196	1.57	70.8	0.0344	36	130	90.0	3
50	TX11Vsyn0197	1.85	70.7	0.0321	43	138	97.9	3
51	TX11Vsyn0199	2.43	71.9	0.0300	45	185	90.4	5
52	TX11Vsyn0201	1.65	69.2	0.0317	35	146	85.0	3
53	TX11Vsyn0208	1.41	73.3	0.0305	46	114	94.0	3
54	TX11Vsyn0211	1.70	72.1	0.0297	44	133	90.0	3
55	TX11Vsyn0212	2.07	66.3	0.0260	51	158	87.0	3
56	TX11Vsyn0213	2.47	71.0	0.0287	49	189	90.0	3
57	TX11Vsyn0216	1.96	70.1	0.0240	43	191	85.0	3
58	TX11Vsyn0217	1.41	71.5	0.0348	39	101	92.0	2
59	TX11Vsyn0219	1.85	70.6	0.0321	33	159	76.5	2
60	TX11Vsyn0225	1.92	69.9	0.0290	40	171	74.1	3
61	TX11Vsyn0226	1.78	70.6	0.0298	37	152	81.0	2
62	TX11Vsyn0228	1.46	76.1	0.0322	37	123	79.0	3
63	TX11Vsyn0229	1.68	70.8	0.0285	41	151	90.0	2
64	TX11Vsyn0230	2.28	71.6	0.0292	42	187	88.5	3
65	TX11Vsyn0232	1.29	69.8	0.0269	30	175	91.0	2
66	TX11Vsyn0234	2.07	71.9	0.0278	33	203	72.0	2
67	TX11Vsyn0238	1.60	68.1	0.0266	45	135	75.4	3
68	TX11Vsyn0240	1.48	71.7	0.0282	33	157	91.0	2
69	TX11Vsyn0241	1.36	69.1	0.0267	32	169	83.0	2
70	TX11Vsyn0243	1.87	67.5	0.0340	38	149	85.0	2
71	TX11Vsyn0253	1.27	71.5	0.0280	37	127	84.1	2
72	TX11Vsyn0261	1.22	72.6	0.0313	42	94	86.4	3
73	TX11Vsyn0263	1.92	70.8	0.0296	40	164	86.9	3
74	TX11Vsyn0264	1.99	68.9	0.0285	43	162	87.1	2
75	TX11Vsyn0265	2.17	73.1	0.0339	43	150	83.5	2
76	TX11Vsyn0266	2.42	69.1	0.0294	44	182	89.0	2
77	TX11Vsyn0267	1.72	68.3	0.0253	46	154	79.5	2

Table Continued

ID	Genotype	Grain Yield	Test Weight	Seed Wt	Seeds Head ⁻¹	Head No	HT	H S
78	TX11Vsyn0271	1.26	72.3	0.0301	36	117	86.0	3
79	TX11Vsyn0272	1.89	70.1	0.0294	41	163	82.9	2
80	TX11Vsyn0275	1.60	72.3	0.0276	42	138	83.5	2
81	TX11Vsyn0277	1.83	72.7	0.0319	43	141	86.5	4
82	TX11Vsyn0279	1.62	69.8	0.0244	43	157	81.0	3
83	TX11Vsyn0280	0.68	66.5	0.0266	34	65	94.0	2
84	TX11Vsyn0282	2.26	72.6	0.0249	36	248	94.5	5
85	TX11Vsyn0294	2.49	66.6	0.0247	47	217	86.5	2
86	TX11Vsyn0300	1.85	66.9	0.0244	54	136	81.0	3
87	TX11Vsyn0303	1.82	74.9	0.0292	46	139	99.6	5
88	TX11Vsyn0305	2.55	67.1	0.0246	44	244	87.0	3
89	TX11Vsyn0306	2.65	71.6	0.0308	48	191	90.1	3
90	TX11Vsyn0308	2.27	70.7	0.0286	50	156	86.0	2
91	TX11Vsyn0309	1.41	66.6	0.0287	41	122	96.0	2
92	TX11Vsyn0312	1.72	66.4	0.0229	42	191	89.0	3
93	TX11Vsyn0313	1.72	72.8	0.0277	41	150	100	5
94	TAM111	2.23	72.8	0.0262	46	186	85.0	3
95	TAM112	1.63	70.2	0.0267	37	172	76.5	2
96	TAM113	1.68	72.5	0.0275	43	151	81.0	4
97	TAM304	1.51	68.5	0.0253	44	150	79.0	3
98	TAM305	1.93	70.3	0.0266	48	156	81.1	3
99	TAM401	2.02	70.6	0.0263	49	166	86.0	3
100	TAMW101	2.06	74.6	0.0340	31	205	81.0	5
	Mean	1.88	70.6	0.0288	43	158	87.2	3
	Minimum	0.68	64.6	0.0209	30	65	72.0	2
	Maximum	2.67	76.1	0.0407	55	248	100.0	5
	LSD	0.97	1.9	0.0044	11	94	2.6	1
	CV (%)	26.1	1.4	7.7	13.1	29.8	1.5	10

Mean, range, LSD (least significant difference), CV (coefficient of variation) for grain yield, test weight, seed wt. (single seed weight), seeds head⁻¹, head number, height, heading score at College Station, TX in 2013

