



Research Article

Cluster Analysis of Wheat (*Triticum aestivum* L.) Genotypes Based Upon Response to Terminal Heat Stress

Ankur Poudel¹, Dhruva Bahadur Thapa² and Manoj Sapkota^{3*}

¹Department of Plant Breeding, Institute of Agriculture and Animal Sciences, Tribhuvan University, Nepal

²Agriculture and Botany Division, Nepal Agricultural Research Council, Lalitpur, Nepal

³International Maize and Wheat Improvement Centre (CIMMYT), Nepal

*Corresponding author's email: manoj34sapkota@gmail.com

Abstract

High temperature stress adversely affects plant physiological processes; limiting plant growth and reducing grain yield. Heat stress is often encountered due to late sowing of wheat in winter. Fifty wheat genotypes were studied for days to maturity, thousand kernel weight, grain filling duration, grain filling rate, and SPAD reading in alpha lattice design at Agriculture and Forestry University at Rampur, Chitwan, Nepal with the objective to identify superior heat stress tolerant varieties after clustering them based on their response to heat stress. All the genotypes were clustered using reduction in thousand kernel weight, heat susceptibility index for thousand kernel weight, heat susceptibility index for grain filling duration, area under SPAD retreat curve, maturity duration under normal condition, maturity duration at late sown condition, grain filling rate under normal condition and grain filling rate at late sown condition as variables and dendrogram was prepared. UPGMA revealed that these genotypes formed five distinct clusters. The resistant genotypes and susceptible genotypes formed different clusters. The member of cluster 3 was found to be tolerant to terminal heat stress where as members of cluster 2 were found most susceptible to terminal heat stress. From this study genotype BAJ #1/SUP152 was found most tolerant to terminal heat stress. The genotypes belonging to superior cluster could be considered very useful in developing heat tolerant variety and other breeding activities.

Keywords: cluster analysis; heat susceptibility index; terminal heat stress; wheat; *Triticum aestivum*

Introduction

Wheat (*Triticum aestivum* L.) is a cereal crop which belongs to family Poaceae along with other important cereals like rice, wheat, maize, barley, oat and rye. Worldwide, it is grown on nearly 217 million hectares, with a production of 653 million tons (FAOSTAT, 2013). It is a primary staple food crop for South Asia; it is grown on nearly 38 million hectares, with a production of 139.88 million tons (FAO Stat, 2013). Wheat is the world's most favored staple food. It is nutritious, easy to store and transport and can be processed into various types of food. Wheat is considered a good source of protein, minerals, B-group of vitamins and dietary fiber (Shewry, 2007; Simmonds, 1989). It is an excellent health-building food. Wheat flour is used to prepare bread, produce biscuits, confectionary products and noodles. Wheat is also used as animal feed, for ethanol production and brewing of wheat beer. Nearly 55 % of carbohydrate consumed worldwide is supplied by wheat (Gupta et al., 1999).

There are many biotic and abiotic factors responsible to decrease production of wheat. Among abiotic factors, post anthesis heat stress is very important phenomenon which is caused by increased temperature at reproductive stage of crop that decreases grain filling duration as well as thousand kernel weight thus reducing the productivity of crop. High temperature stress adversely affects plant physiological processes; limiting plant growth and reducing grain yield. At anthesis, high temperatures may result in pollen and sterility and restrict embryo development thereby reducing grain number. High temperature stress after anthesis affects the rate of grain filling, leading to reductions in grain yield (Al-Khatib and Paulsen, 1984; Tashiro and Wardlaw, 1990; Weigand and Cueller, 1981). Wheat crops requires optimum temperature of 15-18°C during grain filling period (Chowdhury and Wardlaw, 1978). Rise in temperature above optimum temperature during grain filling period decreases grain yield (Wardlaw et al., 1989) and grain weight (Wiegand and Cuellar, 1981). Heat stress is often encountered due to late sowing of wheat in winter. This is

because of late maturing rice varieties used in rice wheat cropping system which delays sowing of wheat (Hobbs and Giri, 1997). Also, farmers have to wait long to dry field after harvesting rice crop. Yield of late sown wheat crops are reduced up to 40% (Sharma and Duveiller, 2004).

