CHAPTER- V

DISCUSSION

5.1 Screening of spring wheat genotypes for PHST Sensitiveness in wheat genotypes cultivated in northern Iran

The results of descriptive statistics of agronomic traits responsible for PHS in spring wheat genotypes (Table 4.1) revealed that minimum, maximum, mean, SD and range for selected genotypes may help for effective selection of genotypes for improvement of yield and its important components, as well as PHST in subsequently segregating populations. Success of plant breeding for PHST and yield improvement mainly depends on the existence of genetic variability in wheat for particular agronomic traits (Fu and Somers, 2009). Sheykhi et.al., (2014) studied segregation of some wheat genotypes and also estimated minimum, maximum, mean, SD and range for 30 bread wheat genotypes cultivated at Mazandaran, Bayekola Research Station and Neka (Iran). They also noted that descriptive statistics may help in selection of breeding materials having potential for yield improvement under PHS conditions. Similarly Pirdashti et.al.,(2012) evaluated the most effective variables based on statistical analysis for 60 wheat genotypes grown at Sari and supported the above finding. Present investigation on 40 spring wheat genotypes indicated that the different genotypes had different potential for PHST.

Fu and Somers, 2009 and Lobell et.al.,(2005) reported that grain yield of wheat is the integration of many variables that affect plant growth throughout the growing period. Great efforts have been made to develop proper models that can help to foretell the grain yield in wheat and discriminate the ideal crop. The production efficiency of tillers and kernels of wheat positively improve the yield. Their studies have reflected the importance of either variables, particularly, kernels/spike in breeding programs. Weight of grains/spike was reported by many researchers as the most closely variable related to grain yield per unit area and was often used in selecting high yielding wheat genotypes. Meanwhile, 1000-grain weight had been shown as the main yield component accounting for 20% of variation in wheat grain yield (Kumbhar et.al., 1983; Collaku, 1989).
The results of present investigation regarding variance analysis for different traits related to PHS in selected wheat genotypes (Table 4.2) are in agreement with the finding of Sheykhi et.al.,(2014). They had also studied segregation of some wheat \textit{(Triticum aestivum L.)} genotypes using cluster analysis and reported significant differences between groups in terms of all studied traits. They noted that there was high genetic variation between groups.

Tahmasebi et.al.,(2013) and Ahmadi et.al.,( 2012) had also studied the evaluation of yield and yield components in some promising wheat lines and evaluation of genetic diversity in land races of bread wheat under irrigated and rainfed conditions. They reported that the analysis of variance revealed highly significant difference among lines for all traits studied except for grain filling period and number of grains per spike. The significant difference among genotypes for the traits implies the presence of substantial variation among genotypes which is central for the study of traits giving an opportunity to plant breeders for improvement of these characters through breeding.

Ahmadi et.al.,( 2012) found that all agro-morphological and phenological traits measured, revealed significant differences among the genotypes indicating a high genetic diversity and possible detection of PHS tolerant genotypes. The results of combined analysis of variance showed highly significant differences among environments (irrigated and rainfed conditions) for all traits except spike length(SL), days to flowering (DF), plant height (PH), awn length (AW) and peduncle length/plant height (PL/PH) exhibiting that PHS stress had significant effect on most of the traits. Genotype × environment interaction was significant for number of spike per m\textsuperscript{2} (NSP m\textsuperscript{2}), number of seed per spike (NSPS) and thousand seed weight (TSW) indicating that yield components are sensitive to environmental fluctuations. They concluded that effective crop improvement depends on the extent of genetic diversity in the gene pools.

The results on mean comparison of variables in cluster analysis (Table 4.3 & 4.4; Fig 4.11) are in conformity with Tahmasebi et.al.,(2013) and Ahmadi et.al.,(2012) for different wheat genotypes. Etezadi Jam et.al.,(2005) studied the pre-harvest sprouting resistance, correlation and path analysis of seed characteristics with
pre-harvest sprouting resistance in bread wheat (*Triticum aestivum* L.) and reported significant genetic variations for all traits among cultivars. The results of screening methods indicated that most Iranian bread cultivars had low resistance to pre-harvest sprouting. Correlation analysis revealed negative significant correlation between pre-harvest sprouting, seed dormancy and falling number. The positive significant correlation was observed between seed dormancy and falling number. The study of correlation indicated that after-ripening stage highest correlation with sprouting score as well as with sprouting index. Cluster analysis indicated that there exist a favorable diversity among cultivars. The results of present investigation indicated that genotypes in first cluster had more grain yield and tolerance to PHS during less than 21 days of MI, without any remarkable reduction in kernel weight. However the genotypes in third cluster were highly sensitive to PHS and consequently there was very high reduction in grain yield.

Khodadadi et.al.,(2011) investigated genetic diversity of wheat (*Triticum aestivum* L.) genotypes based on cluster and principal component analyses for breeding strategies and concluded that genetic diversity of plants determines their potential for improved efficiency and hence their use for breeding is very important. They noted that high heritability of flag leaf width and flag leaf sheath distance to the spike indicated that these traits were under genetic control and very few genes control these parameters (Feng et.al.,2006; Fu and Somers, 2009; Mohammadi et.al., 2011). Therefore, for improving these traits breeding program without progeny test can be implemented. Flag leaf length and emergence time exhibited the lowest heritability. Thus these traits were mostly under environment control and for improving these traits selection based on progeny test should be done.

Similar results for these traits correlation was reported in the previous studies (Shahid et.al.,2002; Saleem et.al.,2006; Eivazi et.al.,2007). In present study the characters showing high heritability such as yield, kernel weight and colour seems to be under genetic control while others with low heritability such as duration for heading and maturity, tolerance to PHS and diseases like yellow rust, *Fusarium* and powdery mildew may be under environment control.
Rasaei et al., (2011) studied the traits correlation and path analysis of the grain yield of peas. Similarly Baodi et al., (2008) investigated the relationship between leaf water use efficiency and physio-biochemical traits of winter wheat. The researchers like Mohammadi et al., (2011) and Tahmasebi et al., (2013) also studied the correlation between different variables and grain yield in wheat. They concluded that understanding of the relationship between the traits, for the selection of the important traits, is of utmost importance. For development of high-yielding and stress tolerant varieties thorough knowledge of the existing genetic variation for these parameters is highly essential. In present study correlation between different parameters such as duration of maturity and heading, plant height, kernel colour, kernel weight before and after MI, yield before and after MI, duration of MI and tolerance to PHS had shown positive as well as negative correlations. Grain yield and PHST are positively correlated (Table 4.5).

