WHEAT TRAITS VARIATIONS, ASSOCIATIONS, AND POTENTIAL IMPROVEMENT FROM CROSSES OF ELITE X NON-ADAPTED GERMPLASM

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Wheat Traits Variations, Associations, and Potential Improvement

From Crosses of Elite x Non-adapted Germplasm

By

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ABSTRACT

Mantovani, Eder Eduardo; M.S.; Department of Plant Sciences; College of Agriculture, Food Systems, and Natural Resources; North Dakota State University; August 2011. Wheat Traits Variations, Associations, and Potential Improvement from Crosses of Elite x Non-adapted Germplasm. Major Professor: Dr. Mohamed Mergoum.

Wheat improvement most often has been accompanied by a narrowing germplasm base, as newer cultivars have been derived from intercrosses between elite germplasm. However, there is a concern that narrow germplasm may restrict breeding improvement for important traits such as resistances to new biotic and abiotic stresses. In addition to germplasm base, the wheat kernel is a major component of wheat grain yield and an important factor for milling characteristics. Focusing on wheat kernel characteristics might be a key element to improve wheat genotypes for agronomic and quality traits. With the intention to broaden the wheat germplasm, and to explore the associations between kernel traits and agronomic as well as quality traits, a two-year study was initiated in 2009 to examine the influence of the kernel traits on the agronomic and quality attributes of a 160 Recombinant Inbred Lines (RIL) population developed from an adapted (ND 705) and a non-adapted genotype (PI 414566). The experiment was conducted at Prosper and Carrington, North Dakota, during 2009 and 2010. The RIL population had a better performance at Carrington than Prosper due to favorable climatic conditions at this location, in 2009 and 2010. The results in this study showed that kernel traits had a high correlation among them and they exhibited continuous variations suggesting a polygenic inheritance. Grain yield, kernel volume weight (KVW), and flour extraction were highly correlated with kernel width, length/width ratio, weight, and area. Eight RIL yielded better than the adapted parent ND 705 and two of the RIL along with three checks were

significantly superior for grain yield compared with the other genotypes across all environments. Although the non-adapted parent has a facultative grown habit, several RIL required fewer days to flower compared to the adapted parent. Two RIL had better flour extraction compared to the other genotypes included in this study. These results indicated that kernel traits can play a significant role in improving agronomic and quality traits. Higher values for grain yield, KVW, and flour extraction were significantly associated with spheroid or round shape (short and plump), large, and heavy kernels. The high agronomic and quality attributes showed by some RIL demonstrated that the use of a non-adapted parent can broaden the genetic variability while increasing the genetic gain for certain traits. Also, breeders should pay attention to kernel size and shape during the parental selection for the development of populations with improved agronomic and quality traits.

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"Happiness is not a destination but a state of mind"

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PREFACE

This thesis consists of two papers that will be submitted for publication in the appropriate scientific journals. The 'General Introduction' and 'Literature Review' provide a general review of the importance of this study, some previous research, and how the two papers are related to the main issues: wheat agronomic and quality trait variations in a population originated from a cross between an adapted x non-adapted genotype and the potential uses of kernel characteristics in breeding programs in order to develop new cultivars with higher grain yield and quality attributes. Subsequently each paper includes an abstract, introduction, material and methods, results and discussion, conclusion, references section, and appendix. The references for the 'General Introduction' and 'Literature Review' are cited in the 'General References' section.

GENERAL INTRODUCTION

Wheat (*Triticum aestivum* L.) is one of the most important crops worldwide. In 2010, wheat harvest reached 222.2 million ha with a total production of 645.4 million tons. The average yield was 2.9 tons per ha. The USA represented 9.3 % of world wheat production with 60 million tons. The USA is consistently the world's biggest wheat exporter (28.2 % of the world exports) with 59 % of its production exported. Wheat ranks third among USA field crops planted acreage, behind corn and soybeans. (USDA-ERS, 2011).

Wheat in the USA entered the 21st century dealing with many challenges, despite a strong domestic market for wheat products. This is due to at least two reasons. First, wheat returns are falling compared to other crops, stemming in large part from foreign competition. Second, the government programs provide more support to alternative crops. Consequently, wheat harvested acreage has decreased nearly 12 million ha (35 %) compared to its highest acreages reached in 1981, and USA wheat export share percentage has also significantly declined in the past two decades (USDA-ERS, 2011).

Wheat, along with rice, is the main source of energy for humans. The world population keeps increasing, the production of wheat should follow the same pattern. Wheat production can be increased by expanding planted areas, improving the agronomic techniques, and/or developing improved varieties with higher yield potential. Expanding planted acreage is a limiting factor in many regions, and there are many environmental factors affecting crops, such as climatic conditions. Therefore, the development of new adapted cultivars coupled with improved agronomic practices remains the main factor to increase yield (Briggle and Curtis, 1987).

The wheat consumers are also becoming more exigent for wheat quality. Consequently, scientists are expanding more effort to improve wheat quality characteristics along with grain yield. Today, several traits such as grain protein content and kernel volume weight are being used routinely as markers for indirect selection for quality.

Wheat improvement has been in most cases accompanied by a narrowing germplasm base, as newer cultivars have been derived from intercrosses between elite germplasm. Therefore, wheat breeding programs are continuously improving and releasing varieties with higher agronomic and quality attributes with the use of narrow germplasm base. However, there is a concern that the narrow germplasm may restrict breeding flexibility and slow response to new challenges such as pests, pathogens, and agronomic practices in the future. Also, the tendency of the continuous use of the same germplasm base will at a certain point of time result in an even more restrict germplasm base. Due to these challenges, Breeders are constantly faced with broadening the genetic diversity using elite x non-adapted crosses.

Wheat kernel is the main component of wheat grain yield and quality. In addition, traits such as spikes per unit area, kernels per spike, and kernel weight are major contributing components in determining yield and quality of wheat. Many recent studies (Campbell et al., 1999; Breseghello and Sorrells, 2007; Sun et al., 2009) indicated that wheat kernel characteristics are important quality factors, particularly for milling characteristics. The focus on wheat kernel characteristics in the selection process might be a key element for the wheat quality improvement, therefore, increasing the possibility of achieving higher levels of success in the breeding programs.

The development of high performance cultivars for North Dakota with improved grain yield and milling attributes will raise the value of wheat, which will contribute to the increase of wheat growers and industry incomes. Similarly, high quality of wheat will allow the USA growers to be competitive in the international wheat export market.

OBJECTIVES

The objectives of this study were:

- Explore the potential improvement of wheat agronomic and quality traits from crosses involving elite/adapted and introduced/non-adapted germplasm in wheat breeding programs.
- Study the agronomic and quality trait variations and their correlations in a Recombinant Inbred Lines (RIL) population developed from an elite x non-adapted parental cross.
- Study the kernel characteristics variations using a RIL population derived from an elite x non-adapted parents.
- Determine the associations between kernel characteristics and major agronomic and quality traits, and explore their potential uses in breeding programs in order to improve these traits.

LITERATURE REVIEW

Broadening the germplasm base with the use of exotic lines

Cereal crop species have been the focus of numerous genetic studies and crop improvement efforts due to the global agronomic importance of these species. The lack of genetic diversity, the problems of genetic vulnerability, and selection plateaus has been recognized (Gouesnard et al., 1996). The utilization of exotic germplasm is difficult due to its non-adaptability. However, it has been the most frequently identified procedure to broaden the germplasm base (Holley and Goodman, 1988). There are several reports on the transfer of genes from non-adapted wild relatives into cultivated hexaploid wheat (Miedaner et al., 2006; Nelson and Goodman, 2008); however this approach has not been widely used in wheat breeding programs which in most of the cases use crosses between elite x elite lines to release new cultivars. The reason for this is that favorable attributes are often linked to undesirable traits in non-adapted wild relatives and it is difficult to separate them. These linked groups are called linkage drag (Stalker 1980; Patterson et al. 1991).

A complete integration of exotic and elite chromosomes is required to avoid the loss of useful linked factors. Several generations of random mating, with or without mild selection, are recommended before the application of more rigorous selection (Eagles and Lothrop, 1994). The optimal proportion between exotic and elite germplasm to form the base population is also an important theme. This proportion is influenced by the divergence between the parents, the level of dominance of the considerable traits, the use of back-crosses to the elite material, and the short- and long-term goals. The exotic genes from the non-adapted parent should broaden the germplasm base, improve the considerable traits,

and be incorporated in the new released varieties. However, these goals must be achieved without negatively affecting the desirable traits already presented in the population.

Currently, many breeding programs have been using non-adapted parent to incorporate resistance to Fusarium head blight caused by *Fusarium graminearum* (Miedaner et al., 2006) and other diseases. However, the domesticated crops are narrowing their germplasm base (Zamir, 2001) decreasing their ability to adapt to emerging biotic and abiotic stresses and increasing potential for widespread crop failure (Horsfall et al., 1972). The exotic germplasm can be used as a source of genetic variation lost during the domestication. These non-adapted genotypes, including wild species and landraces, often carry agronomically undesirable alleles. However, genetic studies can identify the valuable genes from the exotic species, and integrate these traits into released varieties (Zamir, 2001).

Kernel characteristics

In recent years, scientists (Dholakia et al., 2003; Breseghello and Sorrells, 2007; Sun et al., 2009) have been studying traits to be used as markers for indirect selection for yield as well as quality. The major components of wheat grain yield are productive spikes per unit area, kernels per spike, and kernel weight. Other traits that can affect kernel number are tillers per plant, spikelets number per spike, and number of viable florets per spikelet (Ma et al., 2007). Since kernel is the main component of wheat grain yield, researchers are interested in studying the properties of kernel characteristics and their genetics. Similarly, they are trying to determine the relationship between kernel traits and wheat quality and agronomic traits. An example is the association between kernel size and seedling vigor. Larger seeds could have a positive association with seedling vigor and,

consequently, increased grain yield. Recently, Breseghello and Sorrells (2007) found that kernel size and shape can contribute to the improvement of grain yield and quality of the final product. In addition, similar results were found by Gaines et al. (1997), showing that size, shape, and consistency of kernels is essential to flour yield, an important trait for the milling industry. The importance of choosing the right variety with the most desirable characteristics for quality attributes has been reinforced by Souza et al. (2004), which found that the genotype selection is more crucial for high quality wheat than environment conditions and nitrogen levels.

Most of the biological characteristics for important kernel traits, such as wheat kernel size, are complex and believed to be genetically quantitative (Campbell et al., 1999; Sun et al., 2009). In addition, wheat kernel characteristics are frequently influenced by environment, having high genotype x environment interaction (Sun et al., 2009). Dholakia et al. (2003) using a population of 106 F_7 recombinant inbred lines (RIL), generated from a cross between the two hexaploid wheat cultivars PH 132 and WL 711, concluded that the kernel length, weight, width and the factor form density (FFD) parameter described by Giura and Saulescu (1996) have polygenic nature. The FFD was expressed as: FFD = kernel weight / (kernel length x kernel width). Their conclusion was based on the continuous variation obtained for these traits. They also observed transgressive segregants for all kernel traits.

Different characteristics such as kernel volume weight or test weight, grain density, kernel sizing, near-infrared reflectance, hardness score among others have been tested for wheat physical quality. Wheat kernel weight and size are important quality factors for evaluation of milling characteristics (Ohm et al., 1998). Kernel volume weight is

considered by semolina millers to be a desirable physical trait in wheat, along with kernel size, vitreousness, and cleanliness (Troccoli and Di Fonzo, 1999).

Kernel characteristics, texture, kernel volume weight, shape of germ, crease, and brush are characteristics that can influence milling quality, which affects the market value of wheat. For bread wheat, kernel shape and uniformity can significantly affect milling quality (Campbell et al., 1999). Gaines et al. (1997) reviewed the effect of kernel size on milling quality in wheat. They found considerable diversity in kernel size among the soft wheat cultivars based on their average kernel size and on the range of kernel size distribution. This is evidenced by some large kernel varieties being twice as large as small kernel varieties. They also reported that there is variation in kernel diameter and weight. The pressure on the development of modern cultivars with superior grain yield can result in wheat cultivars with smaller average kernel size and a wider size range. Gaines et al. (1997) believed that this may happen because modern varieties are released to produce smaller tertiary kernels between two rows of full-size kernels. However, tertiary kernels do not fill out completely during the grain filling period because they have less time to develop, thus they can be shriveled or smaller.

Millers realized that milling a mixture of small and large kernels on a roller mill reduces the flour yield because the break roll gap setting cannot be optimized for either the small or the large kernels (Gaines et al., 1997). Preferably, the break roll spacing in this situation should be set for the average kernel size. Since size mixtures are normal and there is a tendency to associate small kernels with shriveled grains, small kernels are frequently considered to have less flour yield potential and therefore are considered to have lower milling properties. Cleaning (aspiration or sieving) grains to remove shriveled kernels,

usually results in increased homogeneity. However, this cleaning will result in economic loss in quantity because small yet sound and good quality kernels are also discarded. Millers should pay attention to small kernels if they wish to have a high quality mill. A possible solution to improve milling wheat with different kernel size could be, first, to classify and subdivide kernels according to their size, then mill kernels with different kernel sizes separately (Gaines et al., 1997).

Different approaches have been used to study the shape and size of wheat kernels. Marshall et al. (1984), using geometrical models based on theoretical analyses, classified this technique as being precise. The conclusion was drawn because both shape and kernel size can significantly affect the volume per unit surface area of the wheat kernel, consequently milling yield and proportion of flour per unit weight of kernel. They found that the best shape to increase milling yield is a spheroid shape, which unfortunately is negatively correlated to grain yield. However, the same authors added that the geometrical model is not yet realistic, and that further studies should be conducted. For breeding purposes, changes in kernel volume weight have a greater impact on milling yield than changes in kernel shape. Furthermore, kernel volume appears to be easier to manipulate in plant breeding programs than kernel shape. However, breeders should take into consideration the impact of selection for kernel characteristics on yield and quality (Marshall et al., 1984).

Kernel volume weight has been shown to be an important trait for growers and millers (Marshall et al., 1984; Gaines et al., 1997; Campbell et al., 1999; Breseghello and Sorrells, 2007; Sun et al., 2009). It is highly correlated to kernel characteristics and has a higher heritability compared to grain yield. Kernel volume weight is utilized in breeding

programs in order to discard lines in early generations (Patil et al., 1994). In addition, kernel volume weight has demonstrated a high positive correlation with flour quantity. Also, it has been used as an indicator of overall grain quality and it is utilized to establish the price in several countries (Sun et al., 2009).

Kernel characteristics evaluation methodologies

Efficient, precise, realistic, and simple techniques to measure the kernel traits should be used with the intention to understand better the kernel characteristics and how they could generate gains on agronomic traits such as grain yield or quality traits such as kernel volume weight or flour extraction.

Currently, the techniques used are:

- Theoretical geometric model – it is a geometrical model based on theoretical analysis. In this method, there is no use of kernels, thus it was classified by the authors (Marshall et al., 1984) as unrealistic, although, precise. It was used to predict the best shape and size of wheat kernel to enhance flour extraction.
- Kernel size distribution – kernels are separated into three classes; large, medium, and small. Usually, the distribution is done by a wheat sizer equipped with three sieve openings. Top sieve (Tyler #7 with 2.92 mm opening), middle sieve (Tyler #9 with 2.24 mm opening), and bottom sieve (Tyler #12 with 1.65 mm opening). This technique has been used for many years. However, there are no specific measurements of kernel dimension. Several researchers have used this technique (Gaines et., 1997; Ohm et al., 1998; Tsilo et al., 2010).
- Manual – researchers have evaluated kernel length and width using a ruler with precision of 0.1mm (Sun et al., 2009). Another possibility to measure kernel

length and width is the use of Vernier Calipers (Dholakia et al., 2003). This technique can take a long time to evaluate the samples and can have a low precision especially when low amount of kernels are evaluated (usually between 20 and 30 kernels).

- Single kernel characterization system (SKCS) – the SKCS 4100 (Perten Instruments, Springfield, IL) instrument was created at the USDA Grain Marketing and Production Research Center in Manhattan, KS (Osborne and Anderssen, 2003). The SKCS was mainly developed for kernel hardness measurement. However, it also provides kernel weight and width but not kernel length. These features have been added lately as well (Ohm et al., 1998; Lee et al., 2006; and Tsilo et al., 2010).
- Digital image analysis - basically, this evaluation technique consists of three steps. First, a photograph of the kernel sample is taken. Second, this photograph is processed to be able to run in the next program (in some systems the second step is skipped). Third, the processed photograph is analyzed by a program that provides the kernel measurements. Usually, it can measure the kernel length, width, and area. However, it can require a long time to complete all the procedures. There are few studies using digital image analysis technology to measure kernel traits (Campbell et al., 1999; Bergman et al., 2000; Breseghello and Sorrels, 2006; and Breseghello and Sorrels, 2007).

Currently, the available techniques to measure kernel dimensions lack at least one of the principles to be an ideal model. This model has to be efficient, precise, realistic,

simple, and provide all kernel dimensions. Consequently, future efforts should take place on the development of new techniques that incorporate all requirements for an ideal model.

Genetics of kernel characteristics

Dholakia et al. (2003) in their molecular studies on kernel length, weight, width and FFD found that nine markers were associated with kernel length. These markers consisted of five Microsatellite (MS), three Random Amplification of Polymorphic DNA (RAPD), and one Inter Simple Sequence Repeat (ISSR). Two markers were localized on chromosome 2BL, two on chromosome 2DL, one on chromosome 5BL, one on chromosome 6BS, and one on chromosome 7BL. The localization of the remaining two markers was not determined. For kernel weight, seven markers were found: three ISSR, two RAPD, and two MS. Three markers were located on chromosome 2BL, two on chromosome 2DL, and two markers were not localized. In the same study, only two markers were found to be linked to kernel width. One of them was located on chromosome 2DL, and the other was not localized. Six markers were associated with the FFD parameter: four MS, one ISSR, and one RAPD. Two markers were located on chromosome 2DL, one on 5BL, one on 6BS, and the other two markers were not localized.

The same study (Dholakia et al., 2003) indicated that some markers were correlated with several traits. For instance, the marker located on chromosome 2DL for kernel width was also linked to kernel length, weight and FFD. Similarly, for FFD markers, one was associated with all other traits, two markers were associated with kernel length and weight, and two associated with kernel length. Based on the association among these loci, it was proposed that they may have pleiotropic effects on kernel characteristics, or they are tightly linked loci.

Other molecular studies have been conducted to investigate the genetics of kernel characteristics. Breseghello and Sorrells (2007), among others, located several loci on chromosomes 2D, 5A, and 5B as having major effects on kernel size and shape in wheat. These researchers referred to measurement of kernel size using traits such as kernel projection area, cross-section area, surface, volume, and weight. Kernel shape was measured by these researchers using characteristics such as kernel length, width, horizontal axes proportion, sphericity and vertical perimeter.

Correlation among wheat characteristics

Sun et al. (2009) conducted a study using a set of 131 RIL derived from a cross between two Chinese lines (Chuan 35050 x Shannong 483) and they found positive correlation between kernel length and kernel width, and kernel weight (0.216, and 0.424, respectively). Kernel weight and kernel width, and kernel volume weight were also correlated (0.753 and 0.194, respectively). A negative correlation was detected between kernel volume weight and kernel length (-0.222). Kernel volume weight and kernel width had no significant correlation.

In another study conducted by Ohm and his co-researchers (1998), flour yield was significantly correlated with kernel hardness, volume weight, and density (0.50, 0.50, and 0.36, respectively). Large kernel had no significant correlation with flour yield. Contrary to other findings (Troccoli and Di Fonzo, 1999), Ohm et al. (1998) also reported significant positive correlation between kernel volume weight and kernel weight (0.47). However, the positive correlation among these characteristics can be due to environmental effects (Ghaderi and Everson, 1971). Slaughter et al. (1992), Satumbaga et al. (1995) and Ohm et al. (1998) found negative correlations between protein content and wheat kernel size and

weight. According to Morris et al (1946) wheat with heavier and thicker kernels could have a higher portion of endosperm which contains lower amounts of protein content than the aleurone layer.

Therefore, many aspects related to kernel characteristics remain unclear. This study aimed to examine the kernel characteristics, their associations, and explore the potential use of genetically diverse and non-adapted germplasm, to improve agronomic and quality traits in breeding programs, in order to develop new cultivars with high yield and quality attributes, and increase the germplasm base.

PAPER 1. WHEAT AGRONOMIC AND QUALITY TRAITS VARIATIONS, CORRELATIONS, AND POTENTIAL IMPROVEMENT FROM CROSSES OF ELITE X NON-ADAPTED GERMPLASM

ABSTRACT

Wheat improvement has been in most of the cases accompanied by a narrowing germplasm base, as newer cultivars have been derived from intercrosses between elite germplasm. However, there is a concern that narrow germplasm may restrict breeding improvement for important traits such as resistances to new biotic and abiotic stresses in the future. With the intention to broaden the wheat germplasm, a two-year study was initiated in 2009 to examine the agronomic and quality attributes of 160 Recombinant Inbred Lines (RIL) developed from an adapted line (ND 705) and a non-adapted genotype (PI 414566). The experiment was conducted at Prosper and Carrington, North Dakota, during two years. The RIL population had a better performance at Carrington due to favorable climatic conditions at this location, in 2009 and 2010. Eight RIL yielded better than the adapted parent ND 705 and two of the RIL along with three checks were significantly superior for grain yield compared to the other genotypes across all environments. Although the non-adapted parent has a facultative grown habit, several RIL required fewer days to flower compared to the adapted parent. Two RIL had better flour extraction compared to the other genotypes included in this study. This RIL population had also many lines with poor agronomic and quality performance. However, the high agronomic and quality attributes showed by some RIL demonstrated that the use of a non-adapted parent can broaden the genetic variability while increasing the genetic gain for some traits.

INTRODUCTION

Wheat (*Triticum aestivum* L.) is one of the most important staple foods in the world. In 2010, wheat had a global production of 645.4 million tons. The USA is the largest wheat exporter with 28 % of the world exports. However, the wheat production in the USA has decreased. China and India are the largest wheat producers in the world. The Russian wheat production increased drastically in the last years exceeding the USA production which is the fourth largest wheat producer in the world (USDA-ERS, 2011).

Wheat in the USA has entered the 21st century dealing with many challenges, despite a strong domestic market for wheat products. This happened for many reasons including the following. First, wheat returns decreased compared with other crops, stemming in large part from foreign competition. Second, the government programs provide more support to alternative crops. Consequently, wheat acreage harvested has decreased nearly by 12 million ha (35 %) of its highest acreages reached in 1981. The USA wheat export share percentage has also declined in the past two decades (USDA-ERS, 2011).

Along with rice, wheat is the main source of energy for human beings. Wheat is an inexpensive source of carbohydrates, fiber, protein and vitamins. Wheat is used as raw material in making breads, cakes, cookies, pastries, crackers, and in the manufacturing of pasta products such as macaroni and spaghetti. Breeders aim to increase wheat grain yield and quality attributes while keeping it easy to access and inexpensive. Researchers in recent studies (Souza et al., 2004; Otteson et al., 2007; Otteson et al., 2008) tested different factors such as genotype, environment conditions, nitrogen level, and timing, and seeding rate that could influence grain yield and its quality aspects. They found that genotype

selection was the most important factor in determining grain yield and quality, including milling performance.

Alexander et al. (1984) studied the effects of yield component selection on wheat grain yield in three populations. They found that selection for kernel weight has usually enhanced grain yield. They also indicated that using kernel weight could be a desirable trait in early generation selection because this trait can generally be measured with more precision at relatively low cost. However, direct selection for grain yield was usually more effective showing that an optimum balance between the number of spikes per unit area, kernels per spike, and kernel weight may possibly result in high yielding genotypes. Baril (1992) working with 34 winter wheat genotypes also found that kernel weight was the most important yield component.