Since the population of world is increasing very fast, yield of crops should be increased to feed growing population. Yield is generally determined by the genetic potential of a plant but different biotic stress from diseases, pest etc. and abiotic stress like drought, heat, cold, salt etc. tend to decrease yield. A large portion of wheat crop in hill is grown in rainfed system. Irrigated wheat is only 36.9 percent in high hills and 42.14 percent in mid hills (MoAD, 2014) Most of Nepalese farming is rainfed and sowing of winter crops is dependent on winter rain. So, lack of sufficient soil moisture delay sowing of wheat which causes exposure of plant to drought and heat after anthesis. Moreover, in Rice – Wheat based cropping system, late varieties of rice delay sowing of wheat resulting in high temperature stress during post anthesis phenology.

There is ultimate need of developing heat stress lines. Most of cultivated cultivars in Nepal are susceptible to heat stress

and considerable loss is occurring every year. Drought and post anthesis heat stress tolerant lines can be developed once the genetic variability for the traits contributing to yield under such condition are assessed in landrace as well as advanced generation lines (Baral, 2011). Genetic gain in yield potential can be done by recombining elite germplasm followed by selection. The objective of the research was to identify superior heat stress tolerant varieties after clustering them based on their response to heat stress.

Methodology

The field experiment was conducted at the research farm of Agriculture and Forestry University (AFU), Faculty of Agriculture, Rampur, Chitwan, Nepal from November 2014 to April 2014, geographically located at 27° 37' N Latitude and 84° 25' E Longitude at an altitude of 228 meters above sea level. This site contains sandy loam soil with acidic reaction. The research location is characteristics of subtropical climate. The plant materials were obtained from International Maize and Wheat Improvement Centre (CIMMYT). Among 50 genotypes used, Gautam was used as check variety. The list of genotypes included in the study is presented in Table 1.

Table 1: List of the genotypes used for the experiment

Entry No.	Genotype
1	GAUTAM
2	KACHU #1
3	QUAIU #1
4	BAJ #1
5	FRANCOLIN #1
6	KACHU/BECARD//WBLL1*2/BRAMBLING
7	QUAIU #1/SUP152
8	QUAIU #1/SUP152
9	KACHU//KIRITATI/2*TRCH
10	KIRITATI//HUW234+LR34/PRINIA/3/BAJ #1
11	ND643/2*WBLL1//VILLA JUAREZ F2009
12	SUP152/FRNCLN
13	BAJ #1/SUP152
14	WHEAR/KUKUNA/3/C80.1/3*BATAVIA//2*WBLL1/5/PRL/2*PASTOR/4/CHOIX/STAR/3/HE1/3*CNO79//2*SERI
15	CROC_1/AE.SQUARROSA (205)//BORL95/3/PRL/SARA//TSI/VEE#5/4/FRET2/5/2*DANPHE #1
16	FRET2*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ/5/2*FRNCLN
17	BAJ #1/3/2*HUW234+LR34/PRINIA//PFAU/WEAVER
18	KISKADEE #1*2//KIRITATI/2*TRCH
19	MUTUS*2/HARIL #1