The results of stepwise multiple linear regression for different traits and rotated factor analysis and communalities for the selected variables in 40 wheat genotypes (Table 4.6 and 4.7) are in agreement with the findings of Pirdashti et al., (2012). They reported that different wheat genotypes demonstrated regression coefficients and the probability of the estimated variables in predicting wheat genotypes’ grain yield. The results of present study showed the prediction equation for grain yield. The formula explains 99% of the total variation within the grain yield components, while the remaining 1% perhaps was due to residual effects. The t-test showed that stem diameter, number of filled grains and 1000-grains weight unlike other five variables significantly contributed the grain yield. These results emphasized on the importance of the above mentioned three variables in wheat selection for breeding programs. These findings are in agreement with the results obtained by Kumbhar et al., (1983). Furthermore, Asseng et al., (2002) reported that increased kernel number could improve potential yield of wheat.

According to Tahmasebi et al., (2013) multiple linear regression method is used to determine the role of yield components in increasing the yield and selection efficiency by means of few traits as the effective indicator to obtain breeding aims (Farshadfar, 2004). The results of stepwise regression analysis were calculated by considering the grain yield as the dependent variable and other characters as the
independent variables. Results showed that, number of spike, 1000-grain weight and plant height remained in the final model and explaining 73.2% of variation in the yield. Thus, the standard linear regression for grain yield was: 

$$MI \ Y = 0.269 - 0.097 x_{12} - 0.851 x_9.$$ 

Efyoni and Mahloji (2005) used stepwise regression analysis in 42 lines of bread wheat, and showed that the grain yield period, the number of grains per spike, the number of spikes per m² and plant height entered into regression model sooner than other traits and were the most effective traits on grain yield. Determination of genetic variation is useful for plant breeding and hence production of more efficient plant species. Baodi et.al., (2008) reported that regression coefficients and the probability of the estimated variables in predicting wheat leaf water use efficiency (WUE). The results showed that the prediction equation for leaf WUE ($y$) was formulated by using the relative physiological traits. The formula explains 99.8% of the total variation within the WUE relative physiological traits, while the remaining 0.2% may be due to residual effects. The $t$-test suggested that RWL had contributed significantly towards leaf WUE. Tahmasebi et.al., (2013), showed that throughout wheat’s early, middle and late grain filling stages, leaf WUE was extremely positive, correlated with Pn, while it was notably negatively correlated with transpiration rate and internal CO₂. In present study 98.3% of the total variation in grain yield after MI could be attributed two kernel weight after MI (0.969) and days to PHS (0.014). The $t$-test showed that kernel weight after MI($X_9$) and days to PHS ($X_{12}$) contribute significantly to PHS as compared to other variables. These two variables are very useful in selection of wheat genotypes in breeding programs under taken for PHS tolerance in different wheat genotypes (Table 4.6 and 4.7).

5.2 Factor analysis

The data presented in Table 4.8 and Fig 4.12 demonstrated that four main factors were responsible for 76.267 % of the total variability in the dependent structure. The first factor duration of MI(41.020%), second factor yield after MI(13.97%), third factor kernel color(12.206%) and fourth factor yellow rust(9.061%). Heaton and Solo (2002) claimed that there has been much recent interest in forecasting, based on factor analysis models for large observation
dimension (p). The factor analysis in present investigation served the same purpose. Aharizad et al., (2012) studied multivariate analysis of genetic diversity in wheat (*Triticum aestivum* L.) recombinant inbred lines using agronomic traits such as days to heading, flag leaf area, peduncle length, spike length, plant height, number of spikelet per spike, number of spikes, number of grains per spike, 1000 grain weight, grain yield, shoot biomass, percent of grain protein, straw yield and harvest index and reported that cluster analysis based on all the traits had assigned the lines into three groups and group two lines showed highest mean of grain yield. In factor analysis, five first factors explained 80.26% of total variation.

First factor determining 23.94% of the variation was named as grain yield factor. Cluster analysis based on the five factors grouped the lines into three groups. The first group lines were superior with respect to grain yield. Similarly Ahmad et al., (2014) had also studied multivariate analysis of some metric traits in bread wheat and reported the importance of factor analysis in determination of genetic diversity of wheat, useful for plant breeding and production of more efficient plant species under different conditions.

Abdi and Williams (2010) highlighted that the principal component analysis (PCA) is probably the most popular multivariate statistical technique used by almost all scientific disciplines, which is the oldest multivariate technique. PCA analyzes a data table representing observations described by several dependent variables, which are, in general, inter-correlated. Its goal is to extract the important information from the data table and to express this information as a set of new orthogonal variables. The multivariate analysis of different variables related to PHS in selected genotypes of wheat also helped to analyze the genetic diversity, Bai and Ng, (2013) supported above view principal components estimation and identification of static factors.

### 5.3 Field evaluation of selected wheat genotypes under MI

The results on field evaluation of selected wheat genotypes under MI conditions and variance analysis of different traits related to PHS in wheat genotypes shown in Table 4.9 indicated that given genotypes significantly interact to MI period (AB) in terms of duration of MI, damage percentage and severity, biological yield, harvest index, kernel weight and size, number of tillers and yield per plot.
However number of plants per plot, length of spike, number of kernels per spike and density of kernel per spike were non significant. Mist irrigation with humidity (50-70 %.), rainfall (30-50 mm) and temperature (25-30 °C) was the most suitable microclimate for sprouting of wheat genotypes. Generally, increasing of mist irrigation period markedly reduced the kernel size (11.03), kernel weight (1.311), biological yield (58.8 t/ha) and grain yield (32.96t/ha) while damage severity (11.03) and percentage was (0.172) significantly increased.

The results of present investigation are in agreement with the work of many researchers such as DePauw et.al.,(2012) ; Bi et.al.,(2014) ; Chang et.al,(2010); De Laethauwer et.al.,(2012); De Laethauwer et.al.,(2013); Lohwasser et.al(2013); Masojc et.al(2013) and Singh et.al.,(2010). All the above researchers reported that PHS in spring wheat and durum wheat is the serious problem in many countries during prolonged wet weather conditions. The principal factor in PHS is early breakage of seed dormancy which cause reduction in wheat grain yield through reduction in test weight and negatively affect end used quality of flour products which directly affect bread making quality.