Bread wheat quality is a key attribute for wheat milling and baking industry exigencies and broad consumer preferences. Bread wheat has to meet requirements of domestic and export markets. Therefore, end-use quality has been one of the major targets of various wheat breeding programs around the world, especially in Australia, Canada, and USA. Wheat consumers and baking industries are becoming more exigent for wheat quality attributes. Breeders usually focusing on grain yield and resistance/tolerance for abiotic and/or biotic stresses are currently placing more emphasis on quality characteristics (Marshall et al., 1984; Nelson et al., 2006; Kerfal et al., 2010). Some of the indicators for milling quality include kernel volume weight, 1000-kernel weight, and flour extraction yield (Finney et al., 1987). Currently, several quality traits such as grain protein content and kernel volume weight are being used routinely as markers for indirect selection for quality (Nelson et al., 2006).

Milling quality is a function of the proportions of flour extracted and the flour contamination by non-endospermic tissues (brain and germ) of the wheat kernel (Marshall et al., 1984). However, desirable quality attributes vary according to the final use of the wheat flour. Suitable bread production requires cultivars with higher protein concentration, alkaline water retention capacity, and dough strength whereas for cakes and cookies these traits are undesirable (Nelson et al., 2006).

Hard red spring wheat (HRSW) grown in the Northern Great Plains of North America is a premium class of wheat used in end-use purposes requiring high gluten strength. Flour mills worldwide blend HRSW with other wheat classes with lower protein to enhance the gluten strength. These flour blends formed by different concentrations of HRSW are used to classify bread products and Chinese-type noodles. In addition, products such as whole grain breads, pizza crusts, yeast breads, hard rolls, and bagels, produced with HRSW display superior appearance and taste (Underdahl et al., 2008a).

Wheat improvement has been in most of the cases accompanied by a narrowing germplasm base, as newer cultivars have been derived from intercrosses of existing elite germplasm. Wheat breeding programs are continuously improving and releasing varieties with higher agronomic and quality attributes with the use of narrow germplasm base. However, there is a concern that narrow germplasm may restrict breeding flexibility and slow response to new biotic and abiotic stresses. Also, the tendency of the continuous use of the same germplasm base will at a certain point of time result in an even more restrict germplasm base. Due to these restrictions, many wheat breeding programs are broadening the genetic diversity using elite x exotic crosses (Holley and Goodman, 1988). The utilization of exotic germplasm is usually undesirable by breeders due to its non-

adaptability. The reason for this is that favorable attributes are often linked to undesirable traits in non-adapted germplasm and it is difficult to dissociate them. These linked groups are called linkage drag (Stalker 1980; Patterson et al. 1991). A complete integration of exotic and elite chromosomes is required to avoid the loss of useful linked factors. Several generations of random mating, with or without mild selection, are recommended before the application of more rigorous selection (Eagles and Lothrop, 1994).

The exotic germplasm can be used as a source of genetic variation lost during the domestication and selection. These non-adapted genotypes, including wild species and landraces, often carry undesirable alleles. However, genetic studies can identify the agronomic valuable genes of the exotic species, and integrate these traits into released varieties (Zamir, 2001) increasing the genetic variability thereby preventing widespread crop failure.

Therefore, many aspects related to elite x non-adapted (exotic) crosses remain unclear requiring further investigations. The objectives of this study were to explore the possibility to improve the agronomic and quality traits with the use of a cross between elite x non-adapted germplasm and explore the potential use of exotic germplasm in breeding programs, in order to develop new superior germplasm.

MATERIAL AND METHODS

Material

This study was conducted using a population of 160 RIL. This population was developed from a cross between ND 705 and PI 414566. The ND 705 is a HRSW line developed by the HRSW breeding program at North Dakota State University (NDSU). The PI 414566 is originally from China and it was obtained from the USDA National Small Grains Collection. It is described as a facultative type of wheat which requires cold temperature to flower, tall, brown kernels, and susceptible for diseases such as stripe rust caused by *Puccinia striiformis*. The RIL population was advanced through single seed descent method to F_8 generation and it was obtained from the Wheat Germplasm Enhancement program at NDSU. A combination of field nurseries at New Zealand, North Dakota, and greenhouse were used to advance this population. The selection of the parents used to develop this RIL population was based on the genetic divergence between the parents in order to produce a RIL population with significant polymorphism for agronomic and quality traits. The number of RIL used in this study was determined by the amount of seeds available for each RIL.

The checks used in this study were: 'Alsen' (PI 615543) (Frohberg et al., 2006), 'Steele-ND' (PI 634981) (Mergoum et al., 2005), 'Glenn' (PI 639273) (Mergoum et al., 2006), 'Faller' (PI 648350) (Mergoum et al., 2008), 'Barlow' (PI 658018) (Mergoum et al., 2011), 'Briggs' (PI 632970) (Devkota et al., 2007), and 'Alpine'. All the check cultivars are HRSW varieties except Alpine which is a hard white spring wheat variety.

Field experiment

The RIL, their parents (ND 705 and PI 414566), and checks (Alsen, Steele-ND, Glenn, Faller, Barlow, Briggs, and Alpine) were evaluated in the summers of 2009 and 2010 at two field locations in ND; Carrington Research Extension Center near Carrington and Prosper Research Plots near Prosper. These sites represent two diverse environments. Carrington is located in the Central ND region with an altitude of 484 m. The soil type at Carrington is Heimdal-Emrick series (loamy, mixed, superactive, Calcic/Hapludolls). Prosper is located in the Red River Valley region (Eastern ND) with an altitude of 274 m. The soil type at Prosper is Bearden series (fine silty, mixed, superactive, frigid Aeric Calciaquolls) (USDA-NRCS, 2011).

In 2009, the planting dates were May 7 at Carrington, and May 28 at Prosper. The harvesting dates were August 26 at Carrington for all the lines except the parent PI 414566 which was harvested on October 6. At Prosper all the lines were harvested on September 18 except PI 414566 which was harvested on October 13. The winter of 2008-2009 had a high amount of snow causing field flood delaying the planting date. Consequently, PI 414566 as a facultative type requiring a longer period to complete its cycle did not have enough cold temperature to completely flowering. In 2010, the planting dates were April 21 at Carrington, and May 20 at Prosper. The harvesting dates were August 18 and 27 at Carrington and Prosper, respectively. PI 414566 was vernalized for three weeks prior to planting at Prosper. However, the emergence rate of the PI 414566 vernalized seeds was very low because of the poor and heavy soil characteristics at Prosper in 2010. Carrington was planted earlier; consequently, PI 414566 had enough cold temperature to flower,

therefore completing its cycle successfully. Sowing rates was 113 kg ha^{-1} in all environments.

In 2009, the experiments included the $F_{8:9}$ RIL, their parents, and checks. In fall 2009, the RIL were sent to New Zealand winter nursery for seed increase. Consequently, in 2010, the experiments included the $F_{10:11}$ RIL, their parents, and checks. The entries were arranged in a 13 x 13 partially balanced square lattice design, with two replicates. The experimental units consisted of seven rows, 2.44 m long and 12.7 cm apart.

Data collection

The grains from harvested plots were cleaned by using a clipper grain cleaner before measuring the following traits.

Agronomic traits

- Grain yield (kg ha^{-1}) – determined by weighing the cleaned grain harvested from each plot.
- Yield components:
  - ✓ Number of spikes (spikes m^{-2}) – determined by counting the number of spikes in two 61 cm long rows per plot. Counts were then converted to determine total number of spikes per square meter.
  - ✓ Number of kernels (kernels $spike^{-1}$) – calculated by averaging the kernels from ten individual spikes selected at random from each plot. The kernels were counted by the electronic seed counter (Seedburo Equipment Co., Chicago, IL) after threshing the spikes.
  - ✓ 1000-kernel weight (g) determined by 'Digital Image Analysis' (DIA) – calculated based on the number of kernels within 10 g samples from each

plot used in the DIA. The results were converted and the weight of 1000 kernels was determined (Doehlert et al., 2004).

- Plant height (cm) – measured from the soil surface to the top of the spike, excluding the awns at maturity. An average plant height was recorded from each plot.
- Days to heading (days) – recorded when inflorescences fully emerged in at least 50% of the spikes and was calculated as the number of days from planting to heading.
- Lodging (%) – estimated by the percent of lodged plants per plot when lodging occurred at a given environment.
- Maturity (days) – recorded when kernels fully complete the physiologic maturity in at least 50 % of the spikes. It was calculated as the number of days from planting to maturity.
- Emergence (%) – estimated by the percent of fully emerged plants per plot.
- Spike color (1-5) – visually rated by using a scale having equidistant values from 1 (white) to 5 (dark red).

Quality traits

- Grain protein content (%) – measured according to American Association of Cereal Chemists (AACC) standard method 46-30 (AACC International, 2000), using an Infratec 1226 Cold Grain Analyzer expressed on a 12 percent basis.
- Kernel volume weight (kg m^{-3}) – measured according to AACC standard method 55-10 (AACC International, 2000).

- Kernel hardness (0-90+) – measured using the single kernel characterization system (SKCS) 4100 instrument (Perten Instruments, Springfield, IL). This instrument measured the force needed to crush the kernels which is expressed as a hardness index. Table 1 has the kernel hardness category classification according to the hardness index.

Table 1. Kernel hardness category classification according to the hardness index (HI)

Category	HI [a]
Extra hard	90 +
Very hard	81 – 90
Hard	65 – 80
Medium hard	45 – 64
Medium soft	35 – 44
Soft	25 – 34
Very soft	10 – 24
Extra soft	Up to 10

[a] HI=hardness index

- Flour extraction (%) – 150 g cleaned wheat samples were tempered to 16.0 % moisture for 16 hr before being milled on a Brabender Quadrumat Junior Mill according to standard procedure used at the Department of Cereal and Food Sciences at North Dakota State University. The flour extraction percentage was calculated by dividing the flour weight of straight grade flour over the total grain weight milled (150 g).

Data analysis

Data were subjected to the analyses of variance (ANOVA), using the MIXED procedure of the Statistical Analyses System (SAS Institute, 2004). Within the mixed model, the RIL, their parents, and checks were considered fixed effects. Environments and blocks were considered random effects. The ANOVA was conducted within and across

environments. Environments were considered homogeneous when the ratio of the effective error variances for each trait was less than 10-fold (Appendix Table 1) (Hartley, 1950; Patterson and Silvey, 1980; Geng et al., 1982; Mitchell, 1995). Data were subjected to combined analysis if environments were homogeneous. Tables 2 and 3 show the pertinent sources of variation, degrees of freedom, and the appropriate expected mean squares.

Table 2. Single analyses of variance for a lattice design, with treatment as fixed effects, and rep as random effects

SOV	DF [a]	MEAN SQUARE		F-test
		Obs	Expected	
Rep	$r - 1$	M1		non-valid
Blocks within rep	$r(k - 1)$	M2		non-valid
Treatment	$(t - 1)$	M3	$\sigma^2 + r\Sigma\alpha_i^2 / (t-1)$	M3 / M4
Error	$(k -1)(rk - k - 1)$	M4	σ^2	
Total	$rk^2 - 1$			

[a] = The letters k, t, and r refer to the size of the lattice, the number of treatments, and the number of replications, respectively.

Table 3. Combined analyses of variance for a lattice design, across 2 years and 2 locations, with treatment as fixed effects, and rep and environment (year x location) as random effects

SOV	DF [a]	MEAN SQUARE		F-test
		Obs	Expected	
Environment	$e - 1$	M1		non-valid
Treatment	$t - 1$	M2	$\sigma^2 + r\sigma^2_{et} + re\Sigma\alpha_i^2/(t-1)$	M2 / M5
Rep within Env	$e(r - 1)$	M3		non-valid
Block within Rep x Env	$er(k - 1)$	M4		non-valid
Env x Treat	$(e - 1)(t - 1)$	M5	$\sigma^2 + r\sigma^2_{et}$	M5 / M6
Error	$e(k -1)(rk - k - 1)$	M6	σ^2	
Total	$ert - 1$			

[a] = The letters k, t, e, and r refer to the size of the lattice, the number of treatments, the number of environments, and the number of replications per environment, respectively.

Mean separation test was performed using an F-protected least significant difference (LSD) value at $P \leq 0.05$ level of significance for each trait. Correlations between the different agronomic and quality traits were calculated for single

environments using the CORR procedure of SAS (2004). Pooled correlations were determined in case of homogeneous correlations. The homogeneity test of the correlations was determined using the χ^2.

RESULTS AND DISCUSSION

Climatic conditions

The 2009 growing season at Carrington and Prosper was characterized by lower rainfall and colder temperatures compared to 2010 growing season and the long term average. Prosper 2010 had the highest amount of rainfall and air temperature in all four environments (Table 4). Prosper, however suffered with a strong hail storm and wind in both years decreasing significantly wheat grain yield and quality attributes especially for the RIL population due to lodging. Climatic conditions at Carrington were in general favorable for spring wheat in both 2009 and 2010. Carrington in 2010 was the earliest planted experiment (21 April) in all environments. The cold weather during the emergence and early wheat growing stages allowed the facultative parent PI 414566 to flower and complete its growing cycle.

Wheat worldwide is classified into two distinct groups according to the vernalization requirements: winter and spring types. Generally, winter wheat requires cold temperature or vernalization period to be able to flower while spring wheat is normally insensitive to cold temperature. However, there are intermediate wheat types which require cold temperatures classified in between winter and spring types. These intermediates types of wheat are called semi-winter, alternative, or facultative. PI 414566 turns out to be one of these wheat types. According to Stelmakh (1998), they are usually characterized by strong photosensitivity and partial sensitivity to vernalization. Facultative wheats have in general less cold tolerance, a shorter but distinct period required for vernalization, and flower earlier compared to true winter wheats (Stelmakh, 1998). Based on 2009 results (low productivity of PI 414566) the seeds of the parent PI 414566 were germinated and

vernalized prior to planting at Prosper in 2010. Unfortunately, seedling survival rate after transplanting was very low (18%) due to the heavy soil (Bearden series) at Prosper.

Table 4. Total monthly rainfall and total season rainfall (mm) and average air temperature (°C) at Carrington, and Prosper, ND during the 2009 and 2010 growing season

Month	Carrington			Prosper		
	2009	2010	Avg.[a]	2009	2010	Avg.[a]
	Rainfall					
	mm					
April	42.7	36.3	36.6	26.4	29.5	36.3
May	34.5	66.0	63.2	23.1	69.9	67.8
June	40.9	81.8	96.3	66.5	80.8	91.4
July	38.1	49.5	79.0	24.6	103.4	82.3
August	50.5	41.7	63.0	57.4	89.4	68.1
September	55.1	107.4	46.7	53.1	134.6	54.1
Total	261.9	382.8	384.8	251.2	507.5	400.1
	Average air temperature					
	°C					
April	3	9	6	4	10	6
May	11	12	14	12	14	13
June	16	17	18	17	19	18
July	18	20	21	18	21	21
August	18	21	20	18	21	20
September	17	13	14	17	13	14

[a] Total rainfall and air temperature of the long term averages measured since 1990 (Source: NDAWN, 2011).

Data analysis

The ANOVA for each environment was performed. The error variance was homogeneous across all environments allowing a combined analysis across the four environments for all the traits except lodging. The homogeneity test was performed based on the 10-fold concept (Appendix 1). The ANOVA summaries for the agronomic and quality traits are reported in Table 5, while the means of the experiment, parents, RIL, and checks for the combined and single environments are presented in Table 6. The ANOVA (Table 5) shows that the genotypes variances were significant for all the agronomic and quality traits. The environments also had a significant effect on all traits.

Table 5. Source of variation, degrees of freedom, mean square, and level of significant of the agronomic and quality traits of the RIL, their parents, and checks grown at Carrington and Prosper, ND, during 2009 and 2010

Source	df [a]	GY [b]	df [a]	NS [b]	df [a]	KH [b]
Env	3	127917748**	3	2563379**	3	3450.4*
Treatmnt	168	1574369**	168	26102**	168	220.9**
Env*Treat	504	430667**	504	10028*	504	22.9**
Error	571	127547	572	8262	572	4.7
CV (%)		11.7		15.9		3.1

Source	df [a]	TKW [b]	NK [b]	KVW [b]	GPC [b]	FE [b]	df [a]	PH [b]	DH [b]
Env	3	3920.3**	10684**	483247**	99.2**	3932.3**	3	4496.2**	11813**
Treatmnt	168	52.7**	69.1**	3632.1**	5.0**	70.2**	168	438.5**	96.1**
Env*Treat	504	7.5**	31.1**	1005.3**	0.8**	13.7**	504	41.0**	12.5**
Error	573	2.2	13.0	116.2	0.3	4.0	574	15.1	1.1
CV (%)		5.2	10.6	1.4	3.5	3.3		4.3	1.9

Source	df [a]	Mat [b]	df [a]	SC [b]	Df [a]	Eme [b c]
Env	2	18442**	1	131.4**	---	---
Treatmnt	168	72.8**	168	2.5**	168	76.4**
Env*Treat	336	17.5**	168	1.0**	---	---
Error	430	3.6	288	0.3	144	2.4
CV (%)		2.1		16.4		1.5

[a] df=degrees of freedom.

[b] GY=grain yield, NS=number of spikes, KH=kernel hardness, TKW =thousand kernel weight, NK=number of kernels, KVW=kernel volume weight, GPC=grain protein content, FE=flour extraction, PH=plant height, DH=days to heading, Mat=maturity, SC=spike color, and Eme=emergence.

[c] ANOVA for one location (Prosper 2010).

* and ** Significant at the 0.05 and 0.01 probability levels, respectively.

Table 6. Experiment, parents (ND 705 and PI 414566), and RIL means, their ranges (min and max), and checks means for agronomic and quality traits at Carrington and Prosper, ND in 2009 and 2010

Traits [a]	Env	Exp	Parents		RIL			Checks	$LSD_{0.05}$
		Mean	ND 705	PI 414566	Mean	Min	Max	mean	
GY ($kg\ ha^{-1}$)	All	3042	3581	615	3002	1835	4112	4236	645
	Carr 09	3617	4021	568	3590	2586	4626	4622	572
	Carr 10	3405	3924	1373	3363	2014	4719	4568	699
	Pros 09	2919	3399	34	2888	1241	4681	3968	898
	Pros 10	2225	2851	195	2163	87	3885	3834	610
NS (spikes m^{-2})	All	572	652	285	569	449	709	682	98
	Carr 09	447	474	145	444	319	594	549	121
	Carr 10	614	717	529	609	468	891	716	172
	Pros 09	645	642	294	642	303	1000	769	221
	Pros 10	584	755	74	581	379	856	689	190
NK (Kernels $spike^{-1}$)	All	34	30	27	34	27	42	32	5.5
	Carr 09	40	34	24	40	28	52	39	5.9
	Carr 10	37	31	31	37	21	47	34	5.8
	Pros 09	31	29	15	31	19	43	28	9.1
	Pros 10	28	26	36	28	4	40	29	7.2
TKW (g)	All	28.4	30.1	16.0	28.4	22.4	35.1	31.2	2.7
	Carr 09	31.5	32.4	17.8	31.5	24.3	38.5	33.4	1.9
	Carr 10	30.0	31.4	22.1	29.9	23.2	36.7	32.8	3.0
	Pros 09	28.6	30.4	11.6	28.6	22.5	36.5	30.4	3.9
	Pros 10	23.6	25.7	11.4	23.5	10.5	37.6	28.1	2.5
PH (cm)	All	91	97	71	91	64	107	86	6.3
	Carr 09	87	97	67	87	61	111	82	7.5
	Carr 10	94	102	88	94	66	110	90	6.3
	Pros 09	94	92	59	95	63	114	87	9.1
	Pros 10	89	95	70	89	67	109	86	7.6
DH (days)	All	56	55	76	55	49	65	55	3.5
	Carr 09	56	57	82	56	51	62	57	1.8
	Carr 10	63	65	74	63	59	69	64	1.7
	Pros 09	52	51	81	52	43	65	50	2.5
	Pros 10	50	47	68	51	41	69	47	2.1
Lod (%)	All	-	-	-	-	-	-	-	-
	Carr 09	0	0	0	0	0	0	0	0
	Carr 10	1	1	1	1	0	38	0	7
	Pros 09	28	25	0	28	0	97	44	53
	Pros 10	49	7	62	50	0	100	9	29
Mat (days)	All	92	91	105	92	83	103	91	4.7
	Carr 09	-	-	-	-	-	-	-	-
	Carr 10	100	103	109	100	91	104	100	3.4
	Pros 09	89	86	113	88	79	106	88	5.1
	Pros 10	86	83	95	86	78	98	85	2.3

Table 6. (Continued)

Traits [a]	Env	Exp	Parents		RIL			Checks	$LSD_{0.05}$
		Mean	ND 705	PI 414566	Mean	Min	Max	mean	
	All	-	-	-	-	-	-	-	-
Eme	Carr 09	-	-	-	-	-	-	-	-
(%)	Carr 10	-	-	-	-	-	-	-	-
	Pros 09	-	-	-	-	-	-	-	-
	Pros 10	99	100	18	100	90	100	100	3.0
	All	3.1	1.5	4.5	3.1	1.4	4.5	2.0	1.4
SC	Carr 09	-	-	-	-	-	-	-	-
(1-5)	Carr 10	3.5	1.5	5.0	3.6	1.0	5.0	2.1	-
	Pros 09	-	-	-	-	-	-	-	1.0
	Pros 10	2.6	1.5	4.1	2.6	1.9	4.1	1.9	1.0
	All	14.9	15.6	15.1	14.9	13.2	17.7	15.3	0.9
GPC	Carr 09	14.8	15.0	17.5	14.7	12.5	17.6	15.4	0.6
(%)	Carr 10	15.1	17.3	13.7	15.0	12.8	18.5	15.5	0.7
	Pros 09	14.3	14.4	14.3	14.3	11.5	16.9	14.4	1.7
	Pros 10	15.6	15.9	15.2	15.6	13.5	17.8	15.6	0.7
	All	749	791	598	749	669	790	774	31
KVW	Carr 09	787	820	619	787	723	816	798	10
($kg\ m^{-3}$)	Carr 10	761	796	673	761	708	802	777	13
	Pros 09	751	796	577	751	685	786	764	24
	Pros 10	697	751	522	695	486	782	759	31
	All	70	76	63	70	57	86	77	4.7
KH	Carr 09	74	75	49	74	59	88	79	3.3
(HI)	Carr 10	67	73	62	67	42	84	74	4.0
	Pros 09	69	78	74	68	45	87	75	5.5
	Pros 10	72	81	72	71	47	89	79	4.1
	All	61	66	54	61	46	67	63	3.6
FE	Carr 09	63	68	48	64	46	70	63	2.3
(%)	Carr 10	64	67	60	64	46	71	65	3.1
	Pros 09	59	65	41	59	32	66	61	4.3
	Pros 10	57	64	65	57	38	70	62	5.4

[a] GY=grain yield, NS=number of spikes, NK=number of kernels, TKW=thousand kernel weight, PH=plant height, DH=days to heading, Lod=lodging, Mat=maturity, Eme=emergence, SC=spike color, GPC=grain protein content, KVW=kernel volume weight, KH=kernel hardness, FE=flour extraction.
- Traits were not collected at these locations.

The treatment x environment interactions were significant for all agronomic and quality traits as well. At Carrington, the genotypes performed overall much better for most agronomic and quality trait as compared to Prosper. Within the locations, the genotypes had a better performance in 2009 compared to 2010. The parents, ND 705 and PI 414566, showed drastic differences for all the agronomic and quality traits with ND 705 being

superior for all traits (Table 6). This confirmed the decision to develop a RIL population with large diversity, thereby broadening the genetic range due to the use of an elite x exotic type of cross.