ID	Genotype	Grain	Test	Seed	Seeds	Head	Heig	HS
		Yield	Weight	Wt.	Head ⁻¹	No.	ht	
		t ha ⁻¹	kg hL ⁻¹	g	count	Heads m ⁻²	cm	1-5
1	TX11Vsyn0101	3.30	77.1	0.0405	36	225	97.5	3
2	TX11Vsyn0103	2.90	75.8	0.0340	43	202	97.3	3
3	TX11Vsyn0110	2.30	77.6	0.0321	47	151	94.2	4
4	TX11Vsyn0111	2.70	76.8	0.0403	37	188	93.6	3
5	TX11Vsyn0112	3.20	76.3	0.0306	41	253	88.8	4
6	TX11Vsyn0113	2.60	77.0	0.0388	40	166	103.8	3
7	TX11Vsyn0116	3.20	74.2	0.0337	40	248	103.1	3
8	TX11Vsyn0118	2.30	75.7	0.0343	35	197	95.3	3
9	TX11Vsyn0119	3.30	76.4	0.0306	44	251	98.1	3
10	TX11Vsyn0120	2.60	76.7	0.0398	39	159	106.2	4
11	TX11Vsyn0122	3.30	78.4	0.0381	43	202	101.2	3
12	TX11Vsyn0123	3.00	75.5	0.0355	39	217	99.9	3
13	TX11Vsyn0124	2.20	76.2	0.0374	33	193	88.3	4
14	TX11Vsyn0127	3.10	76.8	0.0347	34	274	85.9	4
15	TX11Vsyn0130	2.80	77.5	0.0340	36	234	95.3	4
16	TX11Vsyn0131	3.10	74.6	0.0362	36	248	95.7	2
17	TX11Vsyn0133	2.90	77.0	0.0355	38	220	99.6	4
18	TX11Vsyn0134	3.00	77.3	0.0343	38	231	92.0	3
19	TX11Vsyn0135	3.10	89.6	0.0409	38	215	102.1	4
20	TX11Vsyn0136	2.60	76.0	0.0378	43	160	116.2	3
21	TX11Vsyn0137	3.20	77.5	0.0340	39	245	90.1	3
22	TX11Vsyn0138	3.00	76.8	0.0383	41	185	96.2	3
23	TX11Vsyn0140	3.10	76.6	0.0409	36	216	102.3	4
24	TX11Vsyn0146	3.30	76.4	0.0402	35	248	105.1	4
25	TX11Vsyn0153	2.90	77.5	0.0378	32	244	94.7	3
26	TX11Vsyn0154	2.50	74.7	0.0349	37	200	96.6	4
27	TX11Vsyn0156	1.70	77.5	0.0397	40	114	101.2	3
28	TX11Vsyn0158	2.90	76.5	0.0376	37	216	106.8	3
29	TX11Vsyn0159	1.60	76.1	0.0364	35	137	103.6	3
30	TX11Vsyn0160	2.30	78.1	0.0319	42	183	94.9	2
31	TX11Vsyn0161	3.00	75.1	0.0338	39	229	96.1	4
32	TX11Vsyn0164	3.10	76.9	0.0342	38	247	93.5	3
33	TX11Vsyn0165	2.50	76.0	0.0366	32	228	99.2	4
34	TX11Vsyn0167	3.00	76.4	0.0355	39	219	103.4	4
35	TX11Vsyn0168	2.20	74.6	0.0345	39	173	87.5	3
36	TX11Vsyn0169	1.90	77.5	0.0407	40	117	99.0	2

Table Continued

ID	Genotype	Grain Yield	Test Weight	Seed Wt.	Seeds Head ⁻¹	Head No.	Height	HS
37	TX11Vsyn0174	1.90	73.9	0.0400	32	155	104.7	4
38	TX11Vsyn0175	1.70	76.9	0.0391	36	125	99.4	4
39	TX11Vsyn0178	2.70	76.5	0.0326	40	205	94.6	3
40	TX11Vsyn0179	2.40	76.1	0.0391	32	205	101.6	3
41	TX11Vsyn0180	1.90	75.2	0.0373	36	138	94.4	2
42	TX11Vsyn0182	3.40	79.8	0.0373	43	219	108.1	3
43	TX11Vsyn0185	2.70	75.7	0.0347	41	196	96.2	4
44	TX11Vsyn0188	2.90	74.7	0.0415	31	218	103.0	3
45	TX11Vsyn0189	2.70	75.0	0.0345	41	193	99.0	3
46	TX11Vsyn0190	2.70	75.9	0.0364	36	219	96.2	3
47	TX11Vsyn0191	4.10	90.7	0.0361	37	310	98.1	3
48	TX11Vsyn0195	2.70	69.9	0.0400	38	186	107.9	4
49	TX11Vsyn0196	2.40	76.4	0.0412	33	187	101.8	4
50	TX11Vsyn0197	3.00	77.7	0.0366	38	222	104.4	4
51	TX11Vsyn0199	3.30	76.0	0.0359	37	252	107.2	3
52	TX11Vsyn0201	2.40	74.9	0.0390	31	205	92.7	3
53	TX11Vsyn0208	3.20	76.3	0.0328	35	252	94.7	4
54	TX11Vsyn0211	3.70	77.0	0.0367	38	272	94.0	3
55	TX11Vsyn0212	3.10	75.0	0.0350	39	238	92.7	3
56	TX11Vsyn0213	3.90	79.0	0.0330	40	280	93.5	3
57	TX11Vsyn0216	2.60	76.2	0.0325	39	210	98.2	4
58	TX11Vsyn0217	2.40	76.0	0.0371	35	199	102.5	4
59	TX11Vsyn0219	3.10	75.4	0.0337	43	214	99.8	3
60	TX11Vsyn0225	3.20	75.8	0.0381	36	242	98.1	2
61	TX11Vsyn0226	3.40	77.8	0.0350	40	249	90.7	3
62	TX11Vsyn0228	3.10	77.1	0.0354	30	264	96.8	4
63	TX11Vsyn0229	3.60	78.2	0.0326	40	272	92.7	3
64	TX11Vsyn0230	2.70	76.4	0.0403	40	152	96.0	3
65	TX11Vsyn0232	2.20	75.0	0.0393	40	129	104.9	3
66	TX11Vsyn0234	3.60	77.7	0.0357	45	233	99.2	3
67	TX11Vsyn0238	2.80	76.7	0.0352	43	179	92.9	3
68	TX11Vsyn0240	2.70	74.4	0.0311	41	208	104.8	3
69	TX11Vsyn0241	2.40	76.7	0.0369	36	187	92.7	4
70	TX11Vsyn0243	1.50	75.1	0.0419	35	106	103.8	1
71	TX11Vsyn0253	2.20	76.3	0.0374	32	198	104.2	2
72	TX11Vsyn0261	3.90	76.4	0.0352	44	259	99.5	3
73	TX11Vsyn0263	3.00	75.8	0.0393	36	225	101.8	2
74	TX11Vsyn0264	2.70	75.3	0.0354	35	226	101.4	3
75	TX11Vsyn0265	2.80	74.9	0.0352	36	235	96.0	3
76	TX11Vsyn0266	3.10	76.0	0.0335	35	277	89.4	2
77	TX11Vsyn0267	2.80	76.1	0.0354	39	198	99.2	3