They further explained that development of PHS resistant cultivars of wheat is the most important solution to reduce loss and improve the grain yield. But development of such cultivar is highly complicated as it is under genetic control. The important traits to be focused for developing PHS resistance in wheat genotypes are spike morphology, duration maturity, seed dormancy, seed coat colour, environmental conditions, α-amylase activity etc. In our studies of field evaluation of selected genotypes under MI, the main traits analyzed were damage severity, damage percentage, harvest index, biological yield, kernel weight, number of kernels per spike and length of spike.

The correlation of all above traits with PHS and grain yield is shown in Table 4.11 revealed that selected characters such as duration of MI (-0.42;P<0.01), kernel size (-0.77 ;P<0.01), number of kernels per spike (-0.15; P<0.01), damage severity (-0.91 ;P<0.01), kernel weight (-0.84** p<0.01) harvest index(2.74**P<0.01) and damage percentage (-7.40;P<0.01) had shown significant negative correlation with economic yield under MI. However, number of tillers per plot, number of plants per
plot, length of spike, density of kernels per spike and biological yield had shown non significant correlation with yield. The results clearly indicated that with increasing duration of MI there was considerable reduction in economic yield. The positive correlation between all the characters studied with damage severity under MI may help to select the most tolerant wheat genotypes to PHS.

The findings of present investigation are consistent with results reported by Gerjets et al. (2010) and Knox et al., (2012). They studied the PHS resistance in durum wheat and reported that characterization of pre-harvest sprouting resistance is important for selection in breeding to understand the genetics of the traits. Methods of measuring dormancy and other factors contributing to pre-harvest sprouting resistance are varied. They explained that only three durations of MI such as 7, 14 and 21 days is not sufficient for characterizing sprouting resistance. Similar was the observation of Kulwal et al., (2012) they noted significant correlations as well as rank correlations for the PHS scores across the years. Highly significant correlations and rank correlations were observed for PHS scores for the different durations of MI after ripening period. Non significant negative correlations was noted between the PHS score, plant height and days to heading.

5.4 Path analysis showing direct and indirect effects on selected characters of grain yield in wheat genotypes.

The correlation coefficients were partitioned in to direct and indirect effects (Table 4.12 a&b). The results of path analysis showed that damage percentage (-0.8357), harvest index (-1.908), biological yield (-0.315), kernel density per spike (-0.287) and spike number per plot (-0.529) had high negative direct effects on grain yield. While indirect negative effects on grain yield with duration of MI (-0.684), damage severity (-5.594), kernel weight (-1.489), number of kernels per spike (-0.497) and spike length (-0.166) was recorded. Finally, MI remarkably reduced the wheat grain yield, kernel weight and size, biological yield and harvest index. However, the selected genotypes responded differently to MI conditions and there was a significant variation for selecting the suitable genotypes showing tolerance to PHS. The results of present investigation are in agreement with the studies of many researchers like Rasaei et al., (2011); Cyprien and Kumar, (2011). They indicated that
path coefficient analysis is a statistical technique of partitioning the correlation coefficients into its direct and indirect effects, so that the contribution of each character to yield could be estimated. It is used in plant breeding programs to determine the nature of the relationships between yield and yield components that are useful as selection criteria to improve the crop yield under different environmental conditions such as salt, drought, high temperature, cold and PHS.

The goal of the path analysis is to accept descriptions of the correlation between the traits, based on a model of cause and effect relationship and to estimate the importance of the affecting traits on a specific trait. If the cause and effect relationship is well defined, it is possible to present the whole system of variables in the form of the diagram, known as path-diagram.

In presents study path analysis have helped to understand the relationship between various traits such as nature of spike, damage percentage, harvest index, biological yield, kernel density per spike, spike number per plot, duration of MI, damage severity, kernel weight, number of kernels per spike and spike length and the grain yield in different wheat genotypes. The path analysis has utmost importance in selection of PHST genotypes under different MI conditions (7, 14 and 21 days). The different traits under study may have association with each other that ultimately affect grain yield under MI, indicating the tolerance and sensitiveness of spring wheat cultivars.

The value of Karl Pearson’s correlation coefficient \( r \) helps in identifying the association between two characters. It should be noted that it does not measure the magnitude of the association but gives the idea about it. If it is closer to -1 or +1, there is high degree of linear relationship. If it is closer to 0, there is no linear relationship; there may be other type of relationship between them. The results of present study indicated the inter-character correlations among above mentioned traits such as harvest index, kernel weight, damage severity, damage percentage, MI and grain yield after and before MI. The traits like harvest index and damage percentage had direct effect on grain yield while damage severity had indirect effect on grain yield reduction. Similar studies were conducted by Mohammadi et al.,(2011) and Pirdashti et al.,(2012); Cokking and Colkesen(2007); Dewey and Lu(1959);
Shrivastava et.al.,(2001); Salehi et.al.,(2010); Etezadi Jam et.al.,(2005); Shahid et.al.,(2002); Saleem et.al.,(2006); Rasaei et.al.,(2011); Barnard et.al.,(1998); Collaku (1989); El-Deeb and Mohamed (1999); Naser and Leilah (1993); Subhani and Khaliq (1994); Ehdaie and Waines (1989) on different crops like wheat, pea, barley, wheatgrass, soybean and common bean.

5.5 Physiological and biochemical traits of wheat genotypes during PHS

5.5.1 Carbohydrates

The results shown in (Table 4.13; 4.14 a&b; 4.15; 4.25a&b and 4.26) and (Figure 4.19) revealed that carbohydrate contents showed significant changes under different condition of MI in both PHS sensitive and tolerant genotypes of wheat. The contents of carbohydrates were comparatively higher in sensitive genotypes as compared to PHST. The carbohydrate metabolism plays a crucial role in PHS and changes according to environmental conditions (Price et.al., 2004). During PHS carbohydrates like starch are degraded releasing sugars, which are transported to developing embryo and finally cause emergence of radical and plumule in the process of seed germination (Bewely and Blak, 1994).