Agronomic traits

Grain yield

Considerable segregation was observed for the agronomic traits including grain yield for the RIL population. Grain yield followed relatively a normal distribution (Figure 1), suggesting polygenic inheritance. The mean grain yield overall environments was 3042 kg ha^{-1}. While the RIL population mean for yield across environments was 3002 kg ha^{-1}, ND 705 and PI 414566 parental means were 3581 and 615 kg ha^{-1}, respectively. The grain yield mean for checks across environments was 4236 kg ha^{-1} (Table 6). The checks Faller, Barlow, and Steele-ND, and the RIL 143 and 88 were significantly superior compared to the other genotypes across environments. Faller was the only genotype that had significantly superior yield in all the environments while Barlow and Steele-ND were not significantly superior at Prosper in 2010. The RIL 143 and 88 were among the group that had significantly higher grain yield at two of the four environments (RIL 143 at Carrington and Prosper in 2009, and RIL 88 at Prosper in 2009 and Carrington in 2010) (Table 7). In addition to diseases (mainly Fusarium head blight caused by *Fusarium graminearum* and leaf diseases) lodging and kernel shattering were other main reasons for decreased grain yield especially at Prosper in 2010. Barlow and Steele-ND also had losses in grain yield due to lodging and kernel shattering. Barlow and Steele-ND had 7 and 16 % lodging, respectively, and relatively low kernel shattering, while RIL 143 and 88 had losses in grain yield due to lodging. RIL 143 and 88 had 11 and 24 % lodging, respectively.

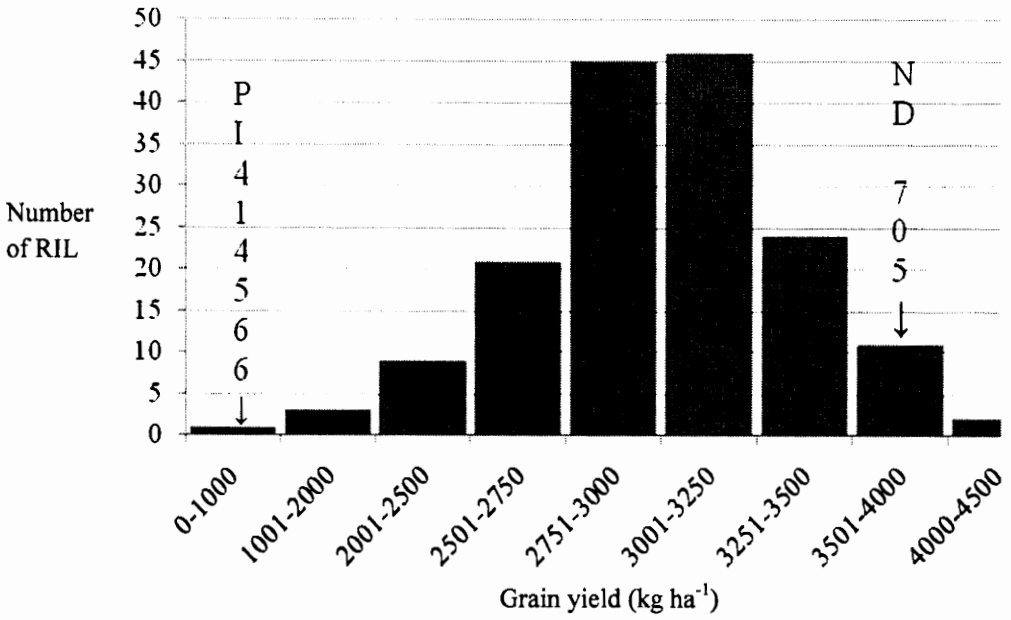

Figure 1. Frequency distribution of 160 RIL and their parents for mean grain yield across all locations (Carrington and Prosper, ND) and years (2009 and 2010). (Parental means are marked with arrows).

PI 414566 had significantly the poorest performance for grain yield in all environments. This parent was not able to complete its cycle in 2009, and had low emergence rate (18 %) and high lodging percentage (62 %) at Prosper in 2010 (Table 6). Three RIL (106, 127, and 96) had also poor performance in at least three out of the four environments (Table 7). These three RIL had also high lodging percentage (100, 80, and 88 % at Prosper in 2010; 56, 55, and 43 % at Prosper 2009; respectively). At Carrington, only RIL 106 had lodging problem (38 %) in 2010. PI 414566 was more susceptible to diseases overall than the adapted parent ND 705. Probably the RIL with low grain yield incorporated the susceptible genes for diseases from the non-adapted parent PI 414566. This could be another factor that resulted in low grain yield for these RIL.

PI 414566, a facultative type of wheat, even with adequate growing conditions at Carrington in 2010 had poor agronomic performance. ND 705, an experimental line, was

Table 7. Grain yield means (kg ha^{-1}) and least square difference (LSD) of the top and bottom ten genotypes across all environments (Carrington and Prosper in 2009 and 2010)

Top 10 genotypes

Carrington 2009		Carrington 2010		Prosper 2009		Prosper 2010		Overall env	
Treatment	GY [a]	Treatment	GY	Treatment	GY	Treatment	GY	Treatment	GY
Faller	5075	Barlow	5001	Barlow	4756	Faller	4791	Faller	4685
Steele-ND	5020	Faller	4789	RIL 143	4681	Barlow	4098	Barlow	4684
Barlow	4897	RIL 6	4719	RIL 86	4209	RIL 88	3885	Steele-ND	4328
RIL 82	4626	Steele-ND	4687	Faller	4168	Glenn	3814	RIL 143	4112
RIL 138	4587	Alpine	4671	Steele-ND	4009	Briggs	3750	RIL 88	4071
Alsen	4551	RIL 91	4621	RIL 167	3966	RIL 139	3729	Briggs	4014
RIL 143	4521	RIL 88	4532	RIL 88	3889	Steele-ND	3697	Alsen	3981
RIL 149	4480	Briggs	4504	RIL 102	3860	RIL 41	3463	Alpine	3980
Briggs	4439	RIL 78	4491	RIL 70	3841	RIL 102	3418	Glenn	3977
RIL 92	4402	RIL 46	4491	Alpine	3783	Alpine	3413	RIL 30	3724

Bottom 10 genotypes

Carrington 2009		Carrington 2010		Prosper 2009		Prosper 2010		Overall env	
Treatment	GY	Treatment	GY	Treatment	GY	Treatment	GY	Treatment	GY
RIL 106	3084	RIL 106	2438	RIL 62	2081	RIL 117	877	RIL 166	2454
RIL 163	2944	RIL 69	2433	RIL 37	2061	RIL 37	797	RIL 117	2360
RIL 105	2917	RIL 118	2400	RIL 15	1949	RIL 96	665	RIL 118	2348
[redacted]	2905	RIL 96	2346	RIL 40	1938	RIL 73	618	RIL 37	2326
RIL 66	2882	RIL 141	2330	RIL 166	1809	RIL 97	586	RIL 87	2271
RIL 142	2871	RIL 87	2220	RIL 106	1758	RIL 118	543	RIL 34	2191
RIL 69	2709	RIL 117	2206	[redacted]	1665	[redacted]	252	RIL 96	1986
RIL 61	2639	RIL 111	2120	RIL 96	1656	RIL 144	249	[redacted]	1909
RIL 141	2586	RIL 58	2014	RIL 34	1241	[redacted]	195	RIL 106	1835
[redacted]	568	[redacted]	1373	[redacted]	34	RIL 106	87	[redacted]	615
$LSD_{0.05}$	572	$LSD_{0.05}$	699	$LSD_{0.05}$	898	$LSD_{0.05}$	610	$LSD_{0.05}$	645

[a] GY=grain yield, kg ha^{-1}.

not released by the HRSW Program, although it went to the last steps of the breeding pipeline demonstrating its high agronomic performance. Most RIL had a grain yield performance ranging between the two parents or statistically equal to ND 705. However, eight RIL yielded better than the best parent indicating transgressive segregation for grain yield in this population. This demonstrates the possibility of selecting genotypes with high yield better than the best parent.

Tsilo et al. (2010) in their study used two well adapted hard red spring wheat lines, MN98550 (Bacup/McVey) and MN99394 (SD3236/SBF0402), to generate 139 RIL population. These parents diverged for water absorption and dough mixing strength. Both parents performed well and very similar for grain yield. However, they were also able to observe transgressive segregation on their RIL population for grain yield. The range for grain yield among their RIL population however, was smaller compared to the RIL used in our study. This can be due to a lack of genetic variation between the RIL parents in their study. McCartney et al. (2005) worked with a population originating from two parents yielding identically. Consequently the segregation for yield was smaller as well. The use of a non-adapted line as one of the parents crossed with an adapted line theoretically should increase the segregation of the population for several traits. However, the chance to find RIL with higher grain yield than the best parent is usually rare because of the undesirable linkages or unforvable alleles for high performance (in this case yield) that come from the non-adapted parent. However, crosses between elite x elite lines usually should generate progenies with better agronomic performance. In most of the RIL population originated from elite x non-adapted lines, back-crosses to the adapted parent are required; however, in this study the eight superior RIL showed that after several generations of recombination,

outstanding lines can be obtained from a cross between elite (ND 705) x non-adapted (PI 414566). Similarly, many RIL in this population had low grain yield.

1000-kernel weight

The three most important yield components are 1000-kernel weight, number of kernels/spike and number of spikes/m^2. The mean for 1000-kernel weight over the four environments was 28.4 g. ND 705, PI 414566, RIL, and checks means were 30.1, 16.0, 28.4, and 31.2 g, respectively. The range among RIL was from 22.4 to 35.1 g (Table 6). Among the RIL, 37 had heavier kernels compared to ND 705 indicating transgressive segregation for this trait as well. The only top yield cultivar among the group with the largest values of 1000-kernel weight was Faller. The other checks and top RIL for grain yield were among the intermediate, close to the superior group, while all the poor yielding RIL (106, 127, and 96) and PI 414566 were among the bottom group with low 1000-kernel weight. As expected, the 1000-kernel weight was positively correlated with grain yield in all environments. The correlation coefficient values for Carrington and Prosper in 2009 were homogeneous and were pooled together (r=0.44). However, non-pooled correlations for Carrington and Prosper in 2010 were 0.19, and 0.75, respectively (Table 8). Similar results were reported by Hsu and Walton (1971), and McCartney et al. (2005) who found a significant correlation between 1000-kernel weight and grain yield. However, Tsilo et al. (2010) did not find significant association between these two traits. Large genetic diversity was observed for this trait facilitating selection. However, it is well known that yield components compensate for each other. Therefore, grain yield is often a function of well balanced yield components. Selecting only for kernel weight does not result in lines with the highest grain yield.

Table 8. Correlation coefficients (r) among grain yield and yield components (1000-kernel weight, number of kernels per spike, and number of spikes per m2) and their significance for the RIL, their parents, and checks grown in four environments (Carrington 2009 and 2010, and Prosper 2009 and 2010). The homogenous r values were pooled and denoted in the table

	Environment	1000-kernel weight	Number of kernels/spike	Number of spikes/m^2
Grain yield	Carr 09	0.44** [a]	0.24** [a]	0.43** [a]
	Pros 09			
	Carr 10	0.19*		
	Pros 10	0.75**	0.57**	

* and ** Significant at the 0.05 and 0.01 probability levels, respectively.
[a] Pooled correlation coefficients across environments.

Number of kernels/spike

The overall mean for number of kernels/spike was 34. ND 705, PI 414566, and RIL means were 30, 27, and 34 kernels/spike, respectively. The range among RIL was from 27 to 42 kernels/spike. The overall mean of the checks for number of kernels/spike was 32 (Table 6). The majority of the RIL produce more kernels/spike than ND 705 indicating transgressive segregation for number of kernels/spike in this population. This demonstrates the possibility of selecting genotypes with higher number of kernels/spike. However, the yield components must be balanced to reach higher levels of grain yield. The top grain yield RIL in the group with the largest values for number of kernels/spike was RIL 30. All the checks and top grain yield RIL were grouped in the intermediate group while the poor grain yield RIL and PI 414566 were among the bottom group except for RIL 96 which was among the intermediate group for the number of kernels/spike. The number of kernels/spike was also positively correlated with grain yield in all environments. The correlation coefficient values for Carrington 2009 and 2010, and Prosper 2009 were pooled and was 0.24, while at Prosper 2010 was 0.57 (Table 8). Previous studies (Waddington et

al., 1986; Donmez et al., 2001; and Underdahl et al., 2008b) also found that increased number of kernels/spike was positively correlated with increased grain yield.

Number of spikes/m^2

Number of spikes/m^2 averaged 572 spikes overall environments. ND 705, PI 414566, RIL, and checks averaged overall environments 652, 285, 569, and 682 spikes/m^2, respectively. The range among RIL was from 449 to 709 spikes/m^2 indicating transgressive segregation for the number of spikes/m^2 in this population (Table 6). All the checks and RIL 88 and 30 were among the group with the significantly largest values of number of spikes/m^2. Line 143 is located in the intermediate group for this trait along with low yielding lines such as 127 and 34. The other low grain yield performance lines (106, 96, and 87) and PI 414566 were among the group with the lowest values for number of spikes per m^2. There was a significant correlation between number of spikes/m^2 and grain yield. Correlation coefficient values between number of spikes/m^2 and grain yield were pooled for all the environments (r=0.43) (Table 8). Our finding concurred with those by Donmez et al. (2001) who found that spike density was also positively correlated with grain yield. However, Underdahl et al. (2008b) did not find a significant association between these two traits.

Plant height

The overall mean of plant height for the genotypes included in this study was 91 cm while, this mean was 97, 71, 91, and 86 cm for ND 705, PI 414566, the RIL, and checks, respectively. Even with the large range among RIL (from 64 to 107 cm) (Table 6), no significant correlation was found between plant height and grain yield in this study (Table 9). Similarly, Underdahl et al. (2008b) and Tsilo et al. (2010) did not find significant

correlation between plant height and grain yield. However, a negative correlation between these two traits has been reported (Cox et al., 1988; Donmez et al., 2001) while another study (McCartney et al., 2005) has reported a positive correlation between plant height and grain yield. The lack of correlation between plant height and grain yield in our study may be due to the narrow genetic variability among the RIL for this trait. In this study, plant height had significant and positive correlation with days to heading at Carrington 2009 and 2010, and Prosper 2009 (r=0.30), maturity at Prosper 2009 and Carrington 2010 (r=0.42) and correlated with spike color (r=-0.26) (Table 9). The negative correlation between plant height and spike color indicates that, in general, the genotypes with white spikes are taller than the genotypes with red dark spikes.

Days to heading and maturity

The overall mean for days to heading was 56 days and the overall mean of parents ND 705 and PI 414566, RIL, and checks were 55, 76, 55, and 55 days, respectively. The facultative grown habit of PI 414566 explains the long period required by this genotype to flower. The range of RIL was from 49 to 65 days (Table 6) indicating transgressive segregation toward the early types (earlier than ND 705) in this population. No RIL with more days to heading than PI 414566 was recorded. This demonstrates the possibility of selecting genotypes with fewer days to flower than the early parent ND 705. The top three RIL for grain yield (143, 88, and 30) and the checks were among the group with intermediate number of days to heading. RIL 127, 106, and 96 needed more days to flower (65, 62, and 58 days, respectively) than the overall average indicating they may carry the same alleles for this trait as their parent PI 414566.

Table 9. Correlation coefficients (r) among the agronomic traits and their significance for the RIL, their parents, and checks grown in four environments (Carrington and Prosper in 2009 and 2010). The homogenous r values were pooled and denoted in the table

	Environment	Days to heading	Spike color	Maturity	Grain yield
Plant height	Carr 09		-	-	
	Pros 09	0.30** [a]	-	0.42** [a]	0.015 [a]
	Carr 10				
	Pros 10	-0.04	-0.26** [a]	0.08	
Days to heading	Carr 09		-	-	-0.14
	Pros 09		-	0.91**	-0.50**
	Carr 10		-0.17*	0.61**	0.20*
	Pros 10		0.35**	0.83**	-0.73**
Spike color	Carr 09			-	-
	Pros 09			-	-
	Carr 10			0.18* [a]	-0.39** [a]
	Pros 10				
Maturity	Carr 09				-
	Carr 10				0.08
	Pros 09				-0.46** [a]
	Pros 10				

* and ** Significant at the 0.05 and 0.01 probability levels, respectively.
- Traits were not collected at these locations.
[a] Pooled correlation coefficients across environments.

The overall mean of number of days to maturity was 92 while the two parents ND 705 and PI 414566, the RIL, and the checks matured in 91, 105, 92, and 91 days, respectively. The range for the RIL was from 83 to 103 days (Table 6) indicating transgressive segregation for this trait. Even with the use of the parent PI 414566, a facultative type, the RIL population had 66 lines maturing earlier than the parent ND 705. This provides the possibility of selecting genotypes with earlier maturity than ND 705. As expected, the results for maturity were very similar to the results of days to heading. The correlations between these two traits were 0.61, 0.91, and 0.83 at Carrington 2010, and Prosper 2009 and 2010, respectively. The correlations between these two traits with grain

yield were significantly negative and higher at Prosper compared to those obtained at Carrington (Table 9). This could be due to the late planting at Prosper compared to Carrington in both years. At Carrington, the genotypes had more time to complete the entire cycle and cooler temperatures during the vegetative phase, whereas at Prosper, some genotypes did not have enough time to fully complete their cycle or the temperatures were high during the vegetative phase triggering the early flowering, consequently decreasing the grain yield. This also could be the reason for the negative correlation between these two traits (days to heading and maturity) and spike color at Prosper (Table 9). The late genotypes could not have enough time to express the dark color of the spikes due to the short agriculture season at Prosper. Recent study by Donmez et al. (2001) also found a negative correlation between days to heading and grain yield. However, other studies did not find any significant correlation between number of days to heading and grain yield (Underdahl et al., 2008b; Tsilo et al., 2010) and between days to maturity and grain yield (McCartney et al., 2005).

Spike color

This trait was collected only in the 2010 environments after we observed that in 2009, lines with the red spike color tend to have low grain yield. On a scale from 1 (white) to 5 (dark red), the overall mean for spike color was 3.1 (yellow color). The RIL parents ND 705 had a white color (1.5) while PI 414566 had a red color (4.5). The RIL spike color ranged between the two parents (from 1.4 to 4.5). The overall mean of the checks for spike color was 2 (Table 6). The 2010 data showed a negative and significant correlation between spike color and grain yield (-0.39) (Table 9) confirming our observation in 2009. Zeven (1983) made a review about spike color of bread wheat and found that most results

indicate one (semi)dominant gene conditioning red spike (*Rg*) and appears to be located on chromosome 1BS. However, Khlestkina et al. (2009) found that the genes *Rg-A1* and *Rg-D1* located at chromosomes 1A and 1D, respectively, also encode for spike color. According to the results in this study, some of the spike color genes may be linked to some grain yield genes.

Quality traits

Grain protein content

The mean for grain protein content (GPC) across the four environments was 14.9 % which was also equal to the RIL population mean. ND 705 and PI 414566 parental GPC means were 15.6, and 15.1 %, respectively. The range of the RIL for GPC was from 13.2 to 17.7 %. The overall mean of the checks for GPC was 15.3 % (Table 6). Although the RIL mean for GPC was lower than both parents, several RIL had higher GPC than both parents ND 705 and PI 414566. Besides the cultivar Glenn at Carrington in 2009, the checks were not among the top 10 genotypes for GPC across environments (Table 10). Overall, the parents had similar results for GPC. However, transgressive segregation among the RIL was observed in both directions. Overall environments, the RIL that had significant superior GPC were RIL 10, 4, 40, 90, and 130. These RIL were consistent across all environments being among the 10 highest GPC genotypes in all the environments. For the low GPC group, the RIL were inconsistent. The genotypes with the lowest values of GPC overall environments were not consistent and not always included among the 10 lowest GPC genotypes (Table 10). In general, Prosper in 2010 had the highest values for GPC because it suffered from a strong hail storm and wind decreasing significantly the grain yield. These environmental conditions, unfavorable to high grain yield usually cause a shift

Table 10. Grain protein content and least square difference (LSD) of the top and bottom ten genotypes across all environments (Carrington and Prosper 2009 and 2010). Data were expressed on a 12 percent basis

Top 10 genotypes									
Carrington 2009		Carrington 2010		Prosper 2009		Prosper 2010		Overall env	
Treatment	GPC [a]	Treatment	GPC	Treatment	GPC	Treatment	GPC	Treatment	GPC
RIL 10	17.62	RIL 10	18.47	RIL 10	16.86	RIL 27	17.83	RIL 10	17.68
PI 414566	17.47	RIL 40	18.12	RIL 4	16.82	RIL 10	17.83	RIL 4	17.41
RIL 4	17.33	RIL 130	18.10	RIL 40	16.61	RIL 104	17.78	RIL 40	17.37
RIL 61	17.21	RIL 4	18.03	RIL 90	16.28	RIL 40	17.69	RIL 90	17.08
RIL 40	17.06	RIL 90	17.76	RIL 61	16.10	RIL 130	17.49	RIL 130	17.08
RIL 90	17.04	RIL 138	17.45	RIL 98	15.99	RIL 4	17.48	RIL 61	16.57
RIL 130	16.95	RIL 104	17.38	RIL 14	15.77	RIL 60	17.27	RIL 138	16.44
RIL 29	16.86	ND 705	17.32	RIL 130	15.76	RIL 90	17.23	RIL 116	16.27
Glenn	16.61	RIL 61	17.05	RIL 135	15.73	RIL 118	17.19	RIL 104	16.24
RIL 138	16.43	RIL 116	16.74	RIL 56	15.70	RIL 22	17.12	RIL 27	16.07
Bottom 10 genotypes									
Carrington 2009		Carrington 2010		Prosper 2009		Prosper 2010		Overall env	
Treatment	GPC	Treatment	GPC	Treatment	GPC	Treatment	GPC	Treatment	GPC
RIL 87	13.19	RIL 127	13.56	RIL 33	12.80	RIL 58	14.26	RIL 82	13.66
RIL 11	13.07	RIL 87	13.54	RIL 55	12.80	RIL 36	14.24	RIL 145	13.63
RIL 107	12.97	RIL 162	13.46	RIL 105	12.78	RIL 96	14.23	RIL 156	13.61
RIL 54	12.96	RIL 142	13.29	RIL 107	12.75	RIL 11	14.06	RIL 5	13.57
RIL 156	12.94	RIL 145	13.25	RIL 82	12.72	RIL 95	14.00	RIL 11	13.57
RIL 146	12.88	RIL 82	13.17	RIL 31	12.70	RIL 87	13.96	RIL 87	13.46
RIL 96	12.84	RIL 72	13.16	RIL 53	12.58	RIL 5	13.81	RIL 20	13.41
RIL 145	12.59	RIL 5	12.97	RIL 28	12.49	RIL 124	13.79	RIL 96	13.41
RIL 3	12.59	RIL 2	12.93	RIL 76	12.22	RIL 134	13.76	RIL 2	13.38
RIL 2	12.55	RIL 107	12.78	RIL 20	11.55	RIL 142	13.50	RIL 107	13.23
$LSD_{0.05}$	0.65	$LSD_{0.05}$	0.72	$LSD_{0.05}$	1.69	$LSD_{0.05}$	0.72	$LSD_{0.05}$	0.88

[a] GPC=grain protein content, %

in nitrogen allocation, increasing the GPC instead of wheat grain yield. Guttieri et al. (2000) and Underdahl et al. (2008a) also found that GPC increased under stress conditions. The GPC, a complex trait, is known to be influenced by environmental conditions such as temperature, soil fertility, and rainfall (Daniel and Triboi, 2000).