Table Continued

ID	Genotype	Grain Yield	Test Weight	Seed Wt.	Seeds Head ⁻¹	Head No.	Height	HS
78	TX11Vsyn0271	2.70	74.9	0.0295	40	222	94.0	2
79	TX11Vsyn0272	2.70	74.6	0.0378	32	223	113.4	3
80	TX11Vsyn0275	3.30	76.6	0.0345	37	280	104.7	4
81	TX11Vsyn0277	3.20	75.2	0.0347	37	254	98.8	3
82	TX11Vsyn0279	2.50	68.8	0.0347	37	199	99.5	3
83	TX11Vsyn0280	2.40	74.7	0.0347	33	231	102.9	3
84	TX11Vsyn0282	1.70	71.1	0.0364	37	130	103.1	4
85	TX11Vsyn0294	4.00	76.2	0.0333	44	280	93.8	5
86	TX11Vsyn0300	2.40	75.6	0.0354	44	151	104.9	3
87	TX11Vsyn0303	2.70	80.1	0.0383	37	198	103.8	3
88	TX11Vsyn0305	2.30	79.0	0.0318	40	188	102.0	5
89	TX11Vsyn0306	2.50	76.9	0.0359	40	166	88.3	3
90	TX11Vsyn0308	3.10	76.5	0.0354	39	228	99.5	2
91	TX11Vsyn0309	2.90	76.4	0.0328	41	223	101.8	3
92	TX11Vsyn0312	2.10	76.8	0.0350	39	154	93.4	4
93	TX11Vsyn0313	2.00	75.5	0.0369	46	129	104.0	3
94	TAM111	4.10	78.3	0.0334	39	309	99.0	4
95	TAM112	2.70	65.7	0.0294	33	211	83.6	3
96	TAM113
97	TAM304
98	TAM305
99	TAM401	1.10	74.1	0.0379	36	89	98.2	2
100	TAMW101
	Mean	2.65	76.1	0.0361	37	203	99.0	3
	Minimum	1.10	65.7	0.0294	30	89	83.6	1
	Maximum	4.10	90.7	0.0419	47	310	116.2	5
	LSD
	CV (%)	20.1	6.0	8.0	10.0	20.4	5.9	22.6

Appendix II

```
*/test of normality/*;
proc sort ; by loc rep;
Proc univariate normal;
Var GY TW SKW SeedsSpike SpikeNo Heading Height Agscore YRCI LRCI YR LR;
run;
*/test of homogeniety of varaiances/*;
proc glm;
proc sort ; by loc rep;
class Genotype;
model GY TW SKW SeedsSpike SpikeNo Heading HeadingScore Height YRCI LRCI
YR LR = Genotype / ss3;
means Genotype / hovtest=levene (type=abs);
means Genotype / hovtest=BARTLETT;
ODS Graphics off;
run;
*/individual location analysis with covariate/*;
proc glm; by loc;
class Genotype rep block;
model GY TW SKW SeedsSpike SpikeNo Heading Headingscore Height Agscore YRCI
LRCI YR LR = Genotype rep block(rep)covariate;
random rep block(rep);
means Genotype / tukey;
lsmeans Genotype;
ODS Graphics off;
run;
*/combined locations analysis with covariate/*;
proc glm;
class loc Genotype rep block;
model GY TW SKW SeedsSpike SpikeNo Heading HeadingScore Height Agscore
YRCI LRCI YR LR = loc Genotype rep block(rep) Genotype*loc rep(loc)covariate;
random rep block(rep) rep(loc);
means Genotype / tukey;
lsmeans Genotype;
ODS Graphics off;
run;
*/correlation analysis/*;
proc corr;
var GY TW SKW SeedsSpike SpikeNo Heading HeadingScore Height YRCI LRCI YR
LR;
run;

*/correlation analysis few/*;
```



```

proc corr;
var GY TW SKW SeedsSpike SpikeNo;
run;

*/regression analysis*/;
proc reg;
model GY = TW SKW SeedsSpike SpikeNo Heading HeadingScore Height YRCI
LRCI YR LR / noint;
run;

*/regression analysis*/;
proc reg;
model GY = TW SKW SeedsSpike SpikeNo / noint;
run;
quit;

*/rcbd combined analysis*/;
;
proc glm;
class ID Rep;
model GY TW SKW SeedsSpike SpikeNo Height HeadingScore LRCI= ID Rep;
lsmeans ID/stderr pdiff;
run;
*/correlation analysis*/;
proc corr;
var GY TW SKW SeedsSpike SpikeNo HeadingScore Height LRCI;
proc corr;
*/regression analysis*/;
proc reg;
model GY = TW SKW SeedsSpike SpikeNo HeadingScore Height LRCI/ noint;
run;
quit;

*/Augmented design*/;
data BS1Aug;
input Genotype      Rep    GY    TW    SKW  SeedsSpike  SpikeNo
      Height Heading    HeadingScore LRCI;
if (Genotype>210)then new=0; else new=1;
if (new) then check=999; else check=Genotype;
datalines;
;
ods html; ods graphics on;

```

```

Proc glm;
class Rep Genotype;
model GY SKW SeedsSpike SpikeNo TW Height Heading HeadingScore
LRCI=Genotype/solution;
lsmeans Genotype;
run;
proc mixed;
class Genotype check;
model GY SKW SeedsSpike SpikeNo TW Height Heading HeadingScore
LRCI=check/solution;
random Genotype/solution;
lsmeans check;
run;
ods graphics off;
ods html close;