Many researchers (Dreccr et.al., 2008 and 2014; Xue et.al., 2008; Rebetzke et.al., 2008) had studied the physiological role of carbohydrates and sugars in development of grains, spike growth, spike carbohydrate pool, grain number per-spikelet and per spike under different abiotic stress conditions. The carbohydrate metabolism influences the final number of fertile florets under stress conditions. The enzymes of carbohydrate metabolism such as invertases, sucrose synthases, sucrose phosphate synthases etc, play key role in grain set in tolerant maize germplasm and pollen viability in wheat (Xue et.al., 2008). The role of glucose in floral abortion is also well documented (Dreccr et.al., 2008 and 2014). The difference in storage carbohydrate accumulation in drought-sensitive and drought tolerant wheat varieties was fully investigated by these workers. The results of present study indicated that PHS sensitive genotypes like N-87-8 and N-87-12 showed low carbohydrates content as compared to tolerant genotypes. The level of accumulation of carbohydrate may act as an indicator in screening of wheat genotypes under PHS and it may serve as the reliable biochemical marker to understand their sensitiveness / tolerance to PHS.
5.5.2 Sugars

The results on sugar contents in advanced lines of spring wheat under MI revealed that the tolerant variety had less sugars as compared PHS sensitive cultivar (Table 4.13; 4.14 a\&b; 4.17; 4.25 a & b and 4.26) and (Fig 4.21), which is attributed to high α-amylase activity in sensitive variety. As this enzyme degrade the starch, releasing glucose and fructose, leading to considerable pre-harvest sprouting of wheat grains in susceptible cultivar (De Laethauwer et al., 2013). According to Mishra and Dubey, 2008-2013; Gupta and Kaur, (2005) soluble sugars, especially sucrose and hexoses are highly sensitive to environmental stresses and serve as substrates for cellular respiration as well as osmolytes to maintain cell homeostasis under stress. Sugars co-ordinately regulate expression of growth and stress related genes in plants and respond to environmental stresses like PHS through sugar sensing mechanisms (Panda et al., 2010; Sharma and Dubey, 2011). Zielinska-Dawidziak et al., (2013) reported that sprouted wheat grains had more reducing sugars due to increasing activity of hydrolytic enzymes. The results of present investigation are in agreement with above finding. Sugars in grains are very important in modulation of plant genes and the sugars- sensing mechanism(s) underlying gene regulation by carbohydrates in plant systems was reported by Jang and Sheen, (1997).

According to Umemura et al., (1998) sugar sensing and α-amylase gene repression in rice embryos showed positive correlation. Kaur et al., (2000) observed that sugar signaling pathway interact with stress pathway to modulate carbohydrate metabolism and indirectly play an important role during plant growth and development under abiotic stress conditions. A large number of stress responsive genes have been reported to be induced by glucose, indicating the role of sugars in environmental responses (Price et al., 2004; Gupta and Kaur 2005).

The environmental stress conditions such as PHS lead to major alterations in carbohydrate metabolism (Gupta and Kaur, 2000). Many researchers such as Ghanbari and Mir (2013); Lunner et al., (2000) and Kaur et al (2005) reported that starch and sugars play key role(s) in PHS of cereals like wheat. Many other factors like reduced water absorption by grains, germination inhibitors, level of α-amylase activity in grains also contribute to PHST (Groos et al., 2002). According to Zhang
et.al., (2011) and Hu et.al., (2012) sugars, starch and antioxidants like proline, phenols, glycine betaine are important factors in PHS. They claimed that glucose and different antioxidants had pivotal role in PHS tolerance/sensitiveness of wheat. Mishra and Dubey (2013) also noted that abiotic stress in rice caused marked perturbations in metabolism of carbohydrates, leading to increased accumulation of soluble sugars. The results of present investigation are in conformity with above findings. The PHS sensitive wheat genotypes such as N-87-8 and N-87-12 had shown very high level of sugars as compared to PHS tolerant genotypes (N-76-12), which may be due to enhanced level of amylase activity.

5.5.3 Starch

According to Dupont and Altenbach (2003) and Rahman et.al.,(2000) starch is a major determinant of yield, accounting for 65–75% of the grain dry weight and up to 80% of the endosperm dry weight. A series of enzymes synthesize the amylose and amyllopectin chains that comprise starch (Ball et.al.,1998). Within the amyloplast, ADP glucose pyrophosphorylase converts glucose 1-phosphate to ADP-glucose, which then is converted into amylose and amyllopectin polymers by starch synthases and branching enzymes. The starch polymers form layered granules within the amyloplasts. Large type A granules are initiated about 4–7 days after anthesis (DPA), and smaller type B granules appear around 10–12 DPA (Peng et.al.,1999; Langeveld et.al.,2000). Many of the genes that encode enzymes required for starch biosynthesis have been sequenced (McCue et.al., 2002; Vrinten and Nakamura, 2000). The information is available on interactions between transcription factors and promoter binding sites of genes encoding starch biosynthetic enzymes in barley and maize endosperm (Kim and Guiltinan, 1999; Zentella and Yamauchi, 2002). Reductions in starch accumulation during extreme environmental conditions like humidity, rainfall and temperature account for significant loss in grain yield (Bhullar and Jenner, 1985; Tashiro and Wardlaw, 1989).

The decline in starch content for Australian varieties exposed to low temperatures (during PHS) was associated with a decrease in rate of conversion of sucrose to starch (Bhullar and Jenner, 1985). High temperature cause decrease in levels of fructose, hexose phosphate, and sugar nucleotides (Jenner, 1991) and reduce
the activity of some enzymes in starch biosynthetic pathways, especially soluble starch synthase (Keeling et al., 1993; Rijven, 1986). Diminished rates of starch production in wheat endosperm due to changes in temperature was hypothesized to be due to inactivation of starch synthase, a key enzyme in the starch biosynthetic pathway (Altenbach et al., 2003; Hurkman et al., 2003).

The observed decrease in starch per kernel resulted from a decrease in duration of starch accumulation. Guedira and Paulsen (2002) observed that high shoot or high root temperature cause reduction in starch accumulation in the grains, mainly through the effect on duration of starch accumulation, suggesting that high temperature influences the duration of grain filling. According to Altenbach et al. (2003) and Guedira and Paulsen (2002) the accumulation of starch began earlier when plants are exposed to abiotic stress like high temperature during grain filling. Environment mainly influences expression of genes for the enzymes involved in starch biosynthesis. It affects formation of starch granules and amylose/amylopectin ratios (Hurkman et al., 2003). The environmental conditions like rainfall at harvest time are pivotal in PHS of many cereals like wheat, ray, sorghum and barley, because it induces metabolic alterations in carbohydrates like starch and sugars. The favorable or adverse impact on grain carbohydrates governs the PHS tolerance or sensitiveness of the genotypes. The major carbohydrates and activity of amylase enzyme decide whether PHS will occur or not in the grains. The results on starch content in selected elite lines of spring wheat indicated significant alterations with duration of MI (Table 4.13; 4.14 a & b; 4.16; 4.25 a & b and 4.26) and (Fig 4.20). The tolerant variety showed more accumulation of starch as compared to PHS sensitive cultivar of wheat. The grain starch is most important end product of cereals as they contain about 70% (w/w) starch and 50% of the calories consumed by humans (Thitisaksakul et al., 2012; WHO, 2003). Studies on changes in starch content may help to improve and avoid its degradation during PHS, because during germination starch is mobilized by the action of hydrolytic enzymes, which are synthesized in the aleurone layer and in the scutellum and secreted into the starchy endosperm of germinating grains (Shaik et al., 2014).