Table 1: List of the genotypes used for the experiment

Entry No.	Genotype
20	BAJ #1*2/TINKIO #1
21	BAJ #1*2/ND643/2*WBLL1
22	WBLL1*2/BRAMBLING*2//BAVIS
23	PRL/2*PASTOR//WHEAR/SOKOLL
24	WHEAR/KUKUNA/3/C80.1/3*BATAVIA//2*WBLL1/4/WAXWING*2/KRONSTAD F2004
25	WHEAR/KIRITATI/3/C80.1/3*BATAVIA//2*WBLL1/4/BECARD
26	FRET2*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ/5/KIRITATI/2*TRCH/6/BAJ #1
27	FRET2*2/BRAMBLING//KIRITATI/2*TRCH/3/FRET2/TUKURU//FRET2
28	KACHU*2/SUP152
29	DANPHE/PAURAQUE #1//MUNAL #1
30	KIRITATI//2*PRL/2*PASTOR/3/CHONTE/5/PRL/2*PASTOR/4/CHOIX/STAR/3/HE1/3*CNO79//2*SERI
31	KIRITATI//HUW234+LR34/PRINIA/3/CHONTE/5/PRL/2*PASTOR/4/CHOIX/STAR/3/HE1/3*CNO79//2*SE RI
32	KIRITATI//HUW234+LR34/PRINIA/3/FRANCOLIN #1/4/BAJ #1
33	MUTUS//KIRITATI/2*TRCH/3/WHEAR/KRONSTAD F2004
34	ND643/2*WBLL1//2*KACHU
35	PAURAQ/5/KIRITATI/4/2*SERI.1B*2/3/KAUZ*2/BOW//KAUZ/6/PAURAQUE #1
36	PAURAQ/4/WHEAR/KUKUNA/3/C80.1/3*BATAVIA//2*WBLL1/5/PAURAQUE #1
37	FRANCOLIN #1*2//ND643/2*WBLL1
38	FRANCOLIN #1/CHONTE//FRNCLN
39	BAJ #1*2/KISKADEE #1
40	WHEAR/KUKUNA/3/C80.1/3*BATAVIA//2*WBLL1*2/4/KIRITATI/2*TRCH
41	TAM200/PASTOR//TOBA97/3/FRNCLN/4/WHEAR//2*PRL/2*PASTOR
42	TOB/ERA//TOB/CNO67/3/PLO/4/VEE#5/5/KAUZ/6/FRET2/7/VORB/8/MILAN/KAUZ//DHARWAR DRY/3/BAV92
43	FALCIN/AE.SQUARROSA (312)/3/THB/CEP7780//SHA4/LIRA/4/FRET2/5/DANPHE #1/11/CROC_1/AE.SQUARROSA
44	(213)//PGO/10/ATTILA*2/9/KT/BAGE//FN/U/3/BZA/4/TRM/5/ALDAN/6/SERI/7/VEE#10/8/OPATA BAVIS/NAVJ07
45	CROC_1/AE.SQUARROSA (213)//PGO/10/ATTILA*2/9/KT/BAGE//FN/U/3/BZA/4/TRM/5/ALDAN/6/SERI/7/VEE#10/8/OPATA/11/ATT ILA*2/PBW65
46	W15.92/4/PASTOR//HXL7573/2*BAU/3/WBLL1/5/DANPHE #1
47	BAVIS/3/ATTILA/BAV92//PASTOR/5/CROC_1/AE.SQUARROSA (205)//BORL95/3/PRL/SARA//TSI/VEE#5/4/FRET2
48	BABAX/LR42//BABAX/3/ER2000/4/PAURAQUE #1
49	VEE/MJI//2*TUI/3/PASTOR/4/BERKUT/5/BAVIS
50	SOKOLL/3/PASTOR//HXL7573/2*BAU/5/CROC_1/AE.SQUARROSA (205)//BORL95/3/PRL/SARA//TSI/VEE#5/4/FRET2

Field experiment was conducted in Alpha Lattice design. There were two conditions; Normal sown irrigated and Late sown irrigated (for heat stress), each condition replicated twice. Each replication comprised five blocks consisting of ten plots each. Each plot was 4 m in length 1.5 m wide. Each plot had 6 rows with spacing 25 cm between rows. Inter-block gap of 0.5 m was maintained. The dose of chemical fertilizers applied was 120:60:60 kg NPK per hectare. Irrigation was done at the three important stages; crown root initiation (CRI) stage, flowering stage and milking stage. The normal season planting was done on 22nd November 2014 and late planting was done in 7th January 2015.