```

```

*/genetic correlations */;
dm 'log;clear;output;clear;';
options ps=500 ls=78;

```

```

data dmdii1;
infile 'C:\Users\bkreddy\Desktop\Gencorr\CH_1.prn' firstobs=2;
*FILENAME DATA1 DDE "EXCEL|[DMDII1.xls]Master!R2C1:R1054C40";/*This
option is used to give a file name and where the data begins including headers,The file
has to be open when using this option*/
*INFILE DATA1 NOTAB DLM= '09'X DSD MISSOEVER lrecl = 10240;/*SAS expects
to see a TAB character placed between each variable that is communicated across the
DDE link. Similarly, SAS places a TAB
character between variables when data are transmitted across the link. When the
NOTAB option is placed in a FILENAME statement that uses the DDE device-type
keyword, SAS accepts character delimiters
other than tabs between variables.The NOTAB option can also be used to store full
character strings, including embedded blanks, in a single spreadsheet cell. For example,
if a link is established
between SAS and the Excel application, and a SAS variable contains a character string
with embedded blanks, each word of the character string is normally stored in a single
cell. */

```

```

INPUT Rep Block Entry GY TW SKW SS SN HT HS;
*USE DATA FROM ONLY ONE ENVIRONMENT FOR THIS EXAMPLE!;
data gencorr1; set dmdii1;
*if entry <70; proc print;

```

```

*first, estimate variance components for each trait separately to compare to multivariate
analysis below;
%macro varcomp(trait);
proc mixed data = gencorr1;
class Rep Block Entry;
model &trait = ;
random rep block(rep) entry;
*also check effect of setting reps and blocks fixed on other variance components;
proc mixed data = gencorr1;
class rep block entry;
model &trait = rep block(rep);
random entry;
run;
%mend;

% varcomp(GY);
% varcomp(TW);
% varcomp(SKW);
% varcomp(SS);
% varcomp(SN);
% varcomp(HT);
% varcomp(HS);

*restructure data set for multivariate reml analysis;
data gencorr2; length trait $ 5; set gencorr1;
trait = "GY"; y = GY; output;
trait = "TW"; y = TW; output;
trait = "SKW"; y = SKW; output;
trait = "SS"; y = SS; output;
trait = "SN"; y = SN; output;
trait = "HT"; y = HT; output;
trait = "HS"; y = HS; output;

drop GY TW SKW SS SN HT HS;

* analyze variables pair-wise;

%macro corr(trait1, trait2);
data traits; set gencorr2; if trait = "&trait1" or trait = "&trait2";
proc mixed asycov data = traits;
class trait rep block entry;
model y = rep(trait) block(rep*trait);
random trait/subject = entry type = un;

```

```

repeated trait/ sub = rep*entry type = un;
ods output covparms = estmat; ods output asycov = covmat;
run;
proc iml;
use estmat; read all into e;
use covmat; read all into cov;
* Note that SAS introduces an extra first column into the covariance matrix which must
be removed;
C = cov(|1:nrow(cov), 2:ncol(cov)|);
* Obtain genotypic and phenotypic covariance and variance components;
CovG = e(|2,1|);
VG1 = e(|1,1|);
VG2 = e(|3,1|);
CovP = CovG + e(|5,1|);
VP1 = VG1 + e(|4,1|);
VP2 = VG2 + e(|6,1|);
* Create a module called "correl" that will estimate genotypic and phenotypic
correlations
and their standard errors;
start correl(C, CovG, VG1, VG2, CovP, VP1, VP2, RG, RP, SERG, SERP);
RG = CovG/sqrt(VG1*VG2);
*Make the derivative vector for rg, note that the order of the rows and columns of the
variance
covariance matrix is VG1, CovG, VG2, VError1, CovError, VError2;
dg = (-1/(2*VG1))/(1/CovG)/(-1/(2*VG2))/0/0/0;
varrg = (RG**2)*dg`*C*dg; serg = sqrt(varrg);
RP = CovP/sqrt(VP1*VP2);
*Make the derivate vector for rp;
d1p = -1/(2*VP1);
d2p = 1/CovP;
d3p = -1/(2*VP2);
dp= d1p//d2p//d3p//d1p//d2p//d3p;
varrp = (RP**2)*dp`*C*dp; serp = sqrt(varrp);
finish correl;
call correl(C, CovG, VG1, VG2, CovP, VP1, VP2, RG, RP, SERG, SERP);
print "Genotypic Correlation Between &trait1 and &trait2";
print RG serg;
print "Phenotypic Correlation Between &trait1 and &trait2";
print RP serp;
quit; run;
%mend;

%corr(GY,TW);
%corr(GY,SKW);

```

```

%corr(GY,SS);
%corr(GY,SN);
%corr(GY,HT);
%corr(GY,HS);
%corr(TW,SKW);
%corr(TW,SS);
%corr(TW,SN);
%corr(TW,HT);
%corr(TW,HS);
%corr(SKW,SS);
%corr(SKW,SN);
%corr(SKW,HT);
%corr(SKW,HS);
%corr(SS,SN);
%corr(SS,HT);
%corr(SS,HS);
%corr(SN,HT);
%corr(SN,HS);
%corr(HT,HS);

```

Run;

Appendix V:

Title1 'testpathsas';

options nodate;

Data test;

Input Name	Loc\$	GY	SKW	SpikeNo	Seeds_Spike;
------------	-------	----	-----	---------	--------------

cards;

;

%inc 'pathsas.sas';

%pathsas (data=test,

indep=skw SpikeNo Seeds_Spike,

dep0=GY,

bylist=loc,

printreg=no,

printout=yes,

corrind=yes,

corrdep=yes,

boot=yes,

random=1234501,

samples=200

);

run;