The increase/decrease in starch content during germination is controlled by activity of α-amylase. In present investigation the wheat cultivar tolerant to PHS (N-
86-12) showed less α-amylase activity and more starch in it. While opposite trend was observed in PHS sensitive variety (N-87-8). Our results on starch contents are in accordance with above findings.

5.5.4 Protein

The results shown in Table 4.13; 4.14 a& b; 4.21; 4.25 a & b and 4.26) and (Fig 4.25) revealed that protein contents were reduced during second and third step of MI in PHS sensitive wheat genotype (N-87-8) as compared to PHST genotype (N-86-12). Protein metabolism during seed germination is highly important, which is degraded by enzyme protease, releasing different amino acids, which are utilized by developing embryo (Bewly and Black, 1994). The breakdown of protein is very fast in wheat grains showing sprouting under MI in PHS sensitive genotypes and it shows considerable reduction in proteins. But in PHS tolerant genotypes due to dormancy inducing compounds like phenols there is no seed germination and no utilization of reserve food material like protein and starch hence the PHST genotypes showed high protein contents. The alterations in protein content may act as biochemical marker for screening the PHST/ sensitiveness of wheat genotypes.

Many researchers like Awole et.al.,(2012); Morris et.al.,(2013); Shaik et.al.,(2014);Oszvald et.al.,(2014); Ade-Omowaye et.al., (2008);Fu et.al., (2014) had indicated importance of protein in grain quality which changes according to environmental conditions and genotype. Mature wheat grains contain 8–20% protein, including the gluten storage proteins that are enriched in proline and glutamine. The abundant gluten proteins constitute up to 80% of total flour protein, and confer properties of elasticity and extensibility that are essential for functionality of wheat flours (Shewry et.al., 1994; Shewry, 1995).

The gluten proteins consist of monomeric gliadins and polymeric glutenins. The gliadins constitute from 30 to 40% of total flour protein and are a polymorphic mixture of proteins. The glutenin polymers consist of low molecular weight glutenin subunits (LMW-GS) of about 40 kDa linked by interchain disulphide bonds to high molecular weight glutenin subunits (HMW-GS) of about 90 kDa. The LMW-GS most closely resemble g-gliadins in sequence (Muller et.al., 1998) and comprise
about 20–30% of the total protein (Gupta et al., 1992) while the HMW-GS account for about 5–10% of the total protein (Payne, 1986).

Three to five HMW-GS and 15–20 different LMW-GS proteins are recognized in 2D gels of hexaploid wheat (Lew et al., 1992). The roles of the individual gluten components in dough functionality are complex (Khatkar et al., 2002). Although HMW-GS constitute no more than 10% of total flour protein, they may be the most important determinants of bread making quality because of their importance in forming the glutenin polymer.

Water-soluble albumins and salt-soluble globulins constitute 10 to 22% of total flour protein (Singh and Skerritt, 2001; Gupta et al., 1989). Predominant albumins and globulins such as alpha-amylase/trypsin inhibitors serpins and purothionins (Ostergaard et al., 2000; Garcia-Olmedo et al., 2002) may have dual roles as nutrient reserves for the germinating embryo and as inhibitors of insects and fungal pathogens prior to germination. Triticin is related to storage globulins in oats, rice and legumes (Singh et al., 1991; Pence et al., 1954) and puroindolines influence grain hardness (Morris, 2002; Kobrehel and Alary, 1989).

Unlike rice or maize, there are no recognizable protein bodies in the endosperm of the mature wheat grain. Instead of this protein in the vacuole-like compartments is compressed between the starch granules, with the loss of recognizable compartmentation (Chrispeels and Herman, 2000). Field studies indicated that environmental conditions, particularly fertilizers and temperature, affect the content of protein and its composition (Luo et al., 2000; Johansson et al., 2001; Tabe et al., 2002; Panozzo and Eagles, 2000).

The quantity as well as quality of protein is badly affected by PHS in wheat grains and cause serious losses in end use quality. Like carbohydrates, protein also plays major role in PHS, hence it may also act as biochemical marker for PHS in wheat and other cereals.
5.6 Antioxidants

5.6.1 Proline

Proline is a multifunctional amino acid, accumulated during stress conditions and acting as a compatible osmolyte/antioxidant, helping the plants in stress adaptation, recovery and signaling as well as modulating responses to abiotic stress like PHS (Szabados and Saroure 2009). In the present study proline was accumulated to maximum level in PHS tolerant genotypes under MI condition for 21 days (Table 4.13; 4.14a & b; 4.18; 4.25 a & b and 4.26) and (Fig 4.22). Kishor et.al.,(2005) also reported significant accumulation of proline under stress conditions due to increased synthesis.

The tolerant variety was showing less PHS due to increasing proline content as compared to susceptible cultivar showing very high PHS. As stated by Verbruggen and Hermans (2008); Xiao et.al.,(2012) ; Kostal et.al.,(2011) any biotic or abiotic stress induce proline accumulation in a wide range of plants. Proline accumulation is believed to be very important in PHS tolerance. The level of proline that accumulates in plants in response to stress varies greatly and is highly dependent on the plant species. PHS tolerant genotype N-86-12 accumulated more proline to ameliorate the PHS stress. The results of present study are in conformance with above findings and proline accumulation in different wheat genotypes may act as a biochemical marker to screen these genotypes for their tolerance / sensitiveness to PHS.