Observation were taken for days to maturity, thousand kernel weight, grain filling duration, grain filling rate, and SPAD reading. The collected data were used to calculate reduction in thousand kernel weight, heat susceptibility index for thousand kernel weight, heat susceptibility index for grain filling duration, area under SPAD retreat curve (AUSRC), maturity duration under normal condition, maturity duration at late sown condition, grain filling rate under normal condition and grain filling rate at late sown

condition as variables for performing the cluster analysis. Data entry and processing was carried out using Microsoft Office Excel 2007. Multivariate analysis was carried out through Minitab 15.

Results and Discussion

All the genotypes were clustered using reduction in thousand kernel weight (RTKW), heat susceptibility index for thousand kernel weight, heat susceptibility index for grain filling duration, area under SPAD retreat curve (AUSRC), maturity duration under normal condition, maturity duration at late sown condition, grain filling rate under normal condition and grain filling rate at late sown condition as variables. The dendrogram is presented in Fig. 1. Distance between different cluster centroids of wheat genotypes is presented in Table 2. The critical examination of the dendrogram revealed five clusters with minimum of 25.91% similarity level in UPGMA clustering. The clusters were divided into two groups; Group A and Group B. Group A consisted of four clusters namely Cluster I, Cluster II, Cluster III and Cluster IV. Group B contained only one cluster: Cluster V

Table 2: Distance among the different cluster centroids of wheat genotypes under terminal heat stress at AFU, Rampur (2014/15)

	Cluster 1	Cluster 2	Cluster 3	Cluster 4	Cluster 5
Cluster 1		40.60	80.59	36.75	138.95
Cluster 2			40.05	77.33	179.52
Cluster 3				117.28	219.54
Cluster 4					102.31

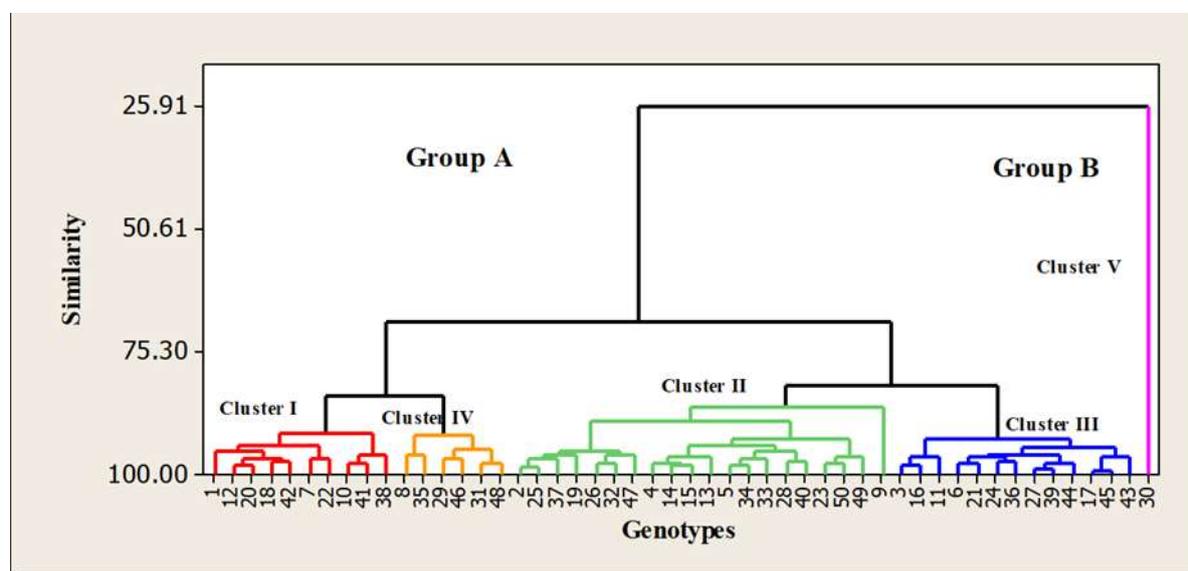


Fig. 1: UPGMA clustering of 50 wheat genotypes under terminal heat stress at AFU, Rampur (2014/15)