GPC had in general no significant association with kernel volume weight except at Carrington in 2010 (0.38) (Table 11). Similarly, Tsilo et al. (2010) reported no significant association between these two traits. GPC was also not associated with kernel hardness except at Carrington 2009 environment (-0.43) (Table 11). Recent study by Kerfal et al. (2010) also did not find significant association between these two traits while Nelson et al. (2006) found positive correlation between them. Negative association was found between GPC and flour extraction (0.24) (Table 11), contrary to Underdahl et al. (2008a) that found no association between these two traits. Hence, further studies may be needed to clarify the relationship between GPC and the other quality traits.

Kernel volume weight

Across locations, the mean for kernel volume weight (KVW) was 749 kg m^{-3} while the overall means of ND 705, PI 414566, RIL, and checks were 791, 598, 749, and 774 kg m^{-3}, respectively. The RIL range for KVW was from 669 to 790 kg m^{-3} (Table 6). The check Glenn was constantly the top genotype for KVW as was previously reported by Mergoum et al. (2006) (Table 12). Carrington 2009 had the highest values for KVW followed by Carrington 2010 environment (Table 6). Carrington environments had better climate conditions favoring this quality indicator trait. Based on KVW overall means, the top 10 genotypes for KVW were three checks (Glenn, Barlow, and Steele-ND), six RIL (47, 149, 30, 124, 167, and 47), and one the parent ND 705. While, the bottom group with

the lowest values of KVW comprised the rest of RIL plus the non-adapted parent, PI 414566 (Table 12). Glenn, ND 705, and the RIL 47 KVW were very consistent across environments. In general, KVW has a higher heritability compared to grain yield (Patil et al., 1994; Tsilo et al., 2010) being more constant across environments.

Table 11. Correlation coefficients (r) among the quality traits and their significance for the RIL, their parents, and checks grown in four environments (Carrington and Prosper in 2009 and 2010). The homogenous r values were pooled and denoted in the table

	Environment	Grain protein content	Kernel hardness	Flour extraction
Kernel Volume Weight	Carr 10	0.38**		
	Carr 09		0.05 [a]	0.30** [a]
	Pros 09	-0.03 [a]		
	Pros 10			
Grain Protein Content	Carr 09		-0.43**	
	Carr 10			-0.24** [a]
	Pros 09		0.09 [a]	
	Pros 10			
Kernel Hardness	Carr 09			
	Carr 10			0.39** [a]
	Pros 09			
	Pros 10			

** Significant at the 0.01 probability level.
[a] Pooled correlation coefficients across environments.

For this trait, transgressive segregation was not observed. The adapted ND 705 parent had the highest value in this study, excluding the check Glenn, while the non-adapted PI 414566 parent had the lowest value for KVW (Table 12). The higher susceptibility for diseases overall of the non adapted parent PI 414566 compared to the adapted parent ND 705 could be a factor, among several others, influencing the large KVW range between these two parents. Also, the disease resistant genes could influence the KVW performance of the RIL population.

Table 12. Kernel volume weight and least square difference (LSD) of the top and bottom ten genotypes across all environments (Carrington and Prosper in 2009 and 2010)

Top 10 genotypes									
Carrington 2009		Carrington 2010		Prosper 2009		Prosper 2010		Overall env	
Treatment	KVW [a]	Treatment	KVW	Treatment	KVW	Treatment	KVW	Treatment	KVW
Glenn	821	RIL 60	802	Glenn	797	Glenn	789	Glenn	801
ND 705	820	RIL 6	801	ND 705	796	RIL 47	782	ND 705	791
RIL 83	816	Glenn	800	RIL 130	786	Barlow	771	RIL 47	790
RIL 74	813	ND 705	796	RIL 44	784	Faller	762	Barlow	783
RIL 94	811	RIL 119	795	RIL 128	782	RIL 124	762	RIL 149	782
RIL 130	811	RIL 88	792	RIL 47	781	RIL 136	760	RIL 30	781
RIL 59	811	RIL 30	788	RIL 89	781	RIL 143	759	RIL 124	778
RIL 58	809	RIL 149	788	RIL 143	779	RIL 149	758	Steele-ND	778
RIL 12	808	RIL 47	787	RIL 43	779	Alsen	757	RIL 167	777
RIL 47	808	RIL 74	787	RIL 149	779	RIL 167	757	RIL 46	776
Bottom 10 genotypes									
Carrington 2009		Carrington 2010		Prosper 2009		Prosper 2010		Overall env	
Treatment	KVW	Treatment	KVW	Treatment	KVW	Treatment	KVW	Treatment	KVW
RIL 37	761	RIL 127	730	RIL 166	719	RIL 32	601	RIL 145	719
RIL 97	758	RIL 166	727	RIL 112	717	RIL 35	596	RIL 32	715
RIL 145	756	RIL 3	725	RIL 127	712	RIL 118	596	RIL 3	708
RIL 127	755	RIL 35	725	RIL 139	708	RIL 144	593	RIL 162	706
RIL 35	752	RIL 110	720	RIL 110	705	RIL 73	590	RIL 127	702
RIL 68	751	RIL 106	716	RIL 35	702	RIL 96	586	RIL 37	701
RIL 166	744	RIL 152	715	RIL 106	695	RIL 37	575	RIL 35	694
RIL 119	737	RIL 162	708	RIL 152	687	RIL 97	536	RIL 97	689
RIL 6	723	RIL 21	708	RIL 34	685	PI 414566	522	RIL 106	669
PI 414566	619	PI 414566	673	PI 414566	577	RIL 106	486	PI 414566	598
$LSD_{0.05}$	10	$LSD_{0.05}$	13	$LSD_{0.05}$	24	$LSD_{0.05}$	31	$LSD_{0.05}$	31

[a] KVW=kernel volume weight, kg m^{-3}.

KVW was not associated with kernel hardness and was positively related with flour extraction in this study (0.30) (Table 11). Previous investigations (Lee et al., 2006) found positive correlation between these KVW and kernel hardness. Similarly, several authors (Ohm et al., 1998; Lyford et al., 2005; Lee et al., 2006) reported a positive correlation between KVW and flour extraction. KVW is known to be correlated with wheat quality (Bergman et al., 2000; Aguirre et al., 2002) and is commonly believed in the wheat market as a valuable trait to enhance flour extraction. Normally, genotypes with greater values of KVW are believed to have more dense kernels with the potential of more endosperm per unit of kernel that will yield in higher flour. KVW has been used in many countries as an indicator of overall grain quality and an important factor to determine wheat prices. However, some authors (Schuler et al., 1995; Bergman et al., 2000) did not find any association between KVW and flour extraction. Lyford et al. (2005) mentioned that two genotypes can "pack" differently due to physical characteristics, affecting KVW values, but having similar flour extraction percentages. Similarly, in ND, the check Glenn, the most grown cultivar in the USA wheat spring region since 2007, has a very high KVW (Mergoum et al., 2006) but average flour extraction (Dr. M. Mergoum, personal communication).

Kernel hardness

Kernel hardness is an important index for differentiation of wheat classes. In this work, the overall mean for kernel hardness was 70 (hard texture). The means of ND 705, PI 414566, RIL, and checks were 76 (hard), 63 (medium hard), 70 (hard), and 77 (hard) respectively. The range of RIL was from 57 (medium hard) to 86 (very hard) (Table 6). This RIL population developed from a medium hard (PI 414566) and a hard (ND 705)

parent, no soft or extra hard texture RIL were found. Based on the overall mean, the top 10 hard genotypes were seven RIL (119, 6, 138, 54, 39, 104, and 19) and three checks (Barlow, Alsen, and Glenn). However, only the RIL 119 and 6, and Barlow were significantly harder compared to the other genotypes. The bottom group of 10 genotypes with the lowest values of kernel hardness included only RIL (Table 13).

The correlation between kernel hardness and flour extraction across all environments was 0.39 (Table 11). This result was in agreement with Ohm et al. (1998) and Nelson et al. (2006) who reported a positive association between these two traits. Increased kernel hardness requires higher milling energy, but based on our results and previous finding (Symes, 1965; Bassett et al., 1989), harder kernels had higher flour yield. Pomeranz and Williams (1990) did further studies and found that higher values of kernel hardness generate better flowing and sifting properties during milling. The main effect of hardness on bread-making qualities is attributed to higher starch damage during milling. This damage increases both water absorption and hydrolysis of starch into fermentable sugars that contribute to loaf volume (Pomeranz and Williams, 1990). However, Lyford et al. (2005) found negative association between these two traits. Their explanation for the negative correlation was due to the fact that working with hard wheat population, softer kernels will generate higher flour extraction rates.

Flour extraction

Flour extraction represents an important economic variable to millers. The overall mean for flour extraction across the four environments was 61 % while the means of ND705, PI 414566, RIL, and the checks were 66, 54, 61, and 63 %, respectively. The flour

Table 13. Kernel hardness and least square difference (LSD) of the top and bottom ten genotypes across all environments (Carrington and Prosper in 2009 and 2010)

Top 10 genotypes									
Carrington 2009		Carrington 2010		Prosper 2009		Prosper 2010		Overall env	
Treatment	KH [a]	Treatment	KH	Treatment	KH	Treatment	KH	Treatment	KH
RIL 6	87.9	RIL 119	83.7	RIL 119	86.9	RIL 119	89.0	RIL 119	86.5
RIL 119	86.9	RIL 104	82.2	RIL 6	84.8	RIL 138	87.0	RIL 6	84.4
RIL 2	86.9	RIL 6	80.0	RIL 138	80.5	RIL 54	85.2	Barlow	82.1
RIL 39	85.6	RIL 19	79.6	Barlow	80.4	Barlow	85.1	RIL 138	81.3
RIL 11	84.9	RIL 138	79.1	RIL 26	79.8	RIL 6	84.5	RIL 54	80.4
Barlow	84.5	Alsen	79.0	RIL 127	79.3	Glenn	84.5	RIL 39	80.2
RIL 111	83.6	Barlow	78.2	Glenn	79.1	RIL 104	84.2	Alsen	79.8
RIL 107	83.2	RIL 17	77.6	RIL 56	78.8	RIL 41	83.6	Glenn	79.3
RIL 145	82.7	Glenn	76.7	RIL 54	78.8	RIL 19	83.3	RIL 104	79.1
RIL 3	82.7	RIL 39	76.4	ND 705	78.5	RIL 66	83.3	RIL 19	78.9
Bottom 10 genotypes									
Carrington 2009		Carrington 2010		Prosper 2009		Prosper 2010		Overall env	
Treatment	KH	Treatment	KH	Treatment	KH	Treatment	KH	Treatment	KH
RIL 8	66.0	RIL 57	58.2	RIL 18	57.9	RIL 118	59.0	RIL 18	60.3
RIL 18	64.5	RIL 8	57.4	RIL 8	56.9	RIL 14	58.5	RIL 45	60.3
RIL 90	64.0	RIL 9	57.2	RIL 57	56.3	RIL 18	58.2	RIL 57	59.7
RIL 81	62.2	RIL 33	57.2	RIL 31	55.5	RIL 20	57.9	RIL 55	59.6
RIL 10	62.0	RIL 45	57.1	RIL 122	55.4	RIL 57	57.5	RIL 33	59.0
RIL 40	62.0	RIL 81	57.1	RIL 81	55.4	RIL 112	57.5	RIL 8	58.4
RIL 4	61.2	RIL 23	57.0	RIL 55	54.0	RIL 81	57.1	RIL 81	58.0
RIL 122	59.5	RIL 122	55.7	RIL 45	53.8	RIL 55	56.0	RIL 122	57.4
RIL 130	59.2	RIL 20	55.6	RIL 33	52.1	RIL 8	53.5	RIL 20	57.4
PI 414566	48.9	RIL 76	42.3	RIL 20	45.1	RIL 76	46.9	RIL 76	56.8
$LSD_{0.05}$	3.3	$LSD_{0.05}$	4.0	$LSD_{0.05}$	5.5	$LSD_{0.05}$	4.1	$LSD_{0.05}$	4.7

[a] KH=kernel hardness is a category classification according to the hardness index (HI) shown in the material and methods section.

extraction of the RIL ranged from 46 to 67%. In general, the genotypes had better flour extraction at Carrington than at Prosper (Table 6).

The parent ND 705 was part of the top 10 genotypes with the highest values of flour extraction. This was probably the main reason why several RIL such as 82 and 141 had high flour extraction, slightly better even than the best check (Faller) and better than the adapted parent ND 705 (Table 14). Flour extraction is related to several factors such as disease resistance. The incorporation of disease resistance in the genotypes will reduce the occurrence of shriveled grain; consequently, flour extraction yield will increase (Cox et al., 1989). ND 705 had satisfactory disease resistance overall. Probably, the RIL 82 and 141 incorporated these resistant genes from ND 705 because PI 414566 was highly susceptible for diseases overall. The non-adapted parent was located among the 10 genotypes with the lowest values for flour extraction along with nine RIL (Table 14).

Associations among agronomic and quality traits

Grain yield was significantly associated with kernel volume weight. The correlations at Carrington 2009 and 2010, and at Prosper 2009 were pooled (0.49). At Prosper 2010, the correlation between these two traits was highly significant (0.78). It is worth to notice that the correlation was higher in the environment under the highest stress conditions (Prosper 2010) (Table 15). Guttieri et al. (2001) found in their study that drought-sensitive genotypes had a significant reduction in grain yield and also KVW. However, the superior genotypes for grain yield were also the superior genotypes for KVW under drought conditions confirming our results. The same study concluded that the genotypes with stable grain yield under stress conditions were also the genotypes with stable KVW. Other studies (McCartney et al., 2005; Lee et al., 2006) also found positive

Table 14. Flour extraction and least square difference (LSD) of the top and bottom ten genotypes across all the environments (Carrington 2009 and 2010, and Prosper 2009 and 2010) and overall environments

Top 10 genotypes									
Carrington 2009		Carrington 2010		Prosper 2009		Prosper 2010		Overall env	
Treatment	FE [a]	Treatment	FE	Treatment	FE	Treatment	FE	Treatment	FE
RIL 82	69.8	RIL 82	70.8	RIL 82	65.8	RIL 127	70.3	RIL 82	66.8
RIL 65	68.3	RIL 89	69.4	ND 705	65.0	Faller	65.3	RIL 141	66.4
RIL 70	68.2	RIL 163	68.8	RIL 141	64.9	PI 414566	65.2	Faller	66.1
RIL 83	67.9	RIL 87	68.7	RIL 70	64.5	Barlow	64.8	ND 705	65.9
RIL 6	67.9	Faller	68.5	Faller	63.9	RIL 135	64.8	RIL 127	65.9
RIL 141	67.9	RIL 2	68.5	RIL 146	63.9	RIL 141	64.7	RIL 89	65.8
ND 705	67.9	RIL 156	68.5	RIL 143	63.6	RIL 152	64.7	RIL 163	65.3
RIL 87	67.8	RIL 65	68.4	RIL 43	63.5	RIL 163	64.5	RIL 2	65.3
RIL 69	67.8	RIL 141	68.3	RIL 58	63.2	RIL 139	64.4	RIL 65	65.3
RIL 146	67.8	RIL 49	67.9	RIL 46	63.1	ND 705	64.0	RIL 152	65.2
Bottom 10 genotypes									
Carrington 2009		Carrington 2010		Prosper 2009		Prosper 2010		Overall env	
Treatment	FE	Treatment	FE	Treatment	FE	Treatment	FE	Treatment	FE
RIL 130	59.0	RIL 101	59.2	RIL 23	52.9	RIL 37	48.5	RIL 98	55.6
RIL 10	58.8	RIL 140	59.2	RIL 128	52.7	RIL 90	48.2	RIL 14	55.4
RIL 61	58.6	RIL 46	59.0	RIL 10	52.5	RIL 118	48.2	RIL 116	54.9
RIL 98	58.5	RIL 130	58.9	RIL 34	52.2	RIL 40	47.5	PI 414566	54.2
RIL 40	58.0	RIL 116	58.8	RIL 40	52.2	RIL 14	47.4	RIL 8	54.1
Alpine	57.6	RIL 33	58.1	RIL 98	51.7	RIL 101	47.0	RIL 40	53.9
RIL 14	57.3	RIL 40	58.0	RIL 116	51.7	RIL 81	45.7	RIL 117	53.9
RIL 81	53.2	RIL 122	55.5	RIL 81	47.8	RIL 8	42.2	RIL 76	53.1
PI 414566	48.0	RIL 81	53.0	PI 414566	40.8	RIL 117	39.6	RIL 81	49.9
RIL 122	45.6	RIL 76	46.4	RIL 122	32.1	RIL 76	38.3	RIL 122	46.0
$LSD_{0.05}$	2.3	$LSD_{0.05}$	3.1	$LSD_{0.05}$	4.3	$LSD_{0.05}$	5.4	$LSD_{0.05}$	3.6

[a] FE=flour extraction, %.

and significant association between these two traits under regular field conditions. However, Tsilo et al. (2010) did not detect significant correlation between grain yield and KVW. Guttieri et al. (2001), McCartney et al. (2005), and Lee et al. (2006) worked with released cultivars, double haploid population developed from two adapted parents, and blended samples from two adapted cultivars, respectively, and they identified positive correlation between these traits. This work showed that with the use of a non-adapted parent, this population had also significant association between these two important traits.

Grain yield was significantly correlated with GPC only at Carrington in 2010 (0.18) (Table 15). Usually, these two traits are known to have negative relationship due to temperature and nitrogen shifts allocating the nitrogen to one or to the other trait (Daniel and Triboi, 2000). Groos et al. (2003) also reported a negative and significant association between these two traits. However, these authors concluded that there was no strong negative pleiotropic effect detected between these two traits, suggesting that both traits can be improved using the same breeding scheme. Other studies (Underdahl et al., 2008a) also did not find negative and significant correlation between grain yield and GPC confirming our findings.

Our results show that grain yield and flour extraction had positive and significant correlations. These correlations across environments were homogeneous and, consequently, pooled together (0.22). Lee et al. (2006) found also a positive and significant correlation between grain yield and flour extraction. These traits represent the most important economic variable to farmers and millers, respectively. The main goal of wheat breeding is to develop cultivars with higher attributes for both traits.

Table 15. Correlation coefficients (r) among the agronomic and quality traits, and their significance for the RIL, their parents, and checks grown in four environments (Carrington and Prosper in 2009 and 2010). The r values for single environment are denoted in the table

		Kernel volume weight	Grain protein content	Kernel hardness	Flour extraction
Grain yield	Carr 10		0.18* [a]	0.17*	
	Carr 09	0.49**			0.22**
	Pros 09		-0.11	-0.09 [a]	
	Pros 10	0.78** [a]		0.17*	
1000-kernel weight	Carr 10	0.38**	0.37** [a]		
	Carr 09			-0.18*	0.27**
	Pros 10	0.79** [a]	-0.03		
	Pros 09	0.61** [a]		-0.45** [a]	
Number of kernels per spike	Carr 09	0.02	-0.45**	0.38** [a]	
	Carr 10				0.14
	Pros 09	0.47**	-0.06	0.01	
	Pros 10				
Number of spikes per m^2	Carr 09				
	Carr 10	0.27**	0.05	0.16*	0.05
	Pros 09				
	Pros 10				
Plant height	Carr 09				
	Carr 10	0.23**	0.07	0.08	0.03
	Pros 09				
	Pros 10		-0.27** [a]		
Days to heading	Carr 10	-0.17*	-0.10		
	Carr 09	-0.58**		0.11	-0.18*
	Pros 10		0.27**		
	Pros 09	-0.17*		0.45** [a]	
Maturity	Carr 09	---	---	---	---
	Carr 10	0.12 [a]		0.14	
	Pros 10		0.16*		-0.15
	Pros 09	-0.29**		0.48** [a]	
Lodging	Carr 09	-	-	-	-
	Carr 10	-0.25** [a]	-0.22** [a]		
	Pros 09	0.15 [a]	0.16*	-0.02	-0.06
	Pros 10	-0.66** [a]			
Spike color	Carr 09	---	---	---	---
	Carr 10	---	---	---	---
	Pros 09	-0.35**	-0.04 [a]	-0.09	-0.14
	Pros 10		0.38** [a]		

* and ** Significant at the 0.05 and 0.01 probability levels, respectively.
- No lodging at this location.
--- Traits were not collected at these locations.
[a] Correlation coefficients for a single environment.

The 1000-kernel weight was positively associated with KVW and flour extraction across all the environments (Table 15). That indicates why 1000-kernel weight was used to predict flour extraction by Lyford et al. (2005). McCartney et al. (2005) reported also a significant and positive correlation between these two traits. According to these results, 1000-kernel weight seems to be the yield component that has the greatest influence on KVW and flour extraction, indicating that the heavier kernels are related to higher values for these two quality traits. However, Bergman et al. (2000) did not find any association between 1000-kernel weight and KVW, and flour extraction. They concluded that 1000-kernel weight cannot be used to predict these two quality traits.

Days to heading and maturity were significantly and negatively correlated with KVW. Days to heading also had a negative association with flour extraction (Table 15). Similarly, Tsilo et al. (2010) reported a significant negative correlation between these two traits confirming our findings. These results might suggest that late heading genotypes apparently had lower grain filling rates than early genotypes; consequently, they had lower values of KVW giving the short grain filling period known in the USA spring wheat region. The late heading genotypes might have the genes for late heading and maturity coming from PI 414566 as well as the genes for low KVW. However, McCartney et al. (2005) found a significant positive correlation between these two traits indicating that the late samples had better KVW than the early plants. This result can be adhered in most environments that have long grain filling period.

The highest negative correlation between lodging and KVW was reported at Prosper in 2010 (0.66) (Table 15). This high correlation at this environment is due to the unfavorable conditions that increased the percentage of lodging at this location;

consequently, affecting significantly the KVW for the genotypes that are susceptible to lodging. Contrary to our findings, McCartney et al. (2005) found a positive relationship between these two traits.

Spike color was negatively associated with KVW (0.35) (Table 15). This indicates that the RIL with darker colored spikes, with the red colored spike genes coming from the non-adapted parent PI 414566, may carry genes for low KVW or some of the spike color genes may be linked to KVW genes.

CONCLUSIONS

The adapted (ND 705) and non-adapted parent (PI 414566) showed drastic differences for all the agronomic and quality traits studied. Consequently, the RIL showed significant variances for these traits. Transgressive segregation occurred for all the agronomic and quality traits except for spike color and KVW, suggesting possible traits improvement using these types of mating.

Overall, the genotypes had better performance for agronomic traits, kernel volume weight, and flour extraction in Carrington compared to Prosper due to favorable environment conditions at Carrington during these two years. Within locations, except for flour extraction that had similar results at Carrington 2009 and 2010, these traits had higher levels in 2009 than 2010.

The checks Faller, Barlow, and Steele-ND along with the RIL 143 and 88 had significant superior grain yield across environments. The RIL 143 and 88 showed relatively low rates of lodging and kernel shattering and overall good agronomic attributes, demonstrating high potential to be included in the HRSW breeding program. Also, the RIL 47 had superior attributes for KVW, and RIL 82 and 141 had superior attributes for flour extraction. These RIL can be valuable for the HRSW breeding program.

Grain yield along with GPC, KVW, and flour extraction are the most important traits for farmers and millers to remain competitive. These four traits are complex and influenced by many factors and will continue to be a major challenge for wheat breeders, farmers, and millers. The use of elite x elite crosses continues to be the most common and effective type of cross to develop new cultivars in breeding programs. However, with the concern that narrow germplasm may restrict breeding flexibility and slow genetic progress

to biotic and abiotic stresses tolerance, it seems that the development of populations originated from adapted x non-adapted germplasm is a common practice by breeders to broaden the genetic variability of their germplasm and consequently increase genetic gain. This study has showed clearly the value of using non-adapted sources of germplasm to broaden the germplasm and to improve several wheat economic traits.

REFERENCES

AACC International. 2000. Approved Methods of the American Association of Cereal Chemists, 10th ed. Methods 46-30, 55-10, and 56-81B. The Association: Saint Paul, MN.