5.6.2 Phenol

The results shown in Table 4.13; 4.14 a & b; 4.19; 4.25 a & b and 4.26 ) and (Fig 4.23) revealed that phenolic contents increased in PHS tolerant genotypes of wheat during 21 days of MI as compared to PHS sensitive genotype. According to Weidner et.al.,(1999) phenol plays a key role in the dormancy of cereal caryopses. They explained that level of phenolic acids in caryopses of wheat, rye and triticale contribute to their tolerance to PHS, because these chemical compounds induce seed dormancy. Weidner et.al.,(2002) also indicated that the seeds contain numerous germination inhibitors like phenols which protect them against decaying and at the same time control their germination by imposing dormancy. It is well documented
that phenolic compounds are often synthesized in response to biotic and abiotic stress, as they act as antioxidant to scavenge free radicals generated under stress full environment (Wiedner, 2001). Gatford et al., (2002) had reported that tolerance to PHS in wild relatives of wheat (Triticum tausehii) was due to grain dormancy imposed by different types of phenolics. The PHS tolerance in wheat genotype N-86-12 during 21 days of MI, might be due to high phenolic content in its grains.

A large body of evidences had demonstrated that phenolic compounds always show negative relationship with the PHS tolerance, therefore the varieties with high phenolic content had very high seed dormancy and these are not prone to germination before harvest as they become resistant to PHS (Weidner et al., 2002; Gao et al., 2013). The finding of present study is in agreement with above researchers, indicating that analysis of phenols may act as an additional reliable physiological indicator to identify the wheat varieties as tolerant / susceptible to PHS.

5.6.3 Amylase

The results shown in Table 4.13; 4.14 a & b; 4.23; 4.24; 4.25 a & b and 4.26) and Fig 4.27; 4.28) clearly indicated that during third step of MI (21 days) the PHS susceptible genotypes had shown very high activity of α, β and total amylase as compared to the PHST genotypes. The results of present investigation are in agreement with many researchers like De Laethauwer et al., (2013), Gao et al., (2013), DePauw and McCaig (1991), Singh et al., (2010), Xing et al., (2010), Jaiswal et al., (2012), Clarke et al., (2005), Singh et al., (2014) and Ghanbari and Mir (2013). The also recorded stimulated activity of amylase in different crops during PHS.

The above workers had concentrated on the integrated study of the genetical and physiological background of PHS, which is a multidisciplinary approach. They pursued monitoring of α-amylase activity, an enzyme that is involved in PHS, both at transcriptional and post-transcriptional levels during kernel development. They further explained that damage caused by PHS has often been associated with increased levels of α-amylase activity in the kernel. By converting starch into soluble sugars, high α-amylase activity levels negatively affect the nutritional and end-use quality of grains (Mares and Mrva, 2008). Several tests assessing this damage due to PHS are based on α-amylase activity levels at harvest ripeness. At present a weak
relationship between the level of seed dormancy and α-amylase activity in wheat, triticale and barley is reported (Lindblom et. al., 1989; Lin et. al., 2008). Although PHS is often the primary source of increased α-amylase activity, several other sources of α-amylase may obscure this weak relationship, e.g. retained pericarp α-amylase activity, late maturity α-amylase (LMA) activity and sprouting before physiological maturity (PM) has been studied in detail (Lunn et. al., 2001).

Although different levels of α-amylase activity have been detected in cereals like wheat, rye and triticale, they all showed a typical pattern during kernel development. Shortly after pollination, α-amylase activity increases and reaches a peak at 12–15 days post anthesis (DPA), followed by a rapid decrease during the early stages of maturation (Oettler 1990; Mares and Oettler 1991). This peak is associated with the production of low pI α-amylase that is controlled by α-Amy2 genes located on the group 7 chromosomes in wheat (Gale et. al., 1983; Mrva and Mares 1999). However, at about 30 DPA, different patterns of α-amylase activity occur, depending on the genotype. In many wheat and rye genotypes, α-amylase activity remains low until harvest ripeness, whereas it may increase to excessive levels at harvest maturity in certain genotypes of triticale and wheat (Lindblom et. al., 1989; Oettler 1990; Mares and Oettler 1991). These high levels are attributed to high pI α-amylase and are regulated by α-Amy1 genes on the long arm of the group 6 chromosomes in wheat (Rentzsch et. al., 2012; Mrva and Mares 1999).

The varying levels of α-amylase activity in a cereal grain are affected by environmental and genetic factors as well as by interactions between these factors. Among the environmental factors, temperature is probably the most influential. Gale et. al., (1983) found that slow drying conditions, usually accompanied by low temperatures, could enhance the production of α-amylases in maturing wheat just before harvest ripeness.

Conversely, high temperatures after PM appeared to reduce α-amylase activity in wheat (Biddulph et. al., 2008). According to Wu et. al., (2002) and Gao et. al., (2013) the relationship between α-amylase activity and PHS resistance was deemed to be very remarkable. This may be due to activity of α-amylase that would increase quickly once absorbed enough water and then promoted the seed sprouting.
The activity of α-amylase was also found to have a significant difference between the resistant and sensitive varieties to PHS in wheat (Wang et al., 2008). Three isoforms of α-amylase in wheat have been identified affecting PHS, namely α-amylase-1, α-amylase-2 and α-amylase-3 (Gale and Ainsworth, 1984). The expression level of α-amylase-1 and α-amylase-2 could be regulated by GA3 (Marchylo et al., 1983). The activity of α-amylase-1 was deemed to correlate with the degree of seed dormancy, which accounted for 84% of seed germination. Besides the variation of α-amylase, the α-amylase/subtilisin inhibitors (ASI) in wheat, barley, rice and rye were identified via restraining the activity of α-amylase to restrain the seeds germination (Gale and Ainsworth, 1984).

Singh et al. (2014) stated that sprouting in wheat produces the enzyme α-amylase, which leads to lower falling number and influences cooked pasta quality. DePauw and McCaig, (1991) detected a significant positive correlation between values for germination of threshed kernels and levels of α-amylase in kernels from unthreshed spikes subjected to a wetting treatment. Singh et al. (2010) showed that PHS is initially recognized by an elevated level of starch hydrolytic enzyme activities that primarily originate from α-amylases. These enzymes catalyze breakdown of endosperm starch and thus provide the initial energy needed for seed germination. The potential resistance to PHS in white wheat cultivars is based on harvest-time, seed dormancy and spike morphology traits, which are important targets for wheat improvement in regions where moist harvest conditions are frequent. Xing et al. (2010) also claimed that the production of α-amylase in the kernel is the direct result of germination, which includes several different biochemical and structural processes, so there are multiple concurrent changes occurring. According to Jaiswal et al. (2012) α-amylase involved in germination, thus resulting in PHS tolerance. Ghanbari and mir (2013) revealed that PHS negatively affect subsequent grain quality, seed viability, seedling vigor and milling and backing properties reduction in grain quality is caused by conversion of starch to glucose (sugar) by the enzyme α-amylase. The enzyme α-amylase is synthesized in the aleurone layer and scutellum and released in the endosperm to decompose the starch into sugars available for germination (Lunn et al., 2001). Several factors can contribute to increased PHS tolerant, such as reduced level of α-amylase activity in grains, the presence of
inhibitors of germination, reduced water absorption by the grains (Mares et al., 2009; Groos et al., 2002). Potokina et al., (2002) analyzed the α-amylase activity in developing caryopses from sorghum varieties showing contrasting susceptibility to pre-harvest sprouting.