Cluster 1 consisted of 10 genotypes, which represents 20% of total genotypes. It includes Gautam, SUP152/FRNCLN, BAJ#1*2/TINKIO#1, KISKADEE#1*2//KIRITATI/2*TRCH, TOB/ERA//TOB/CNO67/3/PLO/4/VEE#5/5/KAUZ/6/FRET2/7/VORB/8/MILAN/KAUZ//DHARWARDRY/3/BAV92, QUAIU#1/SUP152, WBLL1*2/BRAMBLING*2//BAVIS, KIRITATI//HUW234+LR34/PRINIA/3/BAJ#1, TAM200/PASTOR//TOBA97/3/FRNCLN/4/WHEAR//2*PRL/2*PASTOR and FRANCOLIN#1/CHONTE//FRNCLN. This cluster had relatively less reduction in thousand kernel weight, intermediate value of AUSRC and days to maturity in normal sown condition but higher value for days to maturity in late sown condition. Similarly, this cluster had higher value of HSI for thousand kernel weight and grain filling duration.

Cluster 2 consisted of 20 genotypes. 40% of total genotypes fall under this cluster. It includes KACHU#1, WHEAR/KIRITATI/3/C80.1/3*BATAVIA//2*WBLL1/4/BECARD, FRANCOLIN#1*2//ND643/2*WBLL1, MUTUS*2/HARIL#1, FRET2*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ/5/KIRITATI/2*TRCH/6/BAJ#1, KIRITATI//HUW234+LR34/PRINIA/3/FRANCOLIN#1/4/BAJ#1, BAVIS/3/ATTELA/BAV92//PASTOR/5/CROC_1/AE.SQUARROSA(205)//BORL95/3/PRL/SARA//TSI/VEE#5/4/FRET2, BAJ#1, WHEAR/KUKUNA/3/C80.1/3*BATAVIA//2*WBLL1/5/PRL/2*PASTOR/4/CHOIX/STAR/3/HE1/3*CNO79//2*SERI, CROC_1/AE.SQUARROSA(205)//BORL95/3/PRL/SARA//TSI/VEE#5/4/FRET2/5/2*DANPHE#1, BAJ#1/SUP152, FRANCOLIN#1, ND643/2*WBLL1//2*KACHU;MUTUS//KIRITATI/2*TRCH/3/WHEAR/KRONSTADF2004, KACHU*2/SUP152, WHEAR/KUKUNA/3/C80.1/3*BATAVIA//2*WBLL1*2/4/KIRITATI/2*TRCH, PRL/2*PASTOR//WHEAR/SOKOLL, SOKOLL/3/PASTOR//HXL7573/2*BAU/5/CROC_1/AE.SQUARROSA(205)//BORL95/3/PRL/SARA//TSI/VEE#5/4/FRET2, VEE/MJI//2*TUI/3/PASTOR/4/BERKUT/5/BAVIS and KACHU//KIRITATI/2*TRCH. The genotypes had highest reduction in thousand kernel weight, highest value of HSI for thousand kernel weight and grain filling duration. Similarly, genotypes had least number of days to maturity for both normal and late sown condition. Similarly grain filling rate for normal sown condition was least. Genotypes had intermediate AUSRC value. The genotypes of this cluster are susceptible genotypes to heat stress.