Aguirre, A., O. Badiali, M. Cantarero, A. Leon, P. Ribotta, and O. Rubiolo. 2002. Relationship of test weight and kernel properties to milling and baking quality in Argentine triticales. Cereal Research Communications 30:203-208.

Alexander, W.L., E.L. Smith, and C. Dhanasobhan. 1984. A comparison of yield and yield component selection in winter wheat. Euphytica 33:953-961.

Baril, C.P. 1992. Factor regression for interpreting genotype-environment interaction in bread-wheat trials. Theoretical and Applied Genetics 83:1022-1026.

Bassett, L.M., R.E. Allan, and G.L. Rubenthaler. 1989. Genotype x environment interactions on soft white winter wheat quality. Agronomy Journal 81:955-960.

Bergman, C.J., D.G. Gualberto, K.G. Campbell, M.E. Sorrells, and P.L. Finney. 2000. Kernel morphology variation in a population derived from a soft by hard wheat cross and associations with end-use quality traits. Journal of Food Quality 23:391-407.

Cox, T.S., J.P. Shoyer, L. Ben-Hui, R.G. Sears, and T.J. Martin. 1988. Genetic improvement in agronomic traits of hard red winter wheat cultivars from 1919 to 1987. Crop Science 28:756-760.

Cox, T.S., M.D. Shogren, R.G. Sears, T.J. Martin, and L.C. Bolte. 1989. Genetic improvement in milling and baking quality of hard red winter wheat cultivars, 1919 to 1988. Crop Science 29:626-631.

Daniel, C., and E. Triboi. 2000. Effects of temperature and nitrogen nutrition on the grain composition of winter wheat: effects on gliadin content and composition. Journal of Cereal Science 32:45-56.

Devkota, R.N., J.C. Rudd, Y. Jin, K.D. Glover, R.G. Hall, and G.A. Hareland. 2007. Registration of 'Briggs' wheat. Crop Science 47:432-434.

Doehlert, D.C., M.S. McMullen, J. Jannink, S. Panigrahi, H. Gu, and N.R. Riveland. 2004. Evaluation of oat kernel size uniformity. Crop Science 44:1178-1186.

Donmez, E., R.G. Sears, J.P. Shroyer, and G.M. Paulsen. 2001. Genetic gain in yield attributes of winter wheat in the Great Plains. Crop Science 41:1412-1419.

Eagles, H.A., and J.E. Lothrop. 1994. Highland maize from Central México – its origin, characteristics and use in breeding programs. Crop Science 34:11-19.

Finney, K.F., W.T. Yamazaki, V.L. Youngs, and G.L. Rubenthaler. 1987. Quality of hard, soft, and durum wheats. p. 677-748. *In*: Wheat and Wheat Improvement. 2nd Ed. E.G. Heyne, ed. ASA: Madison, WI.

Frohberg, R.C., R.W. Stack, T. Olson, J.D. Miller, and M. Mergoum. 2006. Registration of 'Alsen'. Crop Science 46:2311-2312

Geng, S., P. Schneeman, and W.J. Wang. 1982. An empirical study of the robustness of analysis of variance procedures in the presence of commonly encountered data problems. American Journal of Enology and Viticulture 33:131-134.

Groos, C., N. Robert, E. Bervas, and G. Charmet. 2003. Genetic analysis of grain protein-content, grain yield and thousand-kernel weight in bread wheat. Theoretical and Applied Genetics 106:1032-1040.

Guttieri, M.J., J.C. Stark, K. O'Brien, and E. Souza. 2001. Relative sensitivity of spring wheat grain yield and quality parameters to moisture deficit. Crop Science 41:327-335.

Guttieri, M.J., R. Ahmad, J.C. Stark, and E. Souza. 2000. End-use quality of six hard red spring wheat cultivars at different irrigation levels. Crop Science 40:631-635.

Hartley, H.O. 1950. The maximum F-ratio as a short-cut test for heterogeneity of variances. Biometrika 37:308-312.

Holley, R. N., and M. M. Goodman. 1988. Yield potential of tropical hybrid corn derivatives. Crop Science 28:213-217.

Hsu, P., and P.D. Walton. 1971. Relationship between yield and its components and structures above the flag leaf node in spring wheat. Crop Science 11:190-193.

Kerfal, S., P. Giraldo, M. Rodríguez-Quijano, J.F. Vázquez, K. Adams, O. M. Lukow, M.S. Röder, D.J. Somers, and J.M. Carrillo. 2010. Mapping quantitative trait loci (QTLs) associated with dough quality in a soft x hard bread wheat progeny. Journal of Cereal Science 52:46-52.

Khlestkina, E.K., E.A. Salina, T.A.Pshenichnikova, M.S. Roder, and A. Borner. 2009. Glume coloration in wheat: allelism test, consensus mapping and its association with specific microsatellite allele. Cereal Research Communications 37:37-43.

Lee, K.M., J.P. Shroyer, T.J. Herrman, and J. Lingenfelser. 2006. Blending hard white wheat to improve grain yield and end-use performances. Crop Science 46:1124-1129.

Lyford, C.P., W. Kidd, P. Rayas-Duarte, and C. Deyoe. 2005. Prediction of flour extraction rate in hard red winter wheat using the single kernel characterization. Journal of Food Quality 28:279-288.

Marshall, D.R., F.W. Ellison, and D.J. Mares. 1984. Effects of grain shape and size on milling yields in wheat. 1. Theoretical-analysis based on simple geometric-models. Australian Journal of Agricultural Research 35:619-630.

McCartney, C.A., D.J. Somers, D.G. Humphreys, O. Lukow, N. Ames, J. Noll, S. Cloutier, and B.D. McCallum. 2005. Mapping quantitative trait loci controlling agronomic traits in the spring wheat cross RL4452 × 'AC Domain'. Genome 48:870-883.

Mergoum, M., R.C. Frohberg, J.D. Miller, and R.W. Stack. 2005. Registration of 'Steele-ND' wheat. Crop Science 45:1163-1164.

Mergoum, M., R.C. Frohberg, R.W. Stack, J.W. Rasmussen, and T.L. Friesen. 2008. Registration of 'Faller' spring wheat. Journal of Plant Registrations 2:224-229.

Mergoum, M., R.C. Frohberg, R.W. Stack, T. Olson, T.L. Friesen, and J.B. Rasmussen. 2006. Registration of 'Glenn' wheat. Crop Science 46:473-474.

Mergoum, M., S. Simsek, R.C. Frohberg, J.B. Rasmussen, T.L. Friesen, and T. Adhikari. 2011. 'Barlow': A high-quality and high-yielding hard red spring wheat cultivar adapted to the North Central Plains of the USA. Journal of Plant Registrations 5:62-67.

Mitchell, M. 1995. Using XSLIP-STAT to explore new and unusual statistic paradigms. Perspective 19:28-32.

NDAWN, 2011. Weather data [online]. North Dakota Agricultural Weather Network. Available at http://www.ndawn.ndsu.nodak.edu (verified 24 July 2011).

Nelson, J.C, C. Andreescu, F. Breseghello, P.L. Finney, D.G. Gualberto, C.J. Bergman, R.J. Peña, M.R. Perretant, P. Leroy, C.O. Qualset, and M.E. Sorrells. 2006. Quantitative trait locus analysis of wheat quality traits. Euphytica 149:145-159.

Ohm, J.B., O.K. Chung, and C.W. Deyoe. 1998. Single-kernel characteristics of hard winter wheats in relation to milling and baking quality. Cereal Chemistry 75:156-161.

Otteson, B.N., M. Mergoum, and J.K. Ransom. 2007. Seeding rate and nitrogen management effects on spring wheat yield and yield components. Agronomy Journal 99:1615-1621.

Otteson, B.N., M. Mergoum, and J.K. Ransom. 2008. Seeding rate and nitrogen management on milling and baking quality of hard red spring wheat genotypes. Crop Science 48:749-755.

Patil, V.D., M.V.S. Reddy, and Y.S. Nerkar. 1994. Efficiency of early generation selections for yield and related characters in safflower (*Carthamus tinctorius* L.). Theoretical and Applied Genetics 89:293-296.

Patterson, A.H., S.D. Tanksley, and M.E. Sorrells. 1991. DNA markers in plant improvement. Advances in Agronomy 46:39-90.

Patterson, H.D., and V. Silvey. 1980. Statutory and recommended list trials of crop varieties in the United Kingdom. Journal of the Royal Statistical Society [Series A] 143:219-252.

Pomeranz, Y., and P.C. Williams. 1990. Wheat hardness: its genetic, structural and biochemical background, measurements and significance. *In*: Y. Pomeranz (Ed.), Advances in Cereal Science and Technology, 10:471-544. AACC, St. Paul, MN.

SAS Institute. 2004. SAS Online Doc, version 9.1.2 SAS Institute., Cary, NC.

Schuler, S.F., R.K. Bacon, P.L. Finney, and E.E. Gbur. 1995. Relationship of test weight and kernel properties to milling and baking quality in soft red winter wheat. Crop Science 35:949-953.

Souza, E.J., J.M. Martin, M.J. Guttieri, K. O'Brien, D.K. Habernicht, S.P. Lanning, G.R. Carlson, and L.E. Talbert. 2004. Influence of genotype, environment, and nitrogen management on spring wheat quality. Crop Science 44:425-432.

Stalker, H.T. 1980. Utilization of wild species for crop improvement. Advances in Agronomy 33:111-147.

Stelmakh, A.F. 1998. Genetic systems regulating flowering response in wheat. Euphytica 100:359-369.

Symes, K.J. 1965. The inheritance of grain hardness in wheat as measured by the particle-size index. Australian Journal of Agricultural Research 16:113-123.

Tsilo, T.J., G.A. Hareland, S. Simsek, S. Chao, and J.A. Anderson. 2010. Genome mapping of kernel characteristics in hard red spring wheat breeding lines. Theoretical and Applied Genetics 121:717-730.

Underdahl, J. L., M. Mergoum, B. Schatz, and J. K. Ransom.2008a. Quality trait variation in major hard red spring wheat cultivars released in North Dakota since 1968. Cereal Chemistry 85:507-514.

Underdahl, J.L., M. Mergoum, J.K. Ransom, and B.G. Schatz. 2008b. Agronomic traits improvement and associations in hard red spring wheat cultivars released in North Dakota from 1968 to 2006. Crop Science 48:158-166.

USDA-ERS. 2011. World wheat production, supply and disappearance. Available at http://www.ers.usda.gov/data/wheat/WheatYearbook.aspx (verified 11 Mar 2011). Economic Research Service, Washington, DC.

USDA-NRCS. 2011. Web Soil Survey [online]. Available at http://websoilsurvey.nrcs.usda.gov/app/ (verified 23 February 2011). Natural Resource Conservation Service, Washington, DC.

Waddington, S.R., J.K. Ransom, M. Osmanzai, and D.A. Sauders. 1986. Improvement in the yield potential of bread wheat adapted to Northwest Mexico. Crop Science 26:698-703.

Zamir, D. 2001. Improving plant breeding with exotic genetic libraries. Nature Review-Genetics 2:983-989.

Zeven, A.C. 1983. The character brown ear of bread wheat: a review. Euphytica 32:299-310.

Table A1. Coefficient of variance (CV%), error mean square (EMS), and ratio between EMS of environments for agronomic and quality traits evaluated at Carrington and Prosper, ND, in 2009 and 2010

Trait	Carrington 2009		Prosper 2009		Carrington 2010		Prosper 2010		Ratio [b]
	CV%	EMS	CV%	EMS	CV%	EMS	CV%	EMS	
Grain yield	8.00	83,823	15.56	206,315	10.39	125,186	13.86	95,126	2.46
Spikes number per m^2	13.73	3759	17.35	12521	14.19	7592	16.45	9212	3.33
Kernels number per spike	7.41	8.87	14.97	21.27	7.93	8.65	12.99	13.17	2.46
Thousand kernel weight by DIA	3.10	0.95	6.91	3.91	5.04	2.28	5.41	1.63	4.11
Plant height	4.36	14.36	4.87	21.15	3.41	10.25	4.34	14.87	2.06
Days to heading	1.60	0.82	2.47	1.62	1.33	0.71	2.11	1.13	2.29
Lodging	0.00	0.00	94.33	715.60	317.12	11.66	29.84	210.05	> 10.00
Maturity [a]	-	-	2.90	6.58	1.70	2.88	1.35	1.35	4.87
Emergence [a]	-	-	-	-	-	-	1.55	2.36	-
Spike color [a]	-	-	-	-	14.02	0.24	19.49	0.26	1.08
Grain protein content	2.22	0.11	5.98	0.73	2.41	0.13	2.34	0.13	6.82
Kernel volume weight	0.66	26.65	1.65	153.25	0.88	44.66	2.22	239.97	9.01
Kernel hardness	2.22	2.72	4.09	7.86	3.02	4.11	2.88	4.25	2.90
Flour extraction	1.83	1.34	3.68	4.63	2.43	2.44	4.76	7.47	5.57

[a] Traits were not evaluated in all the environments.

[b] Ratio: test of homogeneity (greatest EMS / smallest EMS) should be smaller than 10-fold.

- Traits were not collected at these locations.

PAPER 2. VARIATION OF WHEAT KERNEL CHARACTERISTICS AND ASSOCIATIONS WITH AGRONOMIC AND QUALITY TRAITS

ABSTRACT

Wheat kernel is the main component of wheat grain yield and an important factor for milling characteristics. Focusing on wheat kernel characteristics might be a key element to improve wheat genotypes for agronomic and quality traits. A study conducted at Carrington and Prosper, North Dakota during 2009 and 2010 evaluated the associations among kernel traits including kernel length, width, length/width ratio, weight and area, and agronomic and quality traits. A population of 160 Recombinant Inbred Lines (RIL) developed between two contrasting genotypes (ND 705 and PI 414566) was used to study these associations. The results showed that kernel traits had high correlation among them and they exhibited a normal distribution suggesting a polygenic inheritance. Grain yield, kernel volume weight, and flour extraction were highly correlated with kernel width, length/width ratio, weight, and area. Results from this study indicate that kernel traits can play a significant role in improving agronomic and quality traits. Higher values for grain yield, kernel volume weight, and flour extraction were significantly associated with spheroid or round shape (short and plump), large, and heavy kernels. Based on this study, breeders should pay attention to kernel traits during the parental selection for the development of populations with improved agronomic and quality attributes.

INTRODUCTION

Wheat kernel is the main component of wheat grain yield. Also, it has been reported by researchers (Campbell et al., 1999; Wiersma et al., 2001; Dholakia et al., 2003; Breseghello and Sorrells, 2007; Sun et al., 2009) that kernel traits including kernel weight and size are important for milling characteristics. The focus on wheat kernel characteristics might be a key point in the selection process for the improvement of wheat; therefore, increasing the possibility of success in the breeding program. However, wheat kernel traits are generally complex and believed to be of quantitative nature of inheritance. In addition, wheat kernel characteristics are frequently influenced by environment, having high genotype x environment interaction (Campbell et al., 1999; Sun et al., 2009). Likewise, most of the wheat quality traits are under complex genetic and environmental control (Nelson et al., 2006; Dornez et al., 2008).

Kernel characteristics, such as kernel length, width, shape, area, and weight are very important, as they are highly related to wheat quality traits. However, these traits are in general not well investigated. Kernel weight can be affected by kernel length and width (Campbell et al., 1999; Dholakia et al., 2003) and based on some studies (Wiersma et al., 2001), it enhances flour yield. Larger kernels were reported to increase flour extraction values (Marshall et al., 1986; Chastain et al., 1995). Similarly, kernel shape affects kernel volume weight (KVW) (Campbell et al., 1999). Also, in a geometrical model study, changes in kernel shape and size were able to enhance flour extraction up to 4.7% (Marshall et al., 1984). High values of flour yield can also be achieved by increased kernel hardness (Symes, 1965; Bassett et al., 1989).

In recent years, scientists (Dholakia et al., 2003; Breseghello and Sorrells, 2007; Sun et al., 2009) have focused on traits to be used as markers for indirect selection to improve wheat quality attributes. Dholakia et al. (2003) and Sun et al. (2009) found similar associations among the kernels characteristics working with two different Recombinant Inbred Lines (RIL) populations. Dholakia et al. (2003) developed a 106 RIL population by crossing two hexaploid wheat cultivars PH 132 and WL 711. In this study, kernel weight showed a significant correlation with kernel length and width (0.51, 0.68, respectively). Kernel length and width were also significant correlated (0.57). Similarly, Sun et al. (2009) worked with a set of 131 RIL population developed from the cross between two Chinese lines (Chuan 35050 x Shannong 483). They also reported that kernel weight had significant correlation with kernel length and width (0.42, and 0.75, respectively) and kernel length and width were also significant correlated (0.22). In addition, Sun et al. (2009) compared the association between KVW with the kernel traits. They found a significant positive association of KVW and kernel weight (0.19), negative correlation with kernel length (0.22), and no significant relationship with kernel width.

Lee et al. (2006) used two hard white wheats, 'Betty' and 'Trego', which were combined at different percentages and average values from the blended percentages and locations were associated. Kernel width had significant positive correlation with flour extraction and KVW (0.50, and 0.28, respectively). This finding was not in agreement with Sun et al. (2009) that did not find any correlation between KVW and kernel width. However, kernel weight was positively associated with flour extraction and KVW (0.74, and 0.56, respectively) which was in agreement with Sun et al. (2009) findings. Kernel weight and width were also strongly correlated (0.83). Lee et al. (2006) reported positive

association between KVW and flour extraction (0.44). A research by Breseghello and Sorrells (2006) using 95 selected soft winter wheat cultivars released by 35 different seed companies or research institutions in the last 30 years studied the associations between kernel traits (kernel length, width, weight, and area) and quality traits such as flour extraction. Contrary to Lee et al. (2006), they did not find significant correlations between kernel traits and flour extraction, except between flour extraction and kernel weight at only one location (0.23).

Breseghello and Sorrells (2007) used two different RIL populations, SxO and RxG. SxO population derived from a cross between the synthetic hexaploid wheat W7984 and the spring wheat cultivar 'Opata 85'. RxG population developed from the cross between a soft white spring wheat cultivar 'AC Reed', and a hard red spring wheat (HRSW) cultivar 'Grandin'. The dimensions of the kernels were determined based on two photos of 20 kernels placed in orthogonal directions symbolizing each RIL. They found strong correlations among the kernel traits: kernel length and kernel area (0.90 and 0.84 in SxO and RxG populations, respectively); kernel length and kernel volume (0.78 and 0.67); kernel length and kernel weight (0.80 and 0.68); kernel width and kernel area (0.84 and 0.58); kernel width and kernel volume (0.68 and 0.69); and kernel width and kernel weight (0.68 and 0.67). However, the correlation between kernel length and kernel width was not as high as previous correlations (0.30 and 0.22). The population SxO showed stronger correlations among the kernel traits than population RxG. Also, there were stronger correlations between kernel length and the other traits (kernel area, volume, and weight) than kernel width and the other traits.

Recently, Tsilo et al. (2010) studied a population of 139 RIL derived from the cross between two HRSW lines MN98550 ('Bacup'/'McVey') and MN99394 (SD3236/SBF0402). The results from this study showed a strong correlation between kernel traits and KVW confirming previously reported results by Sun et al. (2009). KVW was also significantly associated with kernel width and weight (0.35, 0.29, respectively).

Several studies conducted on the kernel characteristics showed the potential role of these traits in the wheat quality. However, several other studies have reported conflicting results. Therefore, this study was designed to (1) examine the kernel characteristics variations derived from an elite x non-adapted parents, (2) the kernel characteristics associations with agronomic and quality traits to explore their potential uses in breeding programs, in order to develop new superior cultivars.

MATERIAL AND METHODS

Material

This study was conducted using a population of 160 RIL. This population was developed from a cross between ND 705 and PI 414566. The ND 705 is a HRSW line developed by the HRSW breeding program at North Dakota State University (NDSU). The PI 414566 is originally from China and it was obtained from the USDA National Small Grains Collection. It is described as a facultative type of wheat which requires cold temperature to flower, tall, brown kernels, and susceptible for diseases such as stripe rust caused by *Puccinia striiformis*. The RIL population was advanced through single seed descent method to F_8 generation and it was obtained from the Wheat Germplasm Enhancement program at NDSU. A combination of field nurseries at New Zealand, North Dakota, and greenhouse were used to advance this population. The selection of the parents used to develop this RIL population was based on the genetic divergence between the parents in order to produce a RIL population with significant polymorphism for kernel, agronomic, and quality traits. The number of RIL used in this study was determined by the amount of seeds available of each RIL.

The checks used in this study were: 'Alsen' (PI 615543) (Frohberg et al., 2006), 'Steele-ND' (PI 634981) (Mergoum et al., 2005), 'Glenn' (PI 639273) (Mergoum et al., 2006), 'Faller' (PI 648350) (Mergoum et al., 2008), 'Barlow' (PI 658018) (Mergoum et al., 2011), 'Briggs' (PI 632970) (Devkota et al., 2007), and 'Alpine'. All the check cultivars are HRSW varieties except Alpine which is a hard white spring wheat variety.

Field experiment

The RIL, their parents (ND 705 and PI 414566), and checks (Alsen, Steele-ND, Glenn, Faller, Barlow, Briggs, and Alpine) were evaluated in the summers of 2009 and 2010 at two field locations in ND; Carrington Research Extension Center near Carrington and Prosper Research Plots near Prosper. These sites represent two diverse environments. Carrington is located in the Central ND region with an altitude of 484 m. The soil type at Carrington is Heimdal-Emrick series (loamy, mixed, superactive, Calcic/Hapludolls). Prosper is located in the Red River Valley region (Eastern ND) with an altitude of 274 m. The soil type at Prosper is Bearden series (fine silty, mixed, superactive, frigid Aeric Calciaquolls) (USDA-NRCS, 2011).

In 2009, the planting dates were May 7 at Carrington, and May 28 at Prosper. The harvesting dates were August 26 at Carrington for all the lines except the parent PI 414566 which was harvested on October 6. At Prosper all the lines were harvested on September 18 except PI 414566 which was harvested on October 13. The winter of 2008-2009 had a high amount of snow causing field flood delaying the planting date. Consequently, PI 414566 as a facultative type requiring a longer period to complete its cycle did not have enough cold temperature to completely flowering. In 2010, the planting dates were April 21 at Carrington, and May 20 at Prosper. The harvesting dates were August 18 and 27 at Carrington and Prosper, respectively. PI 414566 was vernalized for three weeks prior to planting at Prosper. However, the emergence rate of the PI 414566 vernalized seeds was very low because of the poor and heavy soil characteristics at Prosper in 2010. Carrington was planted earlier; consequently, PI 414566 had enough cold temperature to flower,

therefore completing its cycle successfully. Sowing rates was 113 kg ha^{-1} in all environments.

In 2009, the experiments included the $F_{8:9}$ RIL, their parents, and checks. In fall 2009, the RIL were sent to New Zealand winter nursery for seed increase. Consequently, in 2010, the experiments included the $F_{10:11}$ RIL, their parents, and checks. The entries were arranged in a 13 x 13 partially balanced square lattice design, with two replicates. The experimental units consisted of seven rows, 2.44 m long and 12.7 cm apart.

Data collection

The grains from harvested plots were cleaned by using a clipper grain cleaner before measuring the following traits.

Agronomic traits

- Grain yield (kg ha^{-1}) – determined by weighing the cleaned grain harvested from each plot.
- Yield components:
  - ✓ Number of spikes (spikes m^{-2}) – determined by counting the number of spikes in two 61 cm long rows per plot. Counts were then converted to determine total number of spikes per square meter.
  - ✓ Number of kernels (kernels $spike^{-1}$) – calculated by averaging the kernels from ten individual spikes selected at random from each plot. The kernels were counted by the electronic seed counter (Seedburo Equipment Co., Chicago, IL) after threshing the spikes.
  - ✓ 1000-kernel weight (g) determined by 'Digital Image Analysis' (DIA) – calculated based on the number of kernels within 10 g samples from each

plot used in the DIA. The results were converted and the weight of 1000 kernels was determined (Doehlert et al., 2004).