The control of α-amylase activity by gibberellins (GA3) is well documented in cereals (Fincher, 1989). On the other hand, it has been shown that abscisic acid (ABA) can exert an inhibitory effect upon the expression of α-amylase genes. Jacobsen et al., (2002) and Bewley and Black, (1994) showed that sorghum varieties with contrasting pre-harvest sprouting sensitive have different levels of α-amylase activity even when their grains have not shown any signs of sprouting. The sorghum variety IS 9530 very resistant to PHS showed very low α-amylase activity over all the development period. In the present study the PHST cv. N-86-12 also showed very low α-amylase activity, conforming its resistance to PHS. Studies of Kaplan and Guy (2004) have demonstrated that induction in β-amylase in response to abiotic stress which was correlated with maltose accumulation, which has the ability to protect proteins, membranes and the photosynthetic electron transport chain. Therefore β-amylase induction and the resulting maltose accumulation may function as a compatible solute stabilizing cell organelles under stress conditions.

5.6.4 Effect of PHS on grain yield and end use quality

PHS in wheat greatly affect the grain yield in different parts of the world resulting in to substantial financial losses to farmers and food processors. It also decreases the grain value to the producers by impacting four primary grade determinants; (1) test weight (bulk density), (2) vitreousness (translucent properties of kernel), (3) degree of soundness (overall visual grain quality), and (4) percent sprouted kernels, grain quality and end use quality (Gao et al., (2013); Masojć et al., (2013); Jaiswal et al., (2012); Himi et al., (2002); De Laethauwer et al., (2013); De Laethauwer et al., (2012); Zhang et al., (2014). Many researchers reported significant economic losses due to a reduction in grain functionality, grain yield and viability of seed for planting (Liu et al., (2008); Singh et al., (2014); Yang et al., (2014); Kulwal et al., (2012); DePauw et al., (2012).
The results of present study are in close agreement with above findings. Grain yield was very low in PHS sensitive wheat genotypes of spring wheat as compared to tolerant genotypes. This may be due to degradation of starch by the elevated levels of amylase activity and high percentage of PHS and severity. The PHS sensitive genotypes also showed low carbohydrates, protein, phenols and proline under MI (21 days), however sugar content was very high, which might be due to degradation of starch by the enzyme α-amylase. Starch accounts for 64-74% of the total dry weight of wheat grains (McCaig et al. 2006; Kulwa et al., 2012) and its properties are important for determining the end-use quality of wheat flour. PHS has been closely associated with elevated levels of α-amylase. Degradation of native starch granules negatively affect quality of various products made from wheat flour such as breads, cookies and noodles. The primary reason for α-amylase accumulation in the grain is delayed harvest due to wet weather causing breakdown of grain dormancy (De Laethauwer et al., (2012); Kondhare et al., (2014)). Another major cause of excess α-amylase activity is the deposition of α-amylase in the endosperm cavity (Knox et al., 2012). The third source of elevated α-amylase activity is associated with pre-maturity sprouting and involves germination during early grain development when kernels are still at high moisture content (Lunn et al. 2001). Rainfall at harvest, however, is the main cause of PHS inducing α-amylase activity (Mares and Mrva 2008).

Starch from sprout damaged wheat grain exhibit a lower swelling power, gelatinize at a lower temperature and over a narrow temperature range than starches from sound grain (Noda et al. 2004). Breads baked from hard wheats are affected more than other wheat products by PHS. Bread production is complicated by increased stickiness of the dough. Even minor sprout damage can cause significant reductions in gluten strength of wheat flour making it unsuitable for bread making (Noda et al. 2004). Sprout damage affects both the processing and quality of different kinds of noodles. High α-amylase activity in dry noodles weakens the dough so that noodles cannot support their own weight and break during the dehydration process (Hatcher and Symons 2000). Sprout damage also affect the products made from soft white wheat. A reduced thickening power of sprout damaged soft wheat flour results in poor cake baking quality, resulting in cakes with low volume, and a dip in the centre (Knox et al., 2012).
The losses in grain yield, end use quality as well as grain quality during PHS in wheat grains depends on genotype, the environmental conditions during grain development and the interaction between these factors (De Laethauwer et.al., 2009). Hence, cereal breeders constantly seek to improve tolerance to PHS in cereals grown under PHS conducive environmental conditions (De Laethauwer et.al., 2012). In present investigation attempt on some physiological, biochemical, and enzymological traits in spring wheat showing PHS in northern part of Iran was focused. These markers may help the breeders to breed PHS tolerant varieties, because it is the only solution to this problem (DeLaethauwer et.al., 2013). The effect of PHS on grain yield and its quality is very much important issue for farmers which is highly correlated with different traits such as carbohydrates, starch, phenol, proline, protein, sugar, α,β and total amylase. This clearly indicated that PHS in wheat is highly complex and multi-faceted problem (Table 4.13; 4.14 a & b ; 4.20 ; 4.25 a & b and 4.26 ) and (Fig 4.24).

5.6.5 Gene expression of α-amylase during PHS

The results of gene expression of α-amylase during sprouting of grains in all the selected genotypes shown in Table 4.27 ; 4.28) and (Fig 4.29 ; 4.30 and 4.31) clearly revealed that wheat genotypes such as Nai60, N-80-19, N-87-12 and N-87-8 had significant difference between them. Similarly comparison test also revealed that N-87-8 genotype had the highest relative expression for α-amylase gene than other genotypes. Consequently, this genotype was sensitive to sprouting. By contrast, the Nai60 genotype had lower levels of α-amylase gene expression and it is considerably tolerant genotype to sprouting. The enzyme α-amylase was activated during two, four, six and eight days of sprouting period. The bp196 fragments of the amylase gene 3 oxone and bp180 fragment of the beta-actin gene was amplified. Determination index R2 fit linear regression for serial dilutions and higher CT mainly indicates less amount of template or less gene expression. Gachon et.al., (2004) stated that DNA in edible plants, have been the driving force for the introduction of real-time PCR techniques in plant research. This was followed by numerous fundamental research applications aiming to study the expression profiles of endogenous genes and multigene families. Since then, the interest in this technique in the plant scientist community has increased exponentially.
According to De Laethauwer et.al., (2009) there is still a major problem to select pre-harvest sprouting (PHS) tolerant triticale varieties in a reliable, field-independent way.

