

FLAG LEAF STOMATAL FREQUENCY AND ITS INTERRELATIONSHIP WITH YIELD AND YIELD COMPONENTS IN WHEAT (*TRITICUM AESTIVUM* L.)

MEHARUN-NISA KHANUM YOUSUFZAI, K.A. SIDDIQUI AND A.Q. SOOMRO

*Nuclear Institute of Agriculture TandoJam -70060, Pakistan
Department of Botany, University of Sindh, Jamshoro, Pakistan.*

Abstract

Three tall (C591, H-68 and H-23-42) and 5 semi-dwarf (Pavon, Pak-81, Mehran, Kohinoor and Sarsabz) cultivars of bread wheat (*Triticum aestivum* L.) were grown in the experimental field of Nuclear Institute of Agriculture (NIA) TandoJam for 3 consecutive years to study the stomatal frequency in the flag leaf and its interrelationships with yield and yield components. The cultivars showed significant differences with respect to stomatal frequency, yield and yield components. Higher stomatal frequency in flag leaf was positively associated with yield and yield components. The cultivar Sarsabz having significant superiority in yield and yield components also showed significantly ($p \leq 0.05$) higher stomatal frequency at adaxial and abaxial surface of flag leaf as compared to other cultivars. Stomatal frequency of flag leaf thus provided a new selection criterion for screening of high yielding varieties of bread wheat.

Introduction

Two important plant processes viz., photosynthesis and transpiration are influenced by stomatal frequency in many crop plants (Inamdar *et al.*, 1991). Variation in stomatal frequency of cultivars have also been reported within and between *Triticum* species (Tear *et al.*, 1971). The stomatal frequency in adaxial surface of leaves has been found higher than abaxial surface in alfalfa (Cole & Dobrenz, 1971), creeping grass (Shearman & Beard, 1972) and on the flag leaf of wheat (Geok-Yong & Dunn, 1975). Transgressive segregation for high and low stomatal frequency was observed in F_2 generation in bread wheat on the basis of selection of new variants (Bhagwat & Bhatia, 1993). Hecial (1971) and Yoshida (1976) also found that stomatal frequency can be used as an indicator of photosynthetic capacity. Stomatal frequency along with pubescence had served as micro-morphological marker in inheritance studies in interspecific and intergeneric hybridization of barley (Rajendra *et al.*, 1978). Studies of structure, organographic distribution and taxonomic importance of stomata are still in progress in different part of the world (Leela & Rao, 1996; Sahin, 1998; Yousufzai, 1999). The present research provides a new experimental evidence on the flag leaf stomatal frequency in relation to the yield and yield components of bread wheat (*Triticum aestivum* L.) which has remained the central theme of the self-sufficiency programme in Pakistan.

Materials and Methods

Three tall (C591, H-68 and H-23-42) and 5 semi-dwarf (Pavon, Pak-81, Mehran, Kohinoor and Sarsabz) hexaploid bread wheat (*T. aestivum*) cultivars were sown N.I.A., TandoJam. The experiment was laid out in split plot design with 4 replications each with four rows of 1m length planted 30cm apart for every cultivars. At heading stage 10 flag leaves were randomly selected from each replication of every genotypes for the study of stomatal frequency at adaxial and abaxial surface of flag leaf. One cm middle portion of

flag leaves were fixed in a solution of 50% Ethyl alcohol (95%), 5% glacial acetic acid, 10% formaldehyde and 35% distilled water (by volume) (FAA) and stored in a refrigerator at 3°C. The samples 15 days after the fixation were boiled in 70% alcohol ($C_2H_5 OH$) for 20 minutes and then in lactic acid for 15 to 20 minutes. The leaf portion now softened clear and transparent in 90% lactic acid was than mounted with 90% lactic acid for stomatal studies according to the method of Rajendra *et al.*, (1971).

Stomatal frequency at adaxial and abaxial surface of flag on per mm^2 were recorded at 6 positions and each side of the mid rib of flag leaf at 10x magnification of Carl Zeiss Light Microscope with 5mm microscopic field.

The crop was left to maturity and the data was recorded on plant height, first, second and third internode length, spike length, tillers per plant and ear head characteristics. All the data were statistically analyzed using Fisher's analysis of variance technique at the least significant differences (LSD) and 5% probability level was used to test the significance of means following the procedure by Steel & Torrie (1980). Correlation coefficients were worked out by Dewey & Lue (1950) method.

Results and Discussion

Stomatal frequency at adaxial and abaxial surface of flag leaf were significantly ($p \leq 0.05$) different among the cultivars of bread wheat. Higher yielding cultivar Sarsabz showed significantly ($p \leq 0.05$) higher stomatal frequency at adaxial and abaxial surface of flag leaf as compared to other cultivars. The cultivar H-23-42 had lower stomatal frequency as compared to others (Table 1).

The mean value on morphological traits, yield and yield components are provided in Tables 1&2. The tall cultivars