The startup.sas file used for PATHSAS analysis.

```
/* This program was pasted into SAS Program editor and has used a data file,
pathsas.sas, and jackboot.sas files stored in a specified directory for analysis to be
conducted */
```

```
Title1 'testpathsas';
```

```
options nodate;
```

```
Data test;
```

```
    infile 'orig.data';
```

```
    input name $ loc gy kw tp seed_t;
```

```
    run;
```

```
%inc 'pathsas.sas';
```

```
%inc 'jackboot.sas';
```

```
%pathsas    (data=test,
             indep=kw tp seed_t,
             dep0=gy,
             bylist=loc,
             printreg=no,
             printout=yes,
             corrind=yes,
             corrdep=yes,
             boot=yes,
             random=4578091,
             samples=1000);
run;
```

A-9 The pathsas.sas macro used for PATHSAS analysis.

```
/* This file was included in a specified directory that startup.sas was able to recognize */
```

```
%macro analyze(data=, out=);
```

```
data data1; set &data;
```

```
proc standard data=data1 mean=0 std=1 out=_sdata2;
```

```
    by &bylist; var &indep &dep0 &dep;
```

```
proc reg data=_sdata2 noprint
```

```
    outest=_estdep(drop=_model_ _type_ _rmse_ intercept);
```

```
    by &bylist; model &dep0=&indep;
```

```
%if &dep ne %then %do;
```

```
proc reg data=_sdata2 noprint
```

```
    outest=estindep(drop=_model_ _type_ _rmse_ intercept);
```

```
    by &bylist;
```

```
    model &dep=&dep0;
```

```
data _estind2; set estindep end=eof;
```

```
    by &bylist;
```

```
    array _r regc1-regc&nodep;
```

```
    retain regc1-regc&nodep;
```

```
    * if first.&bylast then _i_=0; _i_+1; _r=&dep0;
```

```

    if eof then output; drop &dep0 &dep _depvar_; run; %end;
proc corr data=data1 outp=_corr noprint;
  by &bylist; var &indep; run;
data _corr; set _corr;
  if _type_='CORR';
  drop _type_; run;
data _estdep; set _estdep;
  array _reg &indep;
  array _r2 reg1-reg&noind;
  do over _reg; _r2=_reg; end;
  drop &indep; run;
data _tog;
  if _n_=1 then set _estdep; set _corr; by &bylist;
  array _dir &indep;
  array _corr &indep;
  array _r2 reg1-reg&noind;
  _n+1; &dep0=0; do over _dir;
    if _n=_i then _dir=_r2; else _dir=_r2*corr; &dep0 + _dir; end;
  drop _n;
  * keep &bylist--_name_ &indep &dep0 _depvar_;
  drop reg1-reg&noind ; format &indep &dep0 5.2; run;
data _tog2; set _tog;
  * drop &indep; drop _depvar_;
%if &dep ne %then %do;
data _tog2;
  if _n_=1 then set _estind2;
  set _tog; by &bylist;
  array _r regc1-regc&nodep;
  array _t &dep;
  do over _r; _t=&dep0 * _r; end;
  format &dep &dep0 5.2;
  format regc1-regc&nodep 5.2;
  drop regc1-regc&nodep;
  * drop &indep; drop _depvar_; run; %end;
data &out; set _tog2;
  rename _name_= indep; run;
%mend analyze;

%macro pathsas(data,indep,dep0,dep,bylist,printreg,printout,corrind,
  corrdep,boot,random=1234501,samples=1000);

%local word printr;
%global noind nodep noby bylast;
%let noind=0; %if &indep ne %then %do;

```

```

%let word=%scan(&indep,1); %do %while (&word ne );
%let noind=%eval(&noind+1);
%let word=%scan(&indep,&noind+1);%end;%end;

%let nodep=0;
%if &dep ne %then %do;
%let word=%scan(&dep,1);
%do %while (&word ne );
%let nodep=%eval(&nodep+1);
%let word=%scan(&dep,&nodep+1);
%end;
%end;
%let noby=0;
%if &bylist ne %then %do;
%let word=%scan(&bylist,1);
%do %while (&word ne );
%let noby=%eval(&noby+1);
%let by&noby=%scan(&bylist,&noby);
%let word=%scan(&bylist,&noby+1); %end;
%let bylast=%scan(&bylist,&noby); %end;
%if %upcase(&printreg)=YES %then %let printr=;
%else %let printr=noprint;
%if &bylist eq %then %do;
%let bylist=_dummy; %let noby=%eval(1);
%let by&noby=%scan(&bylist,&noby);
%let bylast=%scan(&bylist,&noby); %end;
data _data1; set &data;
%if &bylist eq _dummy %then _dummy=1;;
keep &bylist &dep0 &dep &indep; run;
proc sort data=_data1;
by &bylist;
proc standard data=_data1 mean=0 std=1 out=_sdata2;
by &bylist;
var &indep &dep0 &dep; run;
proc reg data=_sdata2 &printr
outsscp=_sscp(keep=&bylist intercept _type_)
outest=_estdep(drop=_model_ _type_ _rmse_ intercept);
by &bylist;
model &dep0=&indep; run;
data _sscp; set _sscp;
if _type_='N';
rename intercept=nobs;
drop _type_;
data _estdep; merge _sscp _estdep;

```



```

    by &bylist; array _v &indep; _look='no ';
    if nobs<=&noind then do; _look='yes'; do over _v; _v=.; end; end;run;
proc print data=_estdep;
    where _look='yes'; var &bylist nobs;
title3 'The following identification levels do not have enough obs. for
analysis';
title4 ' and the regression coefficients were set to missing    ';run;
title3 ' ';
title3 'Correlation coefficients for Independent variables';
%if %upcase(&corrind)=YES %then %do;
    %if &bylist eq _dummy %then
        %str(proc print data=_corr(drop=&bylist); format &indep 5.2;
            run;);
    %else %str(proc print data=_corr; format &indep 5.2; run;);
%end;
%if %upcase(&corrdep)=YES and &nodep>0 %then %do;
    title3 'Correlation coefficients for dependent variables';
    proc corr data=_data1 outp=_corrdep noprint;
        by &bylist; var &dep0 &dep;
        data _corrdep; set _corrdep;
        if _type_='CORR'; drop _type_;
        %if &bylist eq _dummy %then %str(
proc print data=_corrdep(drop=&bylist);
format &dep0 &dep 5.2; run;);
%else %str( proc print data=_corrdep; format &dep0 &dep 5.2; run;);
    title3 ' ';%end;
data _estdep; set _estdep;
    array _reg &indep; array _r2 reg1-reg&noind;
    do over _reg; _r2=_reg; end; drop &indep; run;
data _tog;
    merge _corr _estdep; by &bylist;
    array _dir &indep;
    array _corr &indep;
    array _r2 reg1-reg&noind;
    if first.&bylast then do; _totc=0; _n=0; end;
    _n+1; &dep0=.; do over _dir; if _n=_i_ then _dir= _r2; else _dir= _r2*_corr;
&dep0 + _dir; end; drop _n; keep &bylist--_name_ &indep &dep0 _depvar_ nobs;
    format &indep &dep0 5.2; run;
data _tog2; set _tog;
    drop _depvar_;

title3 'Direct Effects, Indirect Effects and Total Correlations';
%if %upcase(&printout)=YES %then %do;
%if &bylist eq _dummy %then %str(proc print data=_tog2(drop=&bylist);run;);