One approach to minimize the influence of environmental conditions and physio-morphological traits for PHS detection is using molecular genetic tools. Due to the influence of genotype and environmental conditions during grain development and the interaction between these factors, the only solution to this problem seems to breed PHS tolerant varieties. However, a lot of genetic variation has been reported for PHS tolerance, due to the complexity of PHS a molecular genetic approach seems more appropriate to select for PHS tolerant triticale varieties. The role of the ‘viviparous’ Vp1 gene on dormancy in cereals is well known today. The VP1 protein acts as a transcription factor and controls the activation of genes, involved in embryo growth, maturation and dormancy (Hattori et.al., 1994), and represses terminative hydrolyses during kernel development and maturation (Hoecker et.al., 1995). The Vp1 gene is detected in mature embryos and shows great homology in several species (Hattori et.al., 1994; Nakamura and Toyama, 2001). A correlation between the expression level of the Vp1 gene and the level of seed dormancy in wild oat and wheat was reported by Jones et.al., (1997); Nakamura and Toyama, (2001).

Also correlation between the sensitivity of embryos to abscisic acid and the level of Vp1 expression was found (McKibbin et. al., 2002). Furthermore, alternatively spliced transcripts of the Vp1 gene were present in PHS sensitive genotypes of wheat, indicating that PHS sensitive genotypes partly fail to encode full-length VP1 proteins (Wilkinson et.al., 2005).

Several research workers such as Nolan et.al.,(2006); Bustin, (2002); Zhang et.al.,(2008); Masojc et.al.,(2007); De Laethauwer.et.al.,(2012); Zhang et.al.,(2014); Yan et.al.,(2008); Chen et.al.,(2008); Yang et.al.,(2014) have focused their studies on use of RT-PCR and QTL analysis involved in seed dormancy and PHST in different crops such as wheat, rice and many other cereals. The results of present study on RT-PCR are in conformity with above findings.

Many researchers like Kondhare et.al.,(2014;2013;2012); Young et.al.,(1997); Ullrich et.al.,(2009); Wilkinson et.al.,(2002); Hwang et.al.,(1998); Osanai
et.al.,(2005); Potokina et.al.,(2002); Masojc and Milczarski, (2009); Mrva and Mares, (1999); Hidalgo et.al.,(2013 ); Huang et.al.,(2012); Mares and Mrva,(2008); Cejudo et.al.,(1995) have studied the correlation between α–amylase activity and its gene expression, seed germination, seed dormancy and PHST or sensitiveness under different conditions of temperature, moisture / wet conditions and applications of different hormones like GA3 and ABA.

Kondhare et.al.,(2014;2013;2012) investigated the effects of exogenous abscisic acid and gibberellic acid on pre-maturity a-amylase formation, induction/ stimulation and inhibition in wheat grains. Young et.al.,(1997) worked in detail about the changes in carbohydrate composition and a-amylase expression during germination and seedling growth of starch-deficient endosperm mutants of maize. Ullrich et.al.,(2009) reported the genetic relationships between pre-harvest sprouting and dormancy in barley. Similarly Wilkinson et.al.,(2002) noted the use of comparative molecular genetics to study pre harvest sprouting in wheat, The use of model species in both plant and animal systems to study genetics and molecular biology has greatly speeded up the processes of identification, understanding and exploitation of gene functions. It has taken less than 50 years from the elucidation of the structure of DNA.The correlation between polyphenol oxidase, alpha-amylase and beta-amylase activities of *Triticum monococcum*, *Triticum turgidum* and *Triticum aestivum* was investigated by Hidalgo et.al.,(2013 ), which helped to screen sensitiveness or tolerance to PHS in above mentioned species.

Mares and Mrva,(2008) reported late-maturity a-amylase low falling number in wheat in the absence of preharvest sprouting. Potokina et.al.,(2002) explain that germination of seeds is a complex, multi-stage process requiring the coordinated expression of numerous genes in different tissues. Due to the various functions of seed tissues and the different biochemical processes, these genes are expected to be coordinately regulated both spatially and temporally (Bewley and Black 1994). The endosperm consists of the aleurone layer and the starchy endosperm. The aleurone layer is a source of hydrolytic enzymes and also an important storage tissue (Briggs 1992). The process of germination starts with the uptake of water by the seed (imbibition) and ends when the embryonic axis starts to elongate and the radicle emerges (Bewley 1997). Upon imbibitions, the quiescent dry seed rapidly resumes
metabolic activity. Respiration, enzymatic activity, RNA and protein synthesis are fundamental cellular activities re-established during germination and are the prerequisite for seedling growth. Hydrolytic enzymes are mainly secreted from the scutellar epithelial and aleurone layer and catalyze the depolymerization of starch and protein reserves in the starchy endosperm. Degradation products are absorbed by the scutellum and translocated to the developing seedling. It has been widely assumed that the mixture of enzymes released from the scutellum and from the aleurone layer contains the same enzymes in similar proportion. Subsequent events, including the mobilization of the major storage reserves, are associated with growth of the seedling. These, however, are considered to be post germination events which culminate in programmed cell death of the aleurone layer (Wang et al., 1998). Most enzymes which are involved in major reserve mobilization are apparently synthesized de novo during seed germination (Hayes and Jones, 2000). These include starch hydrolytic enzymes and some of the peptidases, which are responsible for proteolytic activity in the germinating grain (Briggs 1992). There is some evidence that mRNA is conserved in the dry embryo in a suitable condition for the support of early protein synthesis (Bewley and Black 1994). However, it is still unclear to what extent residual messenger RNAs from previous developmental processes are used transiently during early germination (Bewley 1997).

Thus morphological, physiological, biochemical molecular and enzymological analysis will provide some reliable markers to explore and exploit the different wheat genotypes in northern part of Iran for pre harvest sprouting.