- Plant height (cm) – measured from the soil surface to the top of the spike, excluding the awns at maturity. An average plant height was recorded from each plot.
- Days to heading (days) – recorded when inflorescences fully emerged in at least 50% of the spikes and was calculated as the number of days from planting to heading.
- Lodging (%) – estimated by the percent of lodged plants per plot when lodging occurred at a given environment.
- Maturity (days) – recorded when kernels fully complete the physiologic maturity in at least 50 % of the spikes. It was calculated as the number of days from planting to maturity.
- Emergence (%) – estimated by the percent of fully emerged plants per plot.
- Spike color (1-5) – visually rated by using a scale having equidistant values from 1 (white) to 5 (dark red).

Quality traits

- Grain protein content (%) – measured according to American Association of Cereal Chemists (AACC) standard method 46-30 (AACC International, 2000), using an Infratec 1226 Cold Grain Analyzer expressed on a 12 percent basis.
- Kernel volume weight (kg m^{-3}) – measured according to AACC standard method 55-10 (AACC International, 2000).

- Kernel hardness (0-90+) – measured using the single kernel characterization system (SKCS) 4100 instrument (Perten Instruments, Springfield, IL). This instrument measured the force needed to crush the kernels which is expressed as a hardness index. Table 1 has the kernel hardness category classification according to the hardness index.

Table 1. Kernel hardness category classification according to the hardness index (HI)

Category	HI [a]
Extra hard	90 +
Very hard	81 – 90
Hard	65 – 80
Medium hard	45 – 64
Medium soft	35 – 44
Soft	25 – 34
Very soft	10 – 24
Extra soft	Up to 10

[a] HI=hardness index

- Flour extraction (%) – 150 g cleaned wheat samples were tempered to 16.0 % moisture for 16 hr before being milled on a Brabender Quadrumat Junior Mill according to standard procedure used at the Department of Cereal and Food Sciences at North Dakota State University. The flour extraction percentage was calculated by dividing the flour weight of straight grade flour over the total grain weight milled (150 g).

Kernel traits

- Kernel length (mm) – determined by the DIA. Ten-gram samples were placed on a backlit glass platform within a predefined field of view. An analog camera was mounted at one meter height from the platform. Kernels were manually positioned on the platform ensuring they were not touching each other and a

photograph was taken. The images were processed in a computer program called 'Aphelion' (Amerinex Applied Imaging, Amherst, MA) to determine the kernels major and minor axes. The major axes represented the kernel length. Means were calculated from 10 g samples for each plot (Doehlert et al., 2004).

- Kernel width (mm) – determined by the DIA. In the same manner as kernel length except that in this case the minor axes determined by Aphelion program represented the kernel width. This was determined by the means of kernels in 10 g samples from each plot (Doehlert et al., 2004).
- Kernel length / width by DIA ratio – calculated by dividing the kernel length mean by the kernel width mean for each plot.
- Kernel weight (g) determined by 'Digital Image Analysis' (DIA) – calculated based on the number of kernels within 10 g samples from each plot used in the DIA. The results were converted and the weight of 1000 kernels was determined (Doehlert et al., 2004).
- Kernel area (mm^2) – determined by the DIA. Using the same procedure as kernel length and width. The Aphelion program determined the image area which was the kernel area. A 10 g samples from each plot was used to determine the kernel area (Doehlert et al., 2004).
- Kernel width (mm) – measured using Single Kernel Characterization System (SKCS) 4100 instrument (Perten Instruments, Springfield, IL). Kernel width is measured as the size of the gap formed between the crescent and the rotor at the engagement of the seed.

- 1000-kernel weight (g) – measured using the SKCS 4100 instrument (Perten Instruments, Springfield, IL). Kernel weight is measured as the electrical force required to return the boat into which the individual seeds are dropped to its original horizontal position. This force is proportional to the mass of the seed. The instrument provides kernel weight based on 1000-kernel weight.

Data analysis

Data were subjected to the analyses of variance (ANOVA), using the MIXED procedure of the Statistical Analyses System (SAS Institute, 2004). Within the mixed model, the RIL, their parents, and checks were considered fixed effects. Environments and blocks were considered random effects. The ANOVA was conducted within and across environments. Environments were considered homogeneous when the ratio of the effective error variances for each trait was less than 10-fold (Appendix Table 1) (Hartley, 1950; Patterson and Silvey, 1980; Geng et al., 1982; Mitchell, 1995). Data were subjected to combined analysis if environments were homogeneous. Tables 2 and 3 show the pertinent sources of variation, degrees of freedom, and the appropriate expected mean squares.

Table 2. Single analyses of variance for a lattice design, with treatment as fixed effects, and rep as random effects

SOV	DF [a]	MEAN SQUARE		F-test
		Obs	Expected	
Rep	$r - 1$	M1		non-valid
Blocks within rep	$r(k - 1)$	M2		non-valid
Treatment	$(t - 1)$	M3	$\sigma^2 + r\Sigma\alpha_i^2 / (t-1)$	M3 / M4
Error	$(k -1)(rk - k - 1)$	M4	σ^2	
Total	$rk^2 - 1$			

[a] = The letters k, t, and r refer to the size of the lattice, the number of treatments, and the number of replications, respectively.

Mean separation test was performed using an F-protected least significant difference (LSD) value at $P \leq 0.05$ level of significance for each evaluated trait. Correlations between kernel traits and the different agronomic and quality traits were calculated for single environments using the CORR procedure of SAS (2004). Homogeneity of these correlations was tested, and results were pooled if homogeneous. Otherwise, correlations from single environments were kept. The homogeneity test of the correlations was determined using the χ^2.

Table 3. Combined analyses of variance for a lattice design, across two years and two locations, with treatment as fixed effects, and rep and environment (year x location) as random effects

SOV	DF [a]	MEAN SQUARE		F-test
		Obs	Expected	
Environment	e – 1	M1		non-valid
Treatment	t – 1	M2	$\sigma^2 + r\sigma^2_{et} + re\Sigma\alpha_i^2/(t-1)$	M2 / M5
Rep within Env	e (r - 1)	M3		non-valid
Block within Rep x Env	er (k - 1)	M4		non-valid
Env x Treat	(e - 1) (t - 1)	M5	$\sigma^2 + r\sigma^2_{et}$	M5 / M6
Error	e(k -1) (rk - k - 1)	M6	σ^2	
Total	ert – 1			

[a] = The letters k, t, e, and r refer to the size of the lattice, the number of treatments, the number of environments, and the number of replications per environment, respectively.

RESULTS AND DISCUSSION

Environment effects and data analysis

The ANOVA for each environment was performed. Error variances were homogeneous for all the traits among the four environments except for lodging. Consequently, a combined analysis across the four environments was performed for all the traits except lodging. Homogeneity test was verified using the 10-fold (Appendix 1).

The analysis of variance summaries for the agronomic, quality and kernel traits are reported in Table 4, while the means of the experiment, parents, RIL, and checks for the combined and single environments are shown in Table 5.

Significant variance was observed among the genotypes for all the agronomic, quality, and kernel traits. Also, environment had a significant effect on all traits. Consequently, a significant treatment x environment interaction occurred for all the traits (Table 4). The agronomic and kernel traits, KVW, and flour extraction levels were higher, in general, at Carrington compared to Prosper. Within each location, except for flour extraction that showed similar results at Carrington 2009 and 2010, these traits levels were also higher in 2009 compared to 2010. Grain protein content (GPC) had the highest values at Prosper in 2010. Prosper environments especially in 2010 went through a strong hail storm and wind significantly decreasing several traits including wheat grain yield, kernel traits values, KVW, and flour extraction. Climatic conditions at Carrington were, in general, more favorable than at Prosper for spring wheat production in both 2009 and 2010. The hardest kernels were obtained at Carrington 2009 (Table 5). The adapted checks (Alsen, Steele-ND, Glenn, Faller, Barlow, Briggs, and Alpine) showed, in general, superior performance compared to the RIL and their parents (Table 5).

Table 4. Source of variation, degrees of freedom, mean square, and level of significant of the agronomic, quality, and kernel traits of the RIL, their parents, and checks grown at Carrington and Prosper, ND, during 2009 and 2010

Source	df [a]	GY [b]	df [a]	NS [b]	TKW-SKCS [b]	KW-SKCS [b]	KH [b]
Env	3	127917748**	3	2563379**	1519.3**	2.195**	3450.4*
Treatmnt	168	1574369**	168	26102**	25.1**	0.058**	220.9**
Env*Treat	504	430667**	504	10028*	3.6**	0.007**	22.9**
Error	571	127547	572	8262	1.2	0.002	4.7
CV (%)		11.7		15.9	3.8	1.7	3.1

Source	df [a]	TKW-DIA [b]	NK [b]	KVW [b]	GPC [b]	FE [b]	df [a]	KL [b]	KW-DIA [b]	KL/KW-DIA [b]
Env	3	3920.3**	10684**	483247**	99.21**	3932.3**	3	3.147**	4.125**	1.069**
Treatmnt	168	52.7**	69.1**	3632.1**	5.01**	70.2**	168	0.372**	0.088**	0.103**
Env*Treat	504	7.5**	31.1**	1005.3**	0.80**	13.7**	504	0.022**	0.011**	0.006**
Error	573	2.2	13.0	116.2	0.28	4.0	574	0.005	0.004	0.001
CV (%)		5.2	10.6	1.4	3.5	3.3		1.3	2.2	1.7

Source	df [a]	KA [b]	PH [b]	DH [b]	df [a]	Mat [b]	df [a]	SC [b]	Df [a]	Eme [b c]
Env	3	162.27**	4496.2**	11813**	2	18442**	1	131.37**	---	---
Treatmnt	168	3.49**	438.5**	96.06**	168	72.8**	168	2.54**	168	76.4**
Env*Treat	504	0.48**	41.0**	12.54**	336	17.5**	168	0.99**	---	---
Error	574	0.14	15.1	1.07	430	3.6	288	0.25	144	2.4
CV (%)		3.2	4.3	1.9		2.1		16.4		1.5

[a] df=degrees of freedom.

[b] GY=grain yield, NS=number of spikes m^{-2}, TKW-SKCS=thousand kernel weight by SKCS, KW-SKCS=kernel width by SKCS, KH=kernel hardness, TKW-DIA=thousand kernel weight by DIA, NK=number of kernel $spike^{-1}$, KVW=kernel volume weight, GPC=grain protein content, FE=flour extraction, KL=kernel length, KW-DIA=kernel width by DIA, KL/KW-DIA=kernel length width by DIA ratio, and KA=kernel area, PH=plant height, DH=days to heading, Mat=maturity, SC=spike color, and Eme=emergence.

[c] ANOVA for one location (Prosper 2010).

* and ** Significant at the 0.05 and 0.01 probability levels, respectively.

Table 5. Experiment, parents (ND 705 and PI 414566), and RIL means, their ranges (min and max), and checks means for agronomic, quality, and kernel traits at Carrington and Prosper, ND in 2009 and 2010

Traits [a]	Env	Exp	Parental lines		RIL population (n=160)			Checks	$LSD_{0.05}$
		mean	ND 705	PI 414566	Mean	Min	Max	mean	
GY (kg ha^{-1})	All	3042	3581	615	3002	1835	4112	4236	645
	Carr 09	3617	4021	568	3590	2586	4626	4622	572
	Carr 10	3405	3924	1373	3363	2014	4719	4568	699
	Pros 09	2919	3399	34	2888	1241	4681	3968	898
	Pros 10	2225	2851	195	2163	87	3885	3834	610
NS (spikes m^{-2})	All	572	652	285	569	449	709	682	98
	Carr 09	447	474	145	444	319	594	549	121
	Carr 10	614	717	529	609	468	891	716	172
	Pros 09	645	642	294	642	303	1000	769	221
	Pros 10	584	755	74	581	379	856	689	190
NK (Kernels spike^{-1})	All	34	30	27	34	27	42	32	5.5
	Carr 09	40	34	24	40	28	52	39	5.9
	Carr 10	37	31	31	37	21	47	34	5.8
	Pros 09	31	29	15	31	19	43	28	9.1
	Pros 10	28	26	36	28	4	40	29	7.2
TKW-DIA (g)	All	28.4	30.1	16.0	28.4	22.4	35.1	31.2	2.7
	Carr 09	31.5	32.4	17.8	31.5	24.3	38.5	33.4	1.9
	Carr 10	30.0	31.4	22.1	29.9	23.2	36.7	32.8	3.0
	Pros 09	28.6	30.4	11.6	28.6	22.5	36.5	30.4	3.9
	Pros 10	23.6	25.7	11.4	23.5	10.5	37.6	28.1	2.5
PH (cm)	All	91	97	71	91	64	107	86	6.3
	Carr 09	87	97	67	87	61	111	82	7.5
	Carr 10	94	102	88	94	66	110	90	6.3
	Pros 09	94	92	59	95	63	114	87	9.1
	Pros 10	89	95	70	89	67	109	86	7.6
DH (days)	All	56	55	76	55	49	65	55	3.5
	Carr 09	56	57	82	56	51	62	57	1.8
	Carr 10	63	65	74	63	59	69	64	1.7
	Pros 09	52	51	81	52	43	65	50	2.5
	Pros 10	50	47	68	51	41	69	47	2.1
Lod (%)	All	-	-	-	-	-	-	-	-
	Carr 09	0	0	0	0	0	0	0	-
	Carr 10	1	1	1	1	0	38	0	7
	Pros 09	28	25	0	28	0	97	44	53
	Pros 10	49	7	62	50	0	100	9	29
Mat (days)	All	92	91	105	92	83	103	91	4.7
	Carr 09	-	-	-	-	-	-	-	-
	Carr 10	100	103	109	100	91	104	100	3.4
	Pros 09	89	86	113	88	79	106	88	5.1
	Pros 10	86	83	95	86	78	98	85	2.3

Table 5. (Continued)

Traits [a]	Env	Exp	Parents		RIL			Checks	$LSD_{0.05}$
		Mean	ND 705	PI 414566	Mean	Min	Max	mean	
	All	-	-	-	-	-	-	-	-
Eme	Carr 09	-	-	-	-	-	-	-	-
(%)	Carr 10	-	-	-	-	-	-	-	-
	Pros 09	-	-	-	-	-	-	-	-
	Pros 10	99	100	18	100	90	100	100	3.0
	All	3.1	1.5	4.5	3.1	1.4	4.5	2.0	1.4
SC	Carr 09	-	-	-	-	-	-	-	-
(1-5)	Carr 10	3.5	1.5	5.0	3.6	1.0	5.0	2.1	-
	Pros 09	-	-	-	-	-	-	-	1.0
	Pros 10	2.6	1.5	4.1	2.6	1.9	4.1	1.9	1.0
	All	14.9	15.6	15.1	14.9	13.2	17.7	15.3	0.9
GPC	Carr 09	14.8	15.0	17.5	14.7	12.5	17.6	15.4	0.6
(%)	Carr 10	15.1	17.3	13.7	15.0	12.8	18.5	15.5	0.7
	Pros 09	14.3	14.4	14.3	14.3	11.5	16.9	14.4	1.7
	Pros 10	15.6	15.9	15.2	15.6	13.5	17.8	15.6	0.7
	All	749	791	598	749	669	790	774	31
KVW	Carr 09	787	820	619	787	723	816	798	10
($kg\ m^{-3}$)	Carr 10	761	796	673	761	708	802	777	13
	Pros 09	751	796	577	751	685	786	764	24
	Pros 10	697	751	522	695	486	782	759	31
	All	70	76	63	70	57	86	77	4.7
KH	Carr 09	74	75	49	74	59	88	79	3.3
(HI)	Carr 10	67	73	62	67	42	84	74	4.0
	Pros 09	69	78	74	68	45	87	75	5.5
	Pros 10	72	81	72	71	47	89	79	4.1
	All	61	66	54	61	46	67	63	3.6
FE	Carr 09	63	68	48	64	46	70	63	2.3
(%)	Carr 10	64	67	60	64	46	71	65	3.1
	Pros 09	59	65	41	59	32	66	61	4.3
	Pros 10	57	64	65	57	38	70	62	5.4
	All	5.72	5.47	5.61	5.72	5.15	6.48	5.67	0.15
KL	Carr 09	5.82	5.59	5.67	5.82	5.20	6.59	5.87	0.11
(mm)	Carr 10	5.72	5.44	6.02	5.72	5.14	6.44	5.68	0.20
	Pros 09	5.73	5.47	5.40	5.74	5.18	6.48	5.69	0.14
	Pros 10	5.59	5.35	5.32	5.60	5.02	6.55	5.45	0.13
	All	2.74	2.84	2.29	2.74	2.46	2.99	2.87	0.10
KW-	Carr 09	2.84	2.94	2.41	2.84	2.62	3.10	2.94	0.08
DIA	Carr 10	2.80	2.86	2.53	2.80	2.50	3.02	2.93	0.09
(mm)	Pros 09	2.72	2.83	2.11	2.72	2.40	3.01	2.83	0.17
	Pros 10	2.59	2.70	2.09	2.59	2.00	2.94	2.8	0.11

Table 5. (Continued)

Traits[a]	Env	Exp	Parents		RIL			Checks	$LSD_{0.05}$
		Mean	ND 705	PI 414566	Mean	Min	Max	mean	
KL/KW-DIA	All	2.09	1.93	2.46	2.10	1.84	2.48	1.98	0.08
	Carr 09	2.05	1.90	2.35	2.05	1.85	2.32	2.00	0.04
	Carr 10	2.05	1.90	2.38	2.05	1.80	2.45	1.94	0.05
	Pros 09	2.11	1.93	2.56	2.12	1.85	2.58	2.02	0.10
	Pros 10	2.16	1.98	2.55	2.17	1.81	2.64	1.95	0.07
KA (mm^2)	All	11.76	11.58	9.53	11.76	9.96	13.81	12.24	0.68
	Carr 09	12.47	12.28	10.14	12.46	10.30	14.56	12.92	0.57
	Carr 10	12.07	11.66	11.42	12.06	9.88	14.01	12.49	0.76
	Pros 09	11.67	11.49	8.32	11.68	9.97	14.09	12.08	0.96
	Pros 10	10.84	10.73	8.22	10.83	7.75	14.53	11.48	0.66
TKW-SKCS (g)	All	29.13	30.08	23.31	29.07	25.01	34.20	31.14	1.85
	Carr 09	30.79	32.36	25.61	30.71	26.39	35.36	32.91	1.69
	Carr 10	29.84	31.45	25.26	29.77	24.74	34.67	31.85	2.16
	Pros 09	29.90	30.11	20.38	29.90	25.26	35.73	31.12	2.57
	Pros 10	26.01	26.47	22.00	25.91	17.36	37.37	28.66	2.19
KW-SKCS (mm)	All	2.65	2.77	2.34	2.64	2.44	2.87	2.79	0.08
	Carr 09	2.69	2.85	2.36	2.69	2.48	2.93	2.84	0.06
	Carr 10	2.70	2.86	2.38	2.70	2.48	2.93	2.86	0.08
	Pros 09	2.67	2.73	2.31	2.67	2.47	2.93	2.78	0.11
	Pros 10	2.53	2.65	2.32	2.52	2.10	2.92	2.70	0.09

[a] GY=grain yield, NS=number of spikes m^{-2}, NK=number of kernels $spike^{-1}$, TKW-DIA=thousand kernel weight by DIA, PH=plant height, DH=days to heading, Lod=lodging, Mat=maturity, Eme=emergence, SC=spike color, GPC=grain protein content, KVW=kernel volume weight, KH=kernel hardness, FE=flour extraction, KL=kernel length, KW-DIA=kernel width by DIA, KL/KW-DIA=kernel length width by DIA ratio, KA=kernel area, TKW-SKCS=thousand kernel weight by SKCS, and KW-SKCS=kernel width by SKCS.

- Traits were not collected at these locations.

Agronomic and quality traits

The agronomic and quality data used in this paper are reported in Table 5. However, the analysis and discussion of these data are reported only in paper 1, in order to avoid duplication.

Kernel traits

Based on previous works cited in the literature review, there are different methods to measure kernel traits (theoretical geometric model, kernel size distribution, manual measurements, single kernel characterization system (SKCS), and digital image analysis (DIA)). In this study, kernel width and kernel weight (represented by 1000-kernel weight in

this work) were measured using both SKCS and DIA. The remaining kernel traits (kernel length and area) were measured by DIA.

The two kernel traits measured by SKCS and DIA were highly correlated. The correlation for kernel weight evaluated by the two methods was 0.93 and for kernel width was 0.85. Although, these two methods showed similar results, SKCS demonstrated problems with samples having small kernel size by either over-estimating their weight or size. SKCS uses 300 kernels samples, but the small kernels included in the sample are usually discarded. PI 414566 for instance, had very small kernels and according to SKCS method, the 1000-kernel weight mean of this parent was 16.0 g while in reality it was 23.3 g using the DIA method. The overall mean of kernel width for PI 414566 was 2.34 and 2.29 mm using SKCS and DIA, respectively. At Prosper, the differences between the values of these two methodologies were even larger because of the small kernel size of PI 414566 at this location (Table 5). Due to the lack of accuracy of the SKCS method, particularly for samples with small kernel size, only data by DIA is reported in this study.

Kernel length

The overall mean for kernel length was 5.72 mm. ND 705, PI 414566, and the RIL means were 5.47, 5.61, and 5.72 mm, respectively. The range of the RIL was from 5.15 to 6.48 mm (Table 5). Kernel length showed a continuous variation, suggesting a polygenic inheritance (Fatokun et al., 1992; Tsilo et al., 2010). Transgressive segregation occurred in both directions (Figure 1). Kernels were longer, in general, in 2009 than in 2010. Specifically at Carrington, kernels were longer than those harvested at Prosper. In 2010 at Carrington, PI 414566 completed its entire grown cycle resulting in a high value for kernel length of 6.02 mm (Table 5).

Kernel length was significantly correlated with all the other kernel traits except kernel width. The correlations between kernel length and length/width ratio, area, and 1000-kernel weight were 0.61, 0.69, and 0.42, respectively (Table 6). These correlations showed that kernel characteristics are inter-related and compensate for each other during the kernel formation and development. In fact, previous studies have also reported similar results. Dholakia et al. (2003), and Sun et al. (2009) found significant correlation between kernel length and 1000-kernel weight while Bergman et al. (2000) and Breseghello and Sorrells (2007) found significant correlation between kernel length and kernel area and weight.

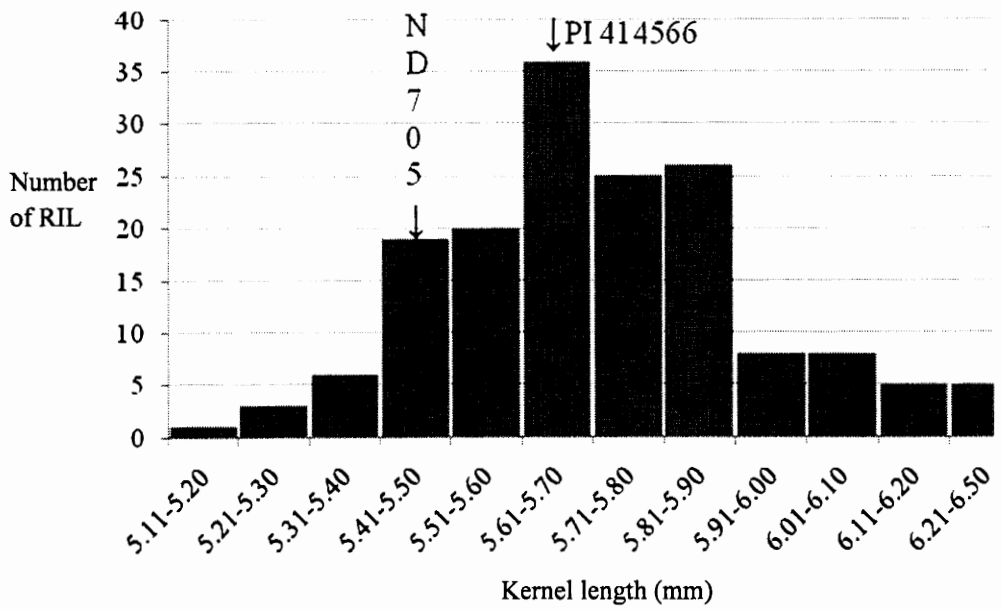

Figure 1. Frequency distribution of 160 RIL and their parents for kernel length means over two locations (Carrington and Prosper), and two years (2009 and 2010) (Parental means are marked with arrows).