```

```

    %else %str(proc print data=_tog2; run;); %end;
title3 ' ';
%if %upcase(&boot)=YES %then %do;
  * %inc 'jackboot.sas';
  proc freq data=_data1;
  tables %do i=1 %to &noby; &&by&i
    %if &i lt &noby %then *; %end;
  / noprint out=_levels; run;
data _null_;
  if 0 then set _levels nobs=total;
  call symput('nlevel',left(put(total,8.)));
  stop; run;
data _out; delete; run;
  %do i=1 %to &nlevel; title3 "&i";
  data _one; set _levels;
  if _n_=&i;
  drop count percent; run;
  data _sub;
  merge _data1 _one(in=yes);
  by &bylist; if yes;
  %boot(data=_sub, samples=&samples,id=indep, chart=0,
    print=0, random=&random,stat=&dep0 &dep);
  %bootci(bc, id=indep, print=0 , stat=&dep0 &dep); run;
  data _ci;
  set bootci;
  data _ci;
  if _n_=1 then set _one;
set _ci(keep=indep name value alcl aucl confid method n); method=scan(method,2);
if not(alcl<=value<=aucl) then check='*'; else check=' ';
if (alcl<-1) or (aucl>1) then check='*'; data _out; set _out _ci; run; %end;
  title3 'Bootstrap 95% confidence intervals - using BC method';
  title4 "Random Seed= &random";
  title5 "Number of Resamples=&samples";
  proc print data=_out label split='*';
%if &bylist eq _dummy %then %str(var indep name alcl value aucl );
  %else %str(var &bylist indep name alcl value aucl );
label indep='Independent*Variables';

  format alcl aucl 6.2; run;
  title3 ' '; %end;
proc datasets library=work memtype=data;
  delete
  _CI_CORR_CORRDEP_DATA1_ESTDEP_ESTINDEP_ESTIND2_LEVELS
  _ONE

```

```

_SDATA2 _SSCP
  _SUB _TOG _TOG2; run;quit;
%mend pathsas;

```

Example of jackboot.sas macro used for bootstrap analysis.

```

/* This file was included in a specified directory that startup.sas was able to recognize */
%macro boot(data=,samples=200,residual=,equation size=,balanced
random=0,stat=_numeric_,id=,biascorr=1,alpha=.05, print=1,chart=1);
  %if %bquote(&data)= %then %do;
    %put ERROR in BOOT: The DATA= argument must be specified.;
    %goto exit; %end; %global _bootdat; %let _bootdat=&data;
  %local by useby; %let useby=0; %global usevardf vardef; %let usevardf=0;
  *** compute the actual values of the statistics;
  %let vardef=DF; %let by=; %analyze(data=&data,out=_ACTUAL_);
  %if &syserr>4 %then %goto exit;
  *** compute plug-in estimates;
  %if &usevardf %then %do;
    %let vardef=N;
    %analyze(data=&data,out=_PLUGIN_);
    %let vardef=DF;
    %if &syserr>4 %then %goto exit;%end;
  %if &useby=0 %then %let balanced=0;
  %if %bquote(&size)^= %then %do;
    %if %bquote(&balanced)= %then %let balanced=0;
    %else %if &balanced %then %do;
      %put %cmpres(ERROR in BOOT: The SIZE= argument may not be used
        with BALANCED=1.);%goto exit;%end;
    %if %bquote(&residual)^= %then %do;
      %put %cmpres;%goto exit;%end;%end;
    %else %if %bquote(&balanced)= %then %let balanced=1;
  *** find number of observations in the input data set;
  %global _nobs; data _null_; call symput('_nobs',trim(left(put(_nobs,12.))));
  if 0 then set &data nobs=_nobs; stop; run;
  %if &syserr>4 %then %goto exit;
  %if &balanced %then
%bootbal(data=&data,samples=&samples,random=&random,print=0);
  %else %if &useby %then
%bootby(data=&data,samples=&samples,random=&random,size=&size,print=0);
  %if &syserr>4 %then %goto exit;%if &balanced | &useby %then %do;
    %let by=_sample_; %analyze(data=BOOTDATA,out=BOOTDIST);
  %end; %else
%bootslow(data=&data,samples=&samples,random=&random,size=&size);
  %if &syserr>4 %then %goto exit;%if &chart %then %do;
    %if %bquote(&id)^= %then %do;