Cluster 3 consisted of 13 genotypes. 26% of genotypes fall under this cluster. It includes QUAIU #1, FRET2*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ/5/2*FRNCLN, ND643/2*WBLL1//VILLAJUAREZ F2009, KACHU/BECARD//WBLL1*2/BRAMBLING, BAJ#1*2//ND643/2*WBLL1, WHEAR/KUKUNA/3/C80.1/3*BATAVIA//2*WBLL1/4/WAXWING*2/KRONSTADF2004, PAURAQ/4/WHEAR/KUKUNA/3/C80.1/3*BATAVIA//2*WBLL1/5/PAURAUQUE#1, FRET2*2/BRAMBLING//KIRITATI/2*TRCH/3/FRET2/TUKURU//FRET2, BAJ#1*2/KISKADEE#1, BAVIS/NAVJ07, BAJ#1/3/2*HUW234+LR34/PRINIA//PFAU/WEAVER, CROC_1/AE.SQUARROSA(213)//PGO/10/ATTELA*2/9/KT/BAGE//FN/U/3/BZA/4/TRM/5/ALDAN/6/SERI/7/VEE#10/8/OPATA/11/ATTELA*2/PBW65 and FALCIN/AE.SQUARROSA(312)/3/THB/CEP7780//SHA4/LIRA/4/FRET2/5/DANPHE#1/11/CROC_1/AE.SQUARROSA(213)//PGO/10/ATTELA*2/9/KT/BAGE//FN/U/3/BZA/4/TRM/5/ALDAN/6/SERI/7/VEE#10/8/OPATA. The genotypes had lowest reduction in thousand kernel weight, lowest value of HSI for thousand kernel weight and grain filling duration. Similarly genotypes had highest AUSRC and grain filling rate for normal sown condition. Since this cluster of genotypes had superior trait values for heat stressed condition, these genotypes may be of interest to researchers.

Cluster 4 consisted of 6 genotypes. This cluster includes 12% of total genotype. It includes QUAIU#1/SUP152, PAURAQ/5/KIRITATI/4/2*SERI.1B*2/3/KAUZ*2/BOW//KAUZ/6/PAURAUQUE#1, DANPHE/PAURAUQUE#1//MUNAL#1, W15.92/4/PASTOR//HXL7573/2*BAU/3/WBLL1/5/DANPHE#1, KIRITATI//HUW234+LR34/PRINIA/3/CHONTE/5/PRL/2*PASTOR/4/CHOIX/STAR/3/HE1/3*CNO79//2*SERI and BABAX/LR42//BABA X/3/ER2000/4/PAURAUQUE#1. The genotypes had intermediate value for reduction in thousand kernel weight, HSI for thousand kernel weight and grain filling duration. Genotypes had longer days to maturity in normal sown condition. Also, genotypes had lower grain filling rate for both normal and late sown condition.

Cluster 5 consisted of only one genotype i.e. KIRITATI//2*PRL/2*PASTOR/3/CHONTE/5/PRL/2*PASTOR/4/CHOIX/STAR/3/HE1/3*CNO79//2*SERI. This genotype had lowest AUSRC value. This genotype had intermediate value for reduction in thousand kernel weight, HSI for thousand kernel weight and grain filling duration. This genotype had relative less days to maturity for normal sown condition.

The distance among the cluster centroids are given in Table 2.

UPGMA revealed that these genotypes formed five distinct clusters. The resistant genotypes and susceptible genotypes formed different clusters. The member of cluster 3 was found to be tolerant to terminal heat stress where as members of cluster 2 were found most susceptible to terminal heat stress. From this study genotype BAJ #1/SUP152 was found most tolerant to terminal heat stress as shown by lowest reduction in kernel weight, low HSI for TKW (0.38) and GFD (0.67). The other promising genotypes are BAJ#1/3/2*HUW234+LR3 4/PRINIA//PFAU/WEAVER,CROC_1/AE.SQUARROSA (205)//BORL95/3/PRL/SARA//TSI/VEE#5/4/FRET2/5/2* DANPHE#1,CROC_1/AE.SQUARROSA(213)//PGO/10/ ATTLA*2/9/KT/BAGE//FN/U/3/BZA/4/TRM/5/ALDAN /6/SERI/7/VEE#10/8/OPATA/11/ATTLA*2/PBW65 which had lower reduction in kernel weight and lower HSI value for TKW and GFD.