Kernel width

Kernel width averaged 2.74 mm across environments. The means of ND 705, PI 414566, and RIL were 2.84, 2.29, and 2.74 mm, respectively. The range of the RIL for

kernel width was from 2.46 to 2.99 mm (Table 5) showing a positive transgressive segregation with values higher than ND 705 for this trait (Figure 2). Transgressive segregation gives a chance to the breeders to improve this trait. Kernel width had a non-discrete variation; consequently, this trait is influenced by many genes (Fatokun et al., 1992; Tsilo et al., 2010). The RIL and their parents had larger values for kernel width in Carrington than Prosper. Within each location, the kernel width, in general, was superior in 2009 than 2010, except for PI 414566 at Carrington 2010 where this parent fully completes its cycle reaching its normal diameter in this environment (Table 5).

Table 6. Correlation coefficients (r) among the kernel traits and their significance for the RIL, their parents, and checks grown in four environments (Carrington and Prosper in 2009 and 2010). The r values for single environment are denoted in the table

	Environment	Kernel width	Kernel length/ width ratio	Kernel area	1000 kernel weight
Kernel length	Carr 09 Carr 10 Pros 09 Pros 10	0.13	0.61**	0.69**	0.42**
Kernel width	Carr 09 Carr 10 Pros 09 Pros 10		-0.70**	0.80**	0.88** 0.93** [a]
Kernel length/ width ratio	Carr 09 Carr 10 Pros 09 Pros 10			-0.004 -0.27**	-0.35** -0.57** [a]
Kernel area	Carr 09 Carr 10 Pros 09 Pros 10				0.91**

* and ** Significant at the 0.05 and 0.01 probability levels, respectively.
[a] Correlation coefficients for a single environment.

Kernel width was significantly correlated with kernel length/width ratio (-0.70) and kernel area (0.80). Similarly, kernel width was also significantly associated with 1000-kernel weight with correlation coefficient values of 0.88 at Carrington 2009 and 2010 and

Prosper 2009; and 0.93 at Prosper 2010 (Table 6). The high correlation among these traits may provide an opportunity for the breeders to select indirectly to improve kernel characteristics. The 1000-kernel weight for instance is used frequently in breeding programs, it is an easy trait to measure, and may be a good trait for indirect selection for kernel width. Our results are in agreement with Bergman et al. (2000), Dholakia et al. (2003), Sun et al. (2009), and Tsilo et al. (2010) who reported high correlation between kernel width and kernel weight. Based on our results and previous studies, kernel width was more indicative than kernel length due to its high correlation with kernel weight (Bergman et al., 2000) and kernel area (Bergman et al., 2000; Sun et al., 2009).

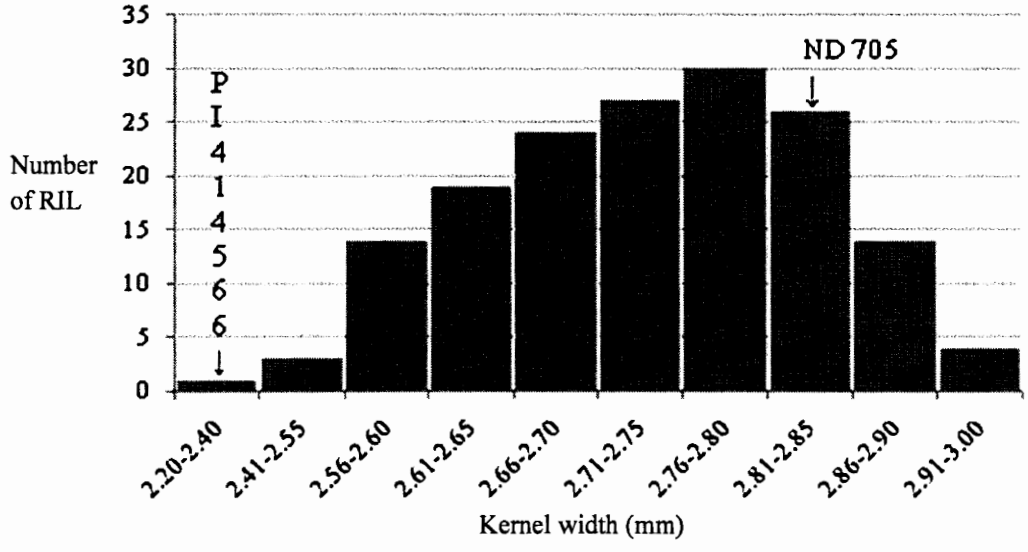

Figure 2. Frequency distribution of 160 RIL and their parents for kernel width means over two locations (Carrington and Prosper), and two years (2009 and 2010) (Parental means are marked with arrows).

Kernel length/width ratio

This trait describes more the shape of kernel. Across environments, the overall mean for kernel length/width ratio was 2.09. The means of this ratio for ND 705, PI 414566, and RIL were 1.93, 2.46, and 2.10, respectively. The RIL ranged from 1.84 to 2.48

(Table 5). This trait variation was continuous, suggesting that the inheritance of it is complex (Fatokun et al., 1992; Tsilo et al., 2010). Transgressive segregation was observed in both directions (Figure 3) indicating a possibility to be used in selection for this and kernel related traits. Based on our results, it seems that low value of this trait means a kernel with a spheroid or round shape (short and plump) while a high value means a cylindrical shape (tall and thin).

The significant and negative correlation with kernel area (r=27 at Prosper 2009 and 2010) and 1000-kernel weight (r=35 at Carrington 2009 and 2010, and at Prosper 2009; and r=57 at Prosper 2010) (Table 6) confirms the fact that spheroid shape seems to have more influence on kernel area and 1000-kernel weight than cylindrical.

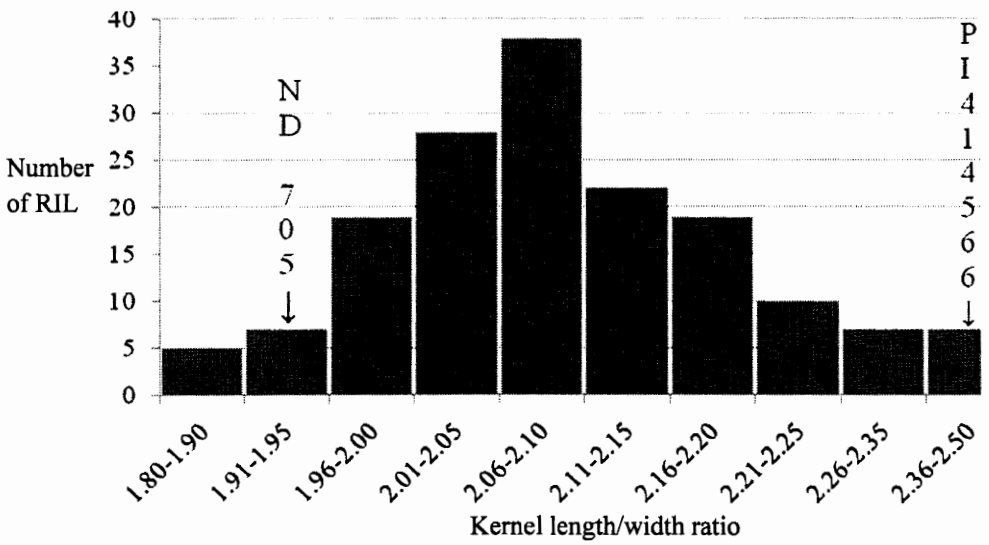

Figure 3. Frequency distribution of 160 RIL and their parents for kernel length/width ratio means over two locations (Carrington and Prosper), and two years (2009 and 2010) (Parental means are marked with arrows).

Kernel area

The kernel area average across all environments was 11.76 mm^2. ND 705, PI 414566, and RIL averages for this trait were 11.58, 9.53, and 11.76 mm^2, respectively. The

RIL mean varied from 9.96 to 13.81 mm^2 (Table 5). The variable for kernel area was continuous, suggesting that this trait is complex and of polygenic inheritance (Fatokun et al., 1992; Tsilo et al., 2010). Transgressive segregation was observed on one direction toward a superior value of the best parent ND 705 (Figure 4). Due to the favorable conditions at Carrington, kernels were able to grow better at this location compared to Prosper. Within locations, the year of 2009 was more favorable for the expression of this trait than in 2010, except for PI 414566.

Kernel area was highly correlated with 1000-kernel weight (0.91) (Table 6). Previous studies by Bergman et al. (2000) and Breseghello and Sorrels (2007) also reported high correlation between these two traits. This correlation is a strong indicator for breeders to use the 1000-kernel weight to select indirectly for improved kernel area.

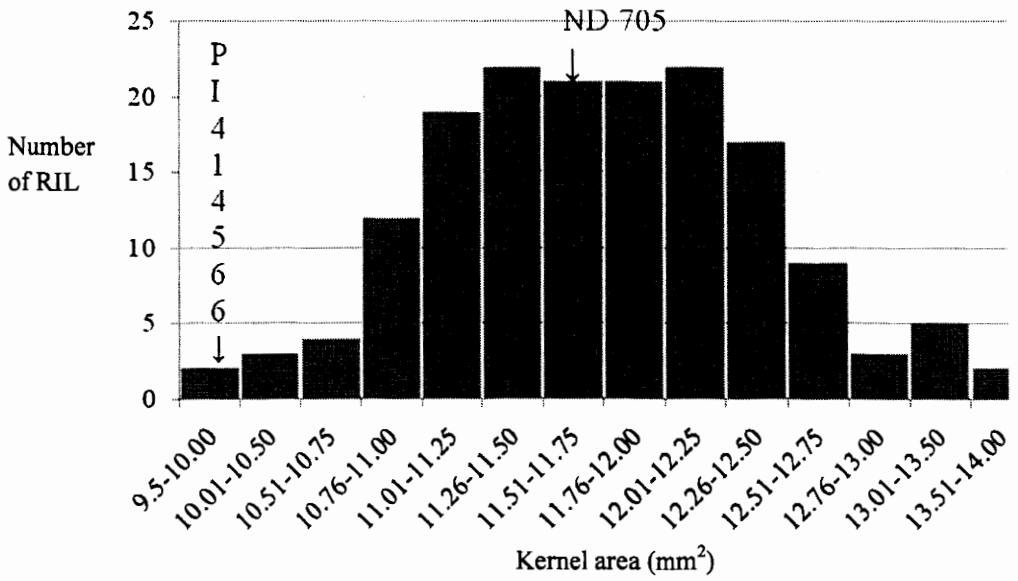

Figure 4. Frequency distribution of 160 RIL and their parents for kernel area means over two locations (Carrington and Prosper), and two years (2009 and 2010) (Parental means are marked with arrows).

Kernel weight

The results of this study (Table 5) indicate that the mean for 1000-kernel weight across environments was 28.4 g while ND 705, PI 414566 and RIL means were 30.1, 16.0, and 28.4 g, respectively. The RIL ranged from 22.4 to 35.1 g. Kernel weight wa, in general, higher at Carrington in 2009 followed by Carrington in 2010. Within the years of 2009 and 2010, Carrington had better environment conditions for wheat development compared to Prosper favoring kernel weight development. Kernel weight was highly associated with all the other kernel traits (Table 6). The kernel weight trait followed a continuous variation, suggesting that the inheritance of this trait is quantitative involving many genes (Fatokun et al., 1992; Tsilo et al., 2010). As for previous traits, transgressive segregation was observed for this characteristic as well (Figure 5) and it is what the breeders usually look for. It will allow them to select for improved kernel weight even when non-adapted parent are used in the cross. Kernel weight is largely influenced by environment conditions such as rainfall, temperature, and soil fertility (Daniel and Triboi, 2000).

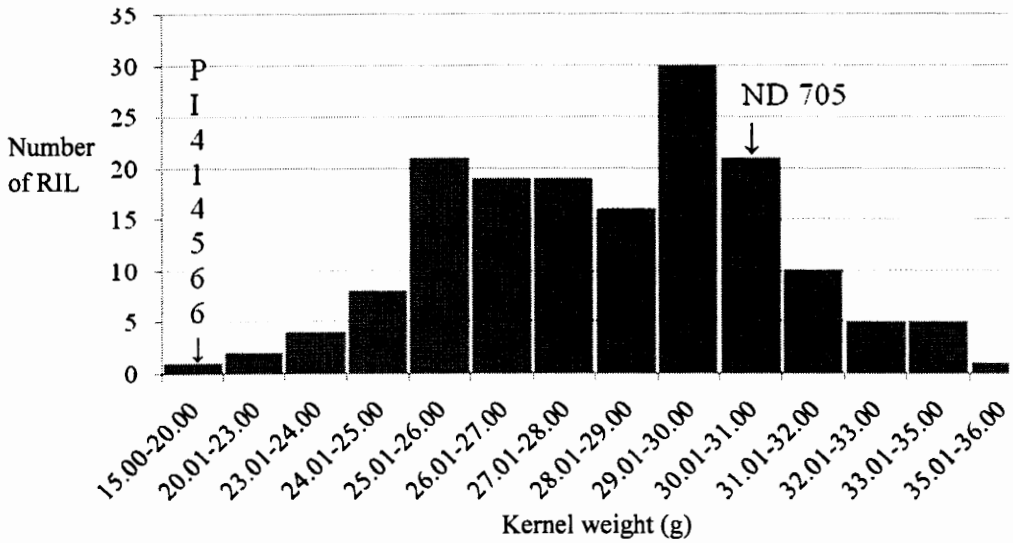

Figure 5. Frequency distribution of 160 RIL and their parents for kernel weight means over two locations (Carrington and Prosper), and two years (2009 and 2010) (Parental means are marked with arrows).

Associations among agronomic and kernel traits

The grain yield mean of the RIL was lower than the adapted checks. This RIL population was developed to generate high divergence for kernel traits for mapping purposes. However, it also had high variation for agronomic traits.

Grain yield was highly associated with kernel traits. Grain yield was positively correlated with kernel width (0.26 at Carrington, and 0.65 at Prosper) and negatively correlated with kernel length/width ratio (0.29 at Carrington and Prosper 2009, and 0.51 at Prosper 2010) (Table 7). Overall, the top yielding genotypes in this study including the checks Faller, Steele-ND, and Barlow along with the RIL 143 had significantly higher values for kernel width. These results, seems to indicate that higher grain yield is associated more to kernels with spheroid shape. Also, the high correlations between grain yield and kernel area, and 1000-kernel weight (Table 7) suggest that these spheroid kernels should be large and/or heavy as well. The correlation between grain yield and kernel traits were higher at Prosper compared to Carrington. This higher correlation at Prosper suggests that under unfavorable conditions, the top grain yield genotypes, in addition to have better agronomic performance (low lodging, low kernel shattering, and diseases resistance), tended to have increased kernel width than kernel length.

Lodging was not a major issue except at Prosper 2010. Therefore, lodging was not correlated with kernel traits, except at Prosper 2010. At Prosper 2010, the significant correlation between lodging and kernel width, length/width ratio, area, and weight were -0.53, 0.45, -0.42, and -0.54, respectively (Table 7). These associations between lodging and kernels traits at Prosper in 2010 were similar to the results obtained for grain yield. The lodging resistant lines, in general, produced larger, heavier, and more spheroid

Table 7. Correlation coefficients (r) among the agronomic and kernel traits, and their significance for the RIL, their parents, and checks grown in four environments (Carrington and Prosper in 2009 and 2010). The r values for single environment are denoted in the table

	Env	Kernel length	Kernel width	Kernel length/width ratio	Kernel area	1000-kernel weight
Grain yield	Carr 10	-0.19* [a]	0.26**	-0.29**	-0.02 [a]	0.27**
	Carr 09				0.29** [a]	
	Pros 09	0.11	0.65**		0.54**	0.54** [a]
	Pros 10			-0.51** [a]		0.75** [a]
Lodging	Carr 09	-	-	-	-	-
	Carr 10	0.01	-0.07	0.06	-0.03	-0.05
	Pros 09		-0.53** [a]	0.45** [a]	-0.42** [a]	-0.54** [a]
	Pros 10					
Number of spikes per m^2	Carr 09	-0.11	-0.07	-0.10	-0.15	-0.07
	Carr 10					
	Pros 09		0.23** [a]		0.17* [a]	0.27** [a]
	Pros 10					
Plant height	Carr 09	0.05	0.07	-0.01	0.19*	0.16*
	Carr 10					
	Pros 10				-0.16* [a]	
	Pros 09					
Days to heading	Carr 09	-0.21**	-0.29** [a]	0.20*	-0.28** [a]	-0.40** [a]
	Carr 10		-0.16* [a]		-0.18* [a]	-0.19* [a]
	Pros 09		-0.56**		-0.62**	-0.63**
	Pros 10					
Number of kernels per spike	Carr 09	-0.07	-0.20*	0.05	-0.23**	-0.26**
	Carr 10					
	Pros 09		0.25** [a]	-0.34** [a]	0.08 [a]	0.15 [a]
	Pros 10		0.41** [a]	-0.31** [a]	0.35** [a]	0.40** [a]
Maturity	Carr 09	---	---	---	---	---
	Carr 10	-0.03 [a]	0.004 [a]	0.10	0.005 [a]	0.04 [a]
	Pros 09	-0.32**	-0.47**		-0.58**	-0.54**
	Pros 10					
Spike color	Carr 09	---	---	---	---	---
	Pros 09	---	---	---	---	---
	Carr 10	0.03	-0.24**	0.20*	-0.16*	-0.26**
	Pros 10					

* and ** Significant at the 0.05 and 0.01 probability levels, respectively.
- No lodging at this location.
--- Traits were not collected at these locations.
[a] Correlation coefficients for a single environment.

type of kernels than those lines that lodged. Similar results were obtained for number of spikes per m^2 under the unfavorable environment of Prosper 2010. At this location, the genotypes with high number of spikes per m^2 tended to be associated with plump, large, and heavy kernels. Also, high number of kernels per spike as well as white spike color was significant correlated with large, heavy, and spheroid kernels. Plant height was significantly associated only with kernel area and weight. Tsilo et al., (2010) did not find significant correlation between plant height and kernel width, and weight.

The late lines tended to be associated with small (length and width) and light kernels. Tsilo et al. (2010) found similar results between days to heading and kernel traits. They suggested that late lines have less capability for grain filling compared to early lines.

Associations among quality and kernel traits

The highest values for grain protein content (GPC) were at Prosper in 2010 where climatic conditions were unfavorable. It seems that most genotypes have allocated nitrogen into protein development instead of grain yield. Kernel volume weight (KVW), kernel hardness, and flour extraction reached their highest values at Carrington in 2009 (Table 5).

GPC, in general, was not associated with kernel traits (Table 8). Compared to other kernel characteristics such as kernel size and/or shape, GPC is more affected by the environmental conditions, such as temperature, soil fertility, and rainfall (Daniel and Triboi, 2000), and certainly cultivar selection. However, some previous works showed significant correlations between GPC and kernel weight, width (Ohm et al., 1998; Tsilo et al., 2010), and length (Breseghello and Sorrells, 2007). These associations between GPC and kernel traits were attributed to a higher proportion of seed coat to the endosperm in

larger kernels by Marshall et al. (1984) or to a higher proportion of embryo size to endosperm as Breseghello and Sorrells (2007) proposed from their study.

KVW was highly correlated with all the kernel traits. It was negatively correlated with kernel length in all environments (0.26) except at Prosper 2010, whereas the correlation between KVW and kernel width was positive and very high (0.37 at Carrington, 0.63 at Prosper 2009, and 0.75 at Prosper 2010). Under unfavorable environment conditions, correlation between these two traits tends to increase. The correlation between KVW and kernel length/width ratio was highly negative (0.58) (Table 8). Based on these results, KVW seems to be closely associated with spheroid type of kernels. The correlations between KVW and kernel area and 1000-kernel weight also increased according to the intensity of environmental stresses. The correlation between KVW and kernel area was 0.36 at Prosper 2009 and 0.62 at Prosper 2010. No significant correlation occurred between these two traits at Carrington. The correlation between KVW and 1000-kernel weight was 0.38 at Carrington, 0.61 at Prosper 2009, and 0.79 at Prosper 2010 (Table 8). These results indicate that the type of kernels with higher KVW tend to be short in length, plump, large in area, and heavy. This type of kernel size and shape enhances the packing ability which results in higher KVW. These desirable kernel characteristics become more necessary as the level of environmental stress increases. Under unfavorable conditions, the kernel development becomes more restricted due to the lack of essential nutrients, water, and/or adequate temperatures. The genotypes with better efficiency for nutrients and water uptake, utilization, and conversion into kernels with the desirable traits described above tend to have good quality traits including KVW.

Table 8. Correlation coefficients (r) among the quality and kernel traits, and their significance for the RIL, their parents, and checks grown in four environments (Carrington and Prosper in 2009 and 2010). The r values for single environment are denoted in the table

	Env	Grain protein content	Kernel volume weight	Kernel hardness	Flour extraction
Kernel length	Carr 09 Carr 10 Pros 09	-0.12	-0.26**	-0.30**	-0.002
	Pros 10		0.07 [a]		
Kernel width	Carr 10	0.31** [a]	0.37**	-0.22**	0.27**
	Carr 09 Pros 09	0.02	0.63** [a]	-0.39** [a]	
	Pros 10		0.75** [a]	-0.01 [a]	
Kernel length/ width ratio	Carr 09 Carr 10 Pros 09 Pros 10	-0.15	-0.58**	-0.04	-0.22**
Kernel area	Carr 09	-0.003	0.08	-0.26**	0.19*
	Carr 10 Pros 10		0.62** [a]		
	Pros 09		0.36** [a]	-0.54** [a]	
1000-kernel weight	Carr 10	0.37** [a]	0.38**	-0.18*	0.27**
	Carr 09 Pros 10	-0.03	0.79** [a]		
	Pros 09		0.61** [a]	-0.45** [a]	

* and ** Significant at the 0.05 and 0.01 probability levels, respectively.
[a] Correlation coefficients for a single environment.

Previous investigations on associations between KVW and kernel traits were in general conflicting. Troccoli and Di Fonzo (1999) found negative correlation between KVW and kernel length on durum wheat. The same authors reported that KVW was not significantly associated with kernel width, area, and weight. These researchers however, used small samples of only 25 kernels to measure the kernel traits. In other studies by Campbell et al. (1999) and Bergman et al. (2000), they used 78 RIL developed from a cross between a soft white (NY18) with hard white wheat (Clark's Cream) and found that KVW

was not associated with kernel length, width, and area. In these studies, kernel traits data were collected from 50 to 60 kernel samples. Lee et al. (2006) working with blended samples from two hard white wheat, 'Betty' and 'Trego', found however, significant positive association between KVW and kernel weight, and width. The kernel traits were measured in this study by the SKCS. Similarly, Ohm et al. (1998) also utilized SKCS for measuring the kernel traits and found significant association between KVW and kernel weight among 12 winter wheat cultivars. However, they did not find any association between KVW and kernel width. These inconsistent observations might have resulted from different growing conditions, wheat varieties/genotypes and classes, kernel traits evaluation methods, and kernel sample size.