```

```

proc sort data=BOOTDIST; by &id; run;
proc chart data=BOOTDIST(drop=_sample_);
  vbar &stat;by &id;run; %end; %else %do;
proc chart data=BOOTDIST(drop=_sample_);vbar &stat; run;%end;%end;
%bootse(stat=&stat,id=&id,alpha=&alpha,biascorr=&biascorr,print=&print)
%exit:;%mend boot;%macro bootbal(data=&_bootdat,samples=200,random=0,print=0,);
data BOOTDATA/view=BOOTDATA;
%bootin; drop _a _cbig _ii _j _jbig _k _s; array _c(&_nobs) _temporary_;
array _p(&_nobs) _temporary_;do _j=1 to &_nobs;_c(_j)=&samples;end;
do _j=1 to &_nobs; _p(_j)=_j;end;_k=&_nobs; _jbig=_k;
_cbig=&samples;do _sample_=1 to &samples;do _i=1 to &_nobs;do until(_s<=_c(_j));
_j=ceil(ranuni(&random)*_k); _s=ceil(ranuni(&random)*_cbig);end;
_l=_p(_j);_obs=_l;_c(_j)+-1;
* put _sample_=_i=_k=_l= @30 %do i=1 %to &_nobs; _c(&i) %end;;
if _j=_jbig then do;
_a=floor((&samples-_sample_ -_k)/_k); if _cbig-_c(_j)>_a then do;
do _ii=1 to _k; if _c(_ii)>_c(_jbig) then _jbig=_ii;end; _
cbig=_c(_jbig); end; end;
if _c(_j)=0 then do; if _jbig=_k then _jbig=_j; _p(_j)=_p(_k); _c(_j)=_c(_k); _k+-1; end;
%bootout(_l);end;end;stop;run;%if &syserr>4 %then %goto exit;
%if &print %then %do; proc print data=BOOTDATA; id _sample_ _obs_; run;
%end;%exit:;
%mend bootbal;
%macro bootby(data=&_bootdat,samples=200,random=0,size=,print=0);
%if %bquote(&size)= %then %let size=&_nobs;
data BOOTDATA/view=BOOTDATA;
%bootin; do _sample_=1 to &samples; do _i=1 to &size;
_p=ceil(ranuni(&random)*&_nobs); _obs=_p; %bootout(_p); end; end; stop; run;
%if &syserr>4 %then %goto exit;
%if &print %then %do; proc print data=BOOTDATA; id _sample_ _obs_; run;
%end;%exit:; %mend bootby;
%macro bootslow(data=&_bootdat,samples=20,random=0,size=);
%put %cmpres; %if %bquote(&size)= %then %let size=&_nobs;
data BOOTDIST; set _ACTUAL_; _sample_=0; delete; run; options nonotes;
%local sample; %do sample=1 %to &samples; %put Bootstrap sample &sample;
data _TMPD_; %bootin; do _i=1 to &size;
_p=ceil(ranuni(%eval(&random+&sample))*&_nobs);
%bootout(_p); end; stop; run;
%if &syserr>4 %then %goto exit; %analyze(data=_TMPD_,out=_TMPS_);
%if &syserr>4 %then %goto exit; data _TMPS_; set _TMPS_; _sample_=&sample;
run;
%if &syserr>4 %then %goto exit; proc append data=_TMPS_ base=BOOTDIST;
run;
%if &syserr>4 %then %goto exit; %end; %exit:; options notes;

```

```

%mend bootslow;
%macro bootci(method, stat=,student=,id=,alpha=.05,print=1);% global _bootdat;
  %if %bquote(&_bootdat)= %then %do; %put ERROR in BOOTCI: You must run
BOOT before BOOTCI; %goto exit;%end;
  data _null_; length method $10; method=upcase(symget('method'));
  if method=' ' then do; put 'ERROR in BOOTCI: You must specify one of the
methods '
'PCTL, HYBRID, T, BC or BCa'; abort; end;
  else if method='PERCENTILE' then method='PCTL';
  else if method not in ('PCTL' 'HYBRID' 'BC' 'BCA' 'T')
then do; put "ERROR in BOOTCI: Unrecognized method '" method "'"; abort; end;
  call symput('qmethod',method); run;
  %if &syserr>4 %then %goto exit; %if &qmethod=T %then %do;
  %if %bquote(&stat)= | %bquote(&student)= %then %do;
data _null_; put 'ERROR: VAR= and STUDENT= must be specified with the T method';
run;
  %goto exit; %end; %end;
  %if %bquote(&id)^= %then %do; proc sort data=BOOTDIST; by &id _sample_; run;
  %if &syserr>4 %then %goto exit; %end;
  proc transpose data=BOOTDIST prefix=col out=BOOTTRAN(rename=(coll=value
_name_=name));
  %if %bquote(&stat)^= %then %do; var &stat; %end;
  by %if %bquote(&id)^= %then &id; _sample_; run;
  %if &syserr>4 %then %goto exit; %if &qmethod=T %then %do;
proc transpose data=BOOTDIST prefix=col
out=BOOTSTUD(rename=(coll=student _name_=studname)); var &student;
  by %if %bquote(&id)^= %then &id; _sample_; run;
  %if &syserr>4 %then %goto exit;
  data BOOTTRAN; merge BOOTTRAN BOOTSTUD;
  label student='Value of Studentizing Statistic'
  studname='Name of Studentizing Statistic'; run;
  %if &syserr>4 %then %goto exit; %end;
proc sort data=BOOTTRAN;
  by %if %bquote(&id)^= %then &id; name
  %if &qmethod=BC | &qmethod=BCA %then value;
  %else %if &qmethod=T %then _sample_;run;
  %if &syserr>4 %then %goto exit; %if &qmethod=T %then %do;
  proc transpose data=_ACTUAL_ out=_ACTTR_ prefix=value;
%if %bquote(&stat)^= %then %do; var &stat; %end;
  %if %bquote(&id)^= %then %do; by &id; %end; run;
  %if &syserr>4 %then %goto exit;
proc transpose data=_ACTUAL_ prefix=col
out=_ACTSTUD(rename=( _name_=studname coll=student)); var &student;
  %if %bquote(&id)^= %then %do; by &id; %end; run;