Since above mentioned promising genotypes have been identified from this heat stress experiment on wheat, further research can be done in this direction by selecting superior genotypes. These genotypes could be very useful in developing heat tolerant variety.

Acknowledgements

The authors are highly indebted to CIMMYT, Mexico and Agriculture Botany Division, Khumaltar for the provision of the research materials. The authors would also like to express their sincere gratitude to Directorate of Research and Extension, Agriculture and Forestry University for providing the research grant.

References

Al-Khatib K and Paulsen GM (1984). Mode of high temperature injury to wheat during grain development. *Physiologia plantarum* **61**(3): 363-368. DOI: [10.1111/j.1399-3054.1984.tb06341.x](https://doi.org/10.1111/j.1399-3054.1984.tb06341.x)

Baral K (2011) Variability study of Nepalese wheat landraces and advanced breeding lines for drought and terminal heat stress. (Master's thesis, Institute of Agriculture and Animal Sciences, Rampur, Chitwan)

Chowdhury SI and Wardlaw IF (1978). The effect of temperature on kernel development in cereals. *Crop and Pasture Science* **29**(2): 205-223. DOI: [10.1071/AR9780205](https://doi.org/10.1071/AR9780205)

FAOSTAT database (2013) Food and Agriculture Organization of the United Nations, Retrieved from faostat.fao.org/default.aspx

Gupta PK, Varshney RK, Sharma PC and Ramesh B (1999) Molecular markers and their applications in wheat breeding. *Plant breeding* **118**(5): 369-390. DOI: [10.1046/j.1439-0523.1999.00401.x](https://doi.org/10.1046/j.1439-0523.1999.00401.x)

Hobbs PR and Giri GS (1997) *Reduced and zero-tillage options for establishment of wheat after rice in South Asia*. Springer Netherlands. pp. 455-465. DOI: [10.1007/978-94-011-4896-2_60](https://doi.org/10.1007/978-94-011-4896-2_60)

MoAD (2014) *Statistical Information on Nepalese Agriculture*. Agri business promotion and statistical division, Agri statistic section, Singhdurbar, Kathmandu.

Sharma RC and Duveiller E (2004) Effect of Helminthosporium leaf blight on performance of timely and late-seeded wheat under optimal and stressed levels of soil fertility and moisture. *Field Crops Research* **89**(2): 205-218. DOI: [10.1016/j.fcr.2004.02.002](https://doi.org/10.1016/j.fcr.2004.02.002)

Shewry PR (2007) Improving the protein content and composition of cereal grain. *Journal of Cereal Science*, **46**(3), 239-250. DOI: [10.1016/j.jcs.2007.06.006](https://doi.org/10.1016/j.jcs.2007.06.006)

Simmonds DH (1989) Inherent quality factors in wheat. *Wheat and Wheat Quality in Australia*. Melbourne: CSIRO 31-61.

Tashiro T and Wardlaw IF (1990) The effect of high temperature at different stages of ripening on grain set, grain weight and grain dimensions in the semi-dwarf wheat 'Banks'. *Annals of Botany*, **65**(1), 51-61. DOI: [10.1093/oxfordjournals.aob.a087908](https://doi.org/10.1093/oxfordjournals.aob.a087908)

Wardlaw IF, Dawson IA, Munibi P and Fewster R (1989) The tolerance of wheat to high temperatures during reproductive growth. I. Survey procedures and general response patterns. *Crop and Pasture Science* **40**(1): 1-13. DOI: [10.1071/AR9890001](https://doi.org/10.1071/AR9890001)

Weigand CL and Cueller JA (1981). Duration of grain filling and kernel weight of wheat as affected by temperature. *Crop Science* **21**: 95-101. DOI: [10.2135/cropsci1981.0011183X001100010027x](https://doi.org/10.2135/cropsci1981.0011183X001100010027x)