The results on kernel hardness showed a negative association between this trait and kernel length (0.30), width (0.22 at Carrington, and 0.39 at Prosper 2009), area (0.26 at Carrington and Prosper 2010, and 0.54 at Prosper 2009), and 1000-kernel weight (0.18 at Carrington and Prosper 2010, and 0.45 at Prosper 2009) (Table 8). Previous study (Bergman et al., 2000) however, did not find any significant association between kernel hardness and kernel traits. Most of the investigations on kernel hardness indicate that this trait is affected mainly by genetic control (Pomeranz and Mattern, 1988), and probably in some cases by the interactions of proteins and starch granules (Greenwell and Schofield, 1986; Greenblatt et al., 1995).

Data from this study showed that flour extraction was significantly correlated with all the kernel traits including kernel width (0.27), length/width ratio (-0.22), area (0.19), and 1000-kernel weight (0.27), except kernel length (Table 8). Similar results were reported by Lee et al. (2006) who found high correlations between flour extraction and

kernel width, and weight. Breseghello and Sorrells (2007) reported the same QTL which resulted in an increase in kernel size and weight, also had a positive effect in flour extraction. Lyford et al. (2005) developed an equation to explain flour extraction in hard red winter wheat. In this study, kernel weight was one of the significant parameters influencing positively the flour extraction rate. Using a recurrent selection scheme for kernel size using 10 spring wheat genotypes, Wiersma et al. (2001) were able to increase flour extraction rate by 5%. Considering similar embryo size, larger kernels are supposed to have larger volume of endosperm, consequently, yielding in higher amount of flour extraction. Dai (2009) studied the changes of the main enzymes during grain filling period for large and small kernels. He found that enzymes related to starch synthase and accumulation, and ADP-glucose activities were significantly higher and more active in large kernels compared to small kernels. Plants with larger kernels maintained a longer period of kernel filling with greater activities of these enzymes during mid and late kernel filling stages generating a greater amount of starch. However, Breseghello and Sorrells (2006) using 95 soft winter wheat cultivars and samples of 24 kernels found significant association between flour extraction and only kernel weight at a single location. Other study (Bergman et al., 2000) did not find any significant association between flour extraction and kernel traits.

As it was reported for the KVW correlations with kernel traits, these inconsistent finding may be the result of different growing conditions where these researchers were situated, using different wheat varieties and belonging to different classes, different kernel traits evaluation method, and kernel sample sizes. However, flour extraction is a complex

trait and is not explained by one or few traits and will continue to be a great challenge for wheat breeders, millers, and industry.

CONCLUSION

The parents, ND 705 and PI 414566, showed drastic differences for all kernel traits. Consequently, the RIL showed significant variation for all kernel traits. All kernel traits demonstrated continuous distributions suggesting the polygenic inheritance of these traits. Also, transgressive segregation occurred for all the kernel traits providing an opportunity for breeders to improve these traits. The kernel traits showed in general high association among themselves.

The genotypes had better performance for agronomic and kernel traits, KVW, and flour extraction at Carrington compared to Prosper due to favorable environment conditions. Within locations, except for flour extraction that had similar results at Carrington in 2009 and 2010, these traits were much higher in 2009 than 2010.

Grain yield, KVW, and flour extraction were highly correlated with kernel traits. These important agronomic and quality traits were correlated with kernel width, length/width ratio, area, and 1000-kernel weight. These results indicate that higher values for grain yield, KVW, and flour extraction are related to kernels with spheroid or round shape (short and plump). Also, these associations suggested that these spheroid kernels should be large and heavy in order to have better performance for grain yield, KVW, and flour extraction. Kernel traits can partially contribute to increase grain yield, KVW, and flour extraction.

Based on our results, breeders should considerer large, heavy, and spheroid kernels when selecting parental genotypes for the development of populations for improved agronomic and quality attributes.

REFERENCES

AACC International. 2000. Approved Methods of the American Association of Cereal Chemists, 10th ed. Methods 46-30, 55-10, and 56-81B. The Association: Saint Paul, MN.

Bassett, L.M., R.E. Allan, and G.L. Rubenthaler. 1989. Genotype x environment interactions on soft white winter wheat quality. Agronomy Journal 81:955-960.

Bergman, C.J., D.G. Gualberto, K.G. Campbell, M.E. Sorrells, and P.L Finney. 2000. Kernel morphology variation in a population derived from a soft by hard wheat cross and associations with end-use quality traits. Journal of Food Quality 23:391-407.

Breseghello, F., and M.E. Sorrells. 2006. Association mapping of kernel size and milling quality in wheat (*Triticum aestivum* L.) cultivars. Genetics 172:1165-1177.

Breseghello, F., and M.E. Sorrells. 2007. QTL analysis of kernel size and shape in two hexaploid wheat mapping populations. Field Crops Research 101:172-179.

Campbell, K.G., C.J. Bergman, D.G. Gualberto, J.A. Anderson, M.J. Giroux, G. Hareland, R.G. Fulcher, M.E. Sorrells, and P.L. Finney. 1999. Quantitative trait loci associated with kernel traits in a soft x hard wheat cross. Crop Science 39:1184-1195.

Chastain, T.G., K.J. Ward, and D.J. Wysocki. 1995. Stand establishment responses of soft white winter wheat to seedbed residue and seed size. Crop Science 35:213-218.

Dai, Z. 2009. Activities of enzymes involved in starch synthesis in grains of large and small kernel type wheat (*Triticum aestivum* L.). Journal of Food, Agriculture & Environment 7:493-495.

Daniel, C., and E. Triboi. 2000. Effects of temperature and nitrogen nutrition on the grain composition of winter wheat: effects on gliadin content and composition. Journal of Cereal Science 32:45–56.

Devkota, R.N., J.C. Rudd, Y. Jin, K.D. Glover, R.G. Hall, and G.A. Hareland. 2007. Registration of 'Briggs' wheat. Crop Science 47:432-434.

Dholakia, B.B., J.S.S. Ammiraju, H. Singh, M.D. Lagu, M.S. Roder, V.S. Rao, H.S. Dhaliwal, P.K. Rankejar, and V.S. Gupta. 2003. Molecular marker analysis of kernel size and shape in bread wheat. Plant Breeding 122:392-395.

Doehlert, D.C., M.S. McMullen, J. Jannink, S. Panigrahi, H. Gu, and N.R. Riveland. 2004. Evaluation of oat kernel size uniformity. Crop Science 44:1178-1186.

Dornez, E., K. Gebruers, I.J. Joye, B. Ketelaere, J. Lenartz, C. Massaux, B. Bodson, J.A. Delcour, and C.M. Courtin. 2008. Effects of genotype, harvest year and genotype-by-harvest year interactions on arabinoxylan, endoxylanase activity and endoxylanase inhibitor levels in wheat kernels. Journal of Cereal Science 47:180-189.

Fatokun, C.A., D.I. Menancio-Hautea, D. Danesh, and N.D. Young. 1992. Evidence for orthologous seed weight genes in cowpea and mung bean based on RFLP mapping. Genetics 132:841-8246.

Frohberg, R.C., R.W. Stack, T. Olson, J.D. Miller, and M. Mergoum. 2006. Registration of 'Alsen'. Crop Science 46:2311-2312

Geng, S., P. Schneeman, and W.J. Wang. 1982. An empirical study of the robustness of analysis of variance procedures in the presence of commonly encountered data problems. American Journal of Enology and Viticulture 33:131-134.

Greenblatt, G.A., A.D. Bettge, and C.F. Morris. 1995. Relationship between endosperm texture and occurrence of friabilin and bound lipids on wheat starch. Cereal Chemistry 72:172-176.

Greenwell, P., and J.D. Schofield. 1986. A starch granule protein associated with endosperm softness in wheat. Cereal Chemistry 63:379-380.

Hartley, H.O. 1950. The maximum F-ratio as a short-cut test for heterogeneity of variances. Biometrika 37:308-312.

Lee, K.M., J.P. Shroyer, T.J. Herrman, and J. Lingenfelser. 2006. Blending hard white wheat to improve grain yield and end-use performances. Crop Science 46:1124-1129.

Lyford, C.P., W. Kidd, P. Rayas-Duarte, and C. Deyoe. 2005. Prediction of flour extraction rate in hard red winter wheat using the single kernel characterization. Journal of Food Quality 28:279-288.

Marshall, D.R., D.J. Mares, H.J. Moss, and F.W. Ellison. 1986. Effects of grain shape and size on milling yields in wheat. II. Experimental studies. Australian Journal of Agricultural Research 37:331-342.

Marshall, D.R., F.W. Ellison, and D.J. Mares. 1984. Effects of grain shape and size on milling yields in wheat. 1. Theoretical-analysis based on simple geometric-models. Australian Journal of Agricultural Research 35:619-630.

Mergoum, M., R.C. Frohberg, J.D. Miller, and R.W. Stack. 2005. Registration of 'Steele-ND' wheat. Crop Science 45:1163-1164.

Mergoum, M., R.C. Frohberg, R.W. Stack, J.W. Rasmussen, and T.L. Friesen. 2008. Registration of 'Faller' spring wheat. Journal of Plant Registrations 2:224-229.

Mergoum, M., R.C. Frohberg, R.W. Stack, T. Olson, T.L. Friesen, and J.B. Rasmussen. 2006. Registration of 'Glenn' wheat. Crop Science 46:473-474.

Mergoum, M., S. Simsek, R.C. Frohberg, J.B. Rasmussen, T.L. Friesen, and T. Adhikari. 2011. 'Barlow': A high-quality and high-yielding hard red spring wheat cultivar adapted to the North Central Plains of the USA. Journal of Plant Registrations 5:62-67.

Mitchell, M. 1995. Using XSLIP-STAT to explore new and unusual statistic paradigms. Perspective 19:28-32.

Nelson, J.C, C. Andreescu, F. Breseghello, P.L. Finney, D.G. Gualberto, C.J. Bergman, R.J. Peña, M.R. Perretant, P. Leroy, C.O. Qualset, and M.E. Sorrells. 2006. Quantitative trait locus analysis of wheat quality traits. Euphytica 149:145-159.

Ohm, J.B., O.K. Chung, and C.W. Deyoe. 1998. Single-kernel characteristics of hard winter wheats in relation to milling and baking quality. Cereal Chemistry 75:156-161.

Patterson, H.D., and V. Silvey. 1980. Statutory and recommended list trials of crop varieties in the United Kingdom. Journal of the Royal Statistical Society [Series A] 143:219-252.

Pomeranz, Y., and P.J. Mattern. 1988. Genotype and genotype x environmental interaction effects on hardness estimates in winter wheat. Cereal Food World 33:371-374.

SAS Institute. 2004. SAS Online Doc, version 9.1.2 SAS Institute, Cary, NC.

Sun, X.Y., K. Wu, Y. Zhao, F.M. Kong, G.Z. Han, H.M. Jiang, X.J. Huang, R.J. Li, H.G. Wang, and S.S. Li. 2009. QTL analysis of kernel shape and weight using recombinant inbred lines in wheat. Euphytica 165:615-624.

Symes, K.J. 1965. The inheritance of grain hardness in wheat as measured by the particle-size index. Australian Journal of Agricultural Research 16:113-123.

Troccoli, A., and N. Di Fonzo.1999. Relationship between kernel size features and test weight in Triticum durum. Cereal Chemistry 76:45-49.

Tsilo, T.J., G.A. Hareland, S. Simsek, S. Chao, and J.A. Anderson. 2010. Genome mapping of kernel characteristics in hard red spring wheat breeding lines. Theoretical and Applied Genetics 121:717-730.

USDA-NRCS. 2011. Web Soil Survey [online]. Available at http://websoilsurvey.nrcs.usda.gov/app/ (verified 23 February 2011). Natural Resource Conservation Service, Washington, DC.

Wiersma, J.J., R.H. Busch, G.G. Fulcher, and G. Hareland. 2001. Recurrent selection for kernel weight in spring wheat. Crop Science 41:999-1005.

Table A1. Coefficient of variance (CV%), error mean square (EMS), and ratio between EMS of environments for agronomic, quality, and kernel traits evaluated at Carrington and Prosper, ND, in 2009 and 2010

Trait	Carrington 2009		Prosper 2009		Carrington 2010		Prosper 2010		Ratio [b]
	CV%	EMS	CV%	EMS	CV%	EMS	CV%	EMS	
Grain yield	8.00	83,823	15.56	206,315	10.39	125,186	13.86	95,126	2.46
Spikes number per m^2	13.73	3759	17.35	12521	14.19	7592	16.45	9212	3.33
Kernels number per spike	7.41	8.87	14.97	21.27	7.93	8.65	12.99	13.17	2.46
Thousand kernel weight by DIA	3.10	0.95	6.91	3.91	5.04	2.28	5.41	1.63	4.11
Plant height	4.36	14.36	4.87	21.15	3.41	10.25	4.34	14.87	2.06
Days to heading	1.60	0.82	2.47	1.62	1.33	0.71	2.11	1.13	2.29
Lodging	0.00	0.00	94.33	715.60	317.12	11.66	29.84	210.05	> 10.00
Maturity [a]	-	-	2.90	6.58	1.70	2.88	1.35	1.35	4.87
Emergence [a]	-	-	-	-	-	-	1.55	2.36	-
Spike color [a]	-	-	-	-	14.02	0.24	19.49	0.26	1.08
Grain protein content	2.22	0.11	5.98	0.73	2.41	0.13	2.34	0.13	6.82
Kernel volume weight	0.66	26.65	1.65	153.25	0.88	44.66	2.22	239.97	9.01
Kernel hardness	2.22	2.72	4.09	7.86	3.02	4.11	2.88	4.25	2.90
Flour extraction	1.83	1.34	3.68	4.63	2.43	2.44	4.76	7.47	5.57
Kernel length	0.94	0.0030	1.22	0.0049	1.73	0.0098	1.15	0.0041	3.24
Kernel width	1.49	0.0018	3.17	0.0074	1.55	0.0019	2.10	0.0030	4.13
Kernel length / width ratio	1.05	0.0005	2.48	0.0028	1.36	0.0008	1.67	0.0013	5.93
Thousand kernel weight by DIA	3.10	0.95	6.91	3.91	5.04	2.28	5.41	1.63	4.11
Kernel width by SKCS	1.13	0.0009	2.12	0.0032	1.55	0.0018	1.78	0.0020	3.46
Thousand kernel weight by SKCS	2.79	0.74	4.34	1.69	3.67	1.20	4.26	1.23	2.29
Kernel area	2.31	0.0826	4.14	0.2338	3.16	0.1459	3.08	0.1115	2.83

[a] Traits were not evaluated in all the environments.

[b] Ratio: test of homogeneity (greatest EMS / smallest EMS) should be smaller than 10-fold.

- Traits were not collected at these locations.

GENERAL CONCLUSIONS

The adapted (ND 705) and non-adapted parent (PI 414566) showed drastic differences for all the kernel, agronomic, and quality traits. Consequently, the RIL population showed significant variation for all these traits as well. All kernel traits had continuous distribution suggesting that the inheritance of these traits is quantitative involving many genes. Transgressive segregation occurred for all the kernel, agronomic and quality traits except for spike color and kernel volume weight (KVW).

Overall, the genotypes had better performance for kernel and agronomic traits, KVW, and flour extraction at Carrington compared to Prosper due to favorable environment conditions at Carrington during these two years. Within locations, except for flour extraction that had similar results at Carrington 2009 and 2010, these traits had higher levels in 2009 than 2010.

The checks Faller, Barlow, and Steele-ND along with the RIL 143 and 88 had significant superior grain yield across environments. The RIL 143 and 88 showed, overall, good agronomic attributes including low rates of lodging and kernel shattering, demonstrating high potential to be included in the HRSW breeding program. Also, the RIL 47 had superior attributes for KVW, and RIL 82 and 141 had superior attributes for flour extraction. These RIL can be valuable for the HRSW breeding program as well.

Grain yield, KVW, and flour extraction were highly correlated with kernel traits. These important agronomic and quality traits were correlated with kernel width, length/width ratio, area, and 1000-kernel weight. These results indicate that higher values for grain yield, KVW, and flour extraction were related with spheroid or round shape (short and plump) kernels. Also, these associations suggested that these spheroid kernels should

be large and heavy in order to develop genotypes with higher grain yield, KVW, and flour extraction. Kernel traits can play a significant role to improve grain yield, KVW, and flour extraction.

The use of elite x elite crosses continues to be the most common and effective type of crosses to develop new cultivars in breeding programs. However, with the concern that narrow germplasm may restrict breeding flexibility and slow genetic progress to biotic and abiotic stresses, it seems that the development of populations originated from adapted x non-adapted germplasm is a common practice by breeders to broaden the genetic variability of their germplasm and consequently increase genetic gain. This study has showed clearly the value of using non-adapted sources of germplasm to improve several wheat economic traits.

Based on this study, breeders should exploit heavy, large, and spheroid kernels during the parental selection for the development of populations with improved agronomic and quality attributes.

GENERAL REFERENCES

Bergman, C.J., D.G. Gualberto, K.G. Campbell, M.E. Sorrells, and P.L Finney. 2000. Kernel morphology variation in a population derived from a soft by hard wheat cross and associations with end-use quality traits. Journal of Food Quality 23:391-407.

Breseghello, F., and M.E. Sorrells. 2006. Association mapping of kernel size and milling quality in wheat (*Triticum aestivum* L.) cultivars. Genetics 172:1165-1177.

Breseghello, F., and M.E. Sorrells. 2007. QTL analysis of kernel size and shape in two hexaploid wheat mapping populations. Field Crops Research 101:172-179.

Briggle, L.W., and B.C. Curtis. 1987. Wheat worldwide. 1-32. *In* Heyne E.G. (ed.). Wheat and wheat improvement. Agronomy No 13, 2nd ed. American Society of Agronomy, Madison, WI.

Campbell, K.G., C.J. Bergman, D.G. Gualberto, J.A. Anderson, M.J. Giroux, G. Hareland, R.G. Fulcher, M.E. Sorrells, and P.L. Finney. 1999. Quantitative trait loci associated with kernel traits in a soft x hard wheat cross. Crop Science 39:1184-1195.

Dholakia, B.B., J.S.S. Ammiraju, H. Singh, M.D. Lagu, M.S. Roder, V.S. Rao, H.S. Dhaliwal, P.K. Rankejar, and V.S. Gupta. 2003. Molecular marker analysis of kernel size and shape in bread wheat. Plant Breeding 122:392-395.

Eagles, H.A., and J.E. Lothrop. 1994. Highland maize from Central México – its origin, characteristics and use in breeding programs. Crop Science 34:11-19.

Gaines, C.S., P.L. Finney, and L. Andrews. 1997. Influence of kernel size and shriveling on soft wheat milling and baking quality. Cereal Chemistry 74:700-704.

Ghaderi, A., and E.H. Everson. 1971. Genotype-environment studies of test weight and its components in soft winter weight. Crop Science 11:617-619.

Giura, A., and N.N. Saulescu. 1996. Chromosomal location of genes controlling grain size in a large grained selection of wheat (*Triticum aestivum* L.) Euphytica 89:77-80.

Gouesnard, B., J. Sanou, A. Panouillé, V. Bourion, and A. Bovat. 1996. Evaluation of agronomic traits and analysis of exotic germ plasm polymorphism in adapted x exotic maize crosses. Theoretical and Applied Genetics 92:368-374.

Holley, R. N., and M. M. Goodman. 1988. Yield potential of tropical hybrid corn derivatives. Crop Science 28:213-217.

Horsfall, J.G., G.E. Brandow, W.L. Brown, P.R. Day, W.H. Gabelman, J.B. Hanson, R.F. Holland, A.L. Hooker, P.R. Jennings, V.A. Johnson, D.C. Peters, M.M. Rhoades, G.F. Sprague, S.G. Stephens, J. Tammon, and W.J. Zaumeyer. 1972. Genetic vulnerability of major crops. National Academy of Sciences, Washington, DC.

Lee, K.M., J.P. Shroyer, T.J. Herrman, and J. Lingenfelser. 2006. Blending hard white wheat to improve grain yield and end-use performances. Crop Science 46:1124-1129.

Ma, Z., D. Zhao, C. Zhang, Z. Zhang, S. Xue, F. Lin, Z. Kong, D. Tian, and Q. Luo. 2007. Molecular genetic analysis of five spike-related traits in wheat using RIL and immortalized F_2 populations. Molecular Genetics and Genomics 277:31-42.

Marshall, D.R., F.W. Ellison, and D.J. Mares. 1984. Effects of grain shape and size on milling yields in wheat. 1. Theoretical-analysis based on simple geometric-models. Australian Journal of Agricultural Research 35:619-630.

Miedaner, T., F. Wilde, B. Steiner, H. Buerstmayr, V. Korzun, and E. Ebmeyer. 2006. Stacking quantitative trait loci (QTL) for Fusarium head blight resistance from non-adapted sources in an European elite spring wheat background and assessing their effects on deoxynivalenol (DON) content and disease severity. Theoretical and Applied Genetics 112:562-569.

Morris, V.H., T.L. Alexander, and E.D. Pascoe. 1946. Studies of the composition of the wheat kernel. III. Distribution for ash and protein in central and peripheral zones of whole kernels. Cereal Chemistry 23:540-547.

Nelson, P.T., and M.M. Goodman. 2008. Evaluation of elite exotic maize inbreds for use in temperate breeding. Crop Science 48:85-92.

Ohm, J.B., O.K. Chung, and C.W. Deyoe. 1998. Single-kernel characteristics of hard winter wheats in relation to milling and baking quality. Cereal Chemistry 75:156-161.

Osborne, B.G., and R.S. Anderssen. 2003. Single-kernel characterization principles and applications. Cereal Chemistry. 80:613-622.

Patil, V.D., M.V.S. Reddy, and Y.S. Nerkar. 1994. Efficiency of early generation selections for yield and related characters in safflower (*Carthamus tinctorius* L.). Theoretical and Applied Genetics 89:293-296.

Patterson, A.H., S.D. Tanksley, and M.E. Sorrells. 1991. DNA markers in plant improvement. Advances in Agronomy 46:39-90.

Satumbaga, R., C. Martin, D. Eustace, and C.W. Deyoe. 1995. Relationship of physical and milling properties of hard red winter wheat using the single kernel wheat characterization system. Association of Operative Millers Bulletin. January 6487-6496.

Slaughter, D.C., K.H. Norris, and W.R. Hruschka. 1992. Quality and classification of hard red wheat. Cereal Chemistry 69:428-432.

Souza, E.J., J.M. Martin, M.J. Guttieri, K. O'Brien, D.K. Habernicht, S.P. Lanning, G.R. Carlson, and L.E. Talbert. 2004. Influence of genotype, environment, and nitrogen management on spring wheat quality. Crop Science 44:425-432.

Stalker, H.T. 1980. Utilization of wild species for crop improvement. Advances in Agronomy 33:111-147.

Sun, X.Y., K. Wu, Y. Zhao, F.M. Kong, G.Z. Han, H.M. Jiang, X.J. Huang, R.J. Li, H.G. Wang, and S.S. Li. 2009. QTL analysis of kernel shape and weight using recombinant inbred lines in wheat. Euphytica 165:615-624.

Troccoli, A., N. Di Fonzo. 1999. Relationship between kernel size features and test weight in Triticum durum. Cereal Chemistry 76:45-49.

Tsilo, T.J., G.A. Hareland, S. Simsek, S. Chao, and J.A. Anderson. 2010. Genome mapping of kernel characteristics in hard red spring wheat breeding lines. Theoretical and Applied Genetics 121:717-730.

USDA-ERS. 2011. World wheat production, supply and disappearance. Available at http://www.ers.usda.gov/data/wheat/WheatYearbook.aspx (verified 11 Mar 2011). Economic Research Service, Washington, DC.

Zamir, D. 2001. Improving plant breeding with exotic genetic libraries. Nature Review. Genetics 2:983-989.