```

```

%if &syserr>4 %then %goto exit;
data _ACT_T_; merge _ACTTR_ _ACTSTUD;
  label student='Value of Studentizing Statistic'
  studname='Name of Studentizing Statistic'; run;
%if &syserr>4 %then %goto exit;
proc sort data=_ACT_T_;
  by %if %bquote(&id)^= %then &id; _name_ ; run;
%if &syserr>4 %then %goto exit;
data BOOTTRAN;
  merge BOOTTRAN _ACT_T_(rename=( _name_ =name));
  by %if %bquote(&id)^= %then &id; name; value=(value-value1)/student;
run; %if &syserr>4 %then %goto exit; %end;
%if &qmethod=BC | &qmethod=BCA %then %do;
%if &qmethod=BCA %then %do;
  %global _jackdat;
  %if %bquote(&_jackdat)^=%bquote(&_bootdat) %then %do;
    %jack(data=&_bootdat,stat=&stat,id=&id,alpha=&alpha,
      chart=0,print=&print); %if &syserr>4 %then %goto exit; %end;
  proc means data=JACKDIST noprint vardef=df;
    %if %bquote(&stat)^= %then %do; var &stat;%end;
    output out=JACKSKEW(drop=_type_ _freq_ _sample_) skewness=;
    %if %bquote(&id)^= %then %do; by &id; %end; run;
  %if &syserr>4 %then %goto exit;
  proc transpose data=JACKSKEW prefix=col
    out=_ACCEL_(rename=(col1=skewness _name_ =name));
    %if %bquote(&stat)^= %then %do; var &stat; %end;
    %if %bquote(&id)^= %then %do; by &id; %end; run;
  %if &syserr>4 %then %goto exit;
  proc sort data=_ACCEL_;
    by %if %bquote(&id)^= %then &id; name ; run;
  %if &syserr>4 %then %goto exit;
%end;
data _BC_; retain _alpha_conf; drop value value1; if _n_=1 then do;
_alpha=&alpha;
_conf=100*(1-_alpha); call symput('conf',trim(left(put(_conf,best8))))); end;
merge _ACTTR_(rename=( _name_ =name)) BOOTTRAN;
by %if %bquote(&id)^= %then &id; name; if first.name then do; n=0; _z0=0; end;
n+1; _z0+(value<value1)+.5*(value=value1);
if last.name then do; _z0=probit(_z0/n); output; end; run;
%if &syserr>4 %then %goto exit;
data BOOTPCTL;
retain _i_lo _up _nplo _jlo _glo _npup _jup _gup alcl aucl;
drop _alpha _sample_ _conf _i _nplo _jlo _glo _npup _jup _gup value;
merge BOOTTRAN _BC_ %if &qmethod=BCA %then _ACCEL_;;

```

```

by %if %bquote(&id)^= %then &id; name;
label _lo='Lower Percentile Point'
   _up='Upper Percentile Point'
   _z0='Bias Correction (Z0)';
if first.name then do;%if &qmethod=BC %then %do;
   _lo=probnorm(_z0+(_z0+probit(_alpha/2)));
   _up=probnorm(_z0+(_z0+probit(1-_alpha/2))); %end;
%else %if &qmethod=BCA %then %do;
drop skewness; retain _accel; label _accel='Acceleration';
   _accel=skewness/(-6*sqrt(&nobs))*(&nobs-2)/&nobs/sqrt((&nobs-1)/&nobs);
   _i=_z0+probit(_alpha/2); _lo=probnorm(_z0+_i/(1-_i*_accel)); _i=_z0+probit(1-
   _alpha/2);
   _up=probnorm(_z0+_i/(1-_i*_accel)); %end;
   _nplo=min(n-.5,max(.5,fuzz(n*_lo))); _jlo=floor(_nplo); _glo=_nplo-_jlo;
   _npup=min(n-.5,max(.5,fuzz(n*_up))); _jup=floor(_npup); _gup=_npup-_jup; _i=0; end;
   _i+1; if _glo then do; if _i=_jlo+1 then alcl=value; end;
else do; if _i=_jlo then alcl=value; else if _i=_jlo+1 then alcl=(alcl+value)/2; end;
   if _gup then do; if _i=_jup+1 then aucl=value; end;
else do; if _i=_jup then aucl=value; else if _i=_jup+1 then aucl=(aucl+value)/2; end;
   if last.name then do; output; end; run;
%if &syserr>4 %then %goto exit;%end;
%else %do; %local conf pctlpts pctlpre pctlname; %let pctlpre=a; %let pctlname=lcl ucl;
   data _null_; _alpha=&alpha; _conf=100*(1-_alpha);
   call symput('conf',trim(left(put(_conf,best8))));
   %if &qmethod=PCTL %then %do; _lo=_alpha/2; _up=1-_lo;
   %end; %else %if &qmethod=HYBRID | &qmethod=T %then %do;
   _up=_alpha/2; _lo=1-_up; %end; _lo=100*_lo; _up=100*_up;
   call symput('pctlpts',trim(left(put(_lo,best8))))||' ||
   trim(left(put(_up,best8))));run; %if &syserr>4 %then %goto exit;
proc univariate data=BOOTTRAN noprint pctldef=5;
   var value; output out=BOOTPCTL n=n
pctlpts=&pctlpts pctlpre=&pctlpre pctlname=&pctlname;
   by %if %bquote(&id)^= %then &id; name; run;%if &syserr>4 %then %goto
exit;%end;
   data BOOTCI; retain &id name value alcl aucl confid method n;
   merge %if &qmethod=T %then _ACT_T(rename=(name=value));
   %else _ACTTR(rename=(name=value));
   BOOTPCTL; by %if %bquote(&id)^= %then &id; name; %if
&qmethod=HYBRID %then %do;
   aucl=2*value-aucl; alcl=2*value-alcl;%end;
%else %if &qmethod=T %then %do;
   aucl=value-aucl*student;alcl=value-alcl*student;%end;
confid=&conf; length method $20; method='Bootstrap' ||symget('method');
label name ='Name'

```

```
value ='Observed Statistic'  
alcl ='Approximate Lower Confidence Limit'  
aucl ='Approximate Upper Confidence Limit'  
confid='Confidence Level (%)'  
method='Method for Confidence Interval'  
n    ='Number of Resamples';run;  
%if &syserr>4 %then %goto exit; %if &print %then %do; proc print data=BOOTCI  
label;  
id %if %bquote(&id)^= %then &id; name; run; %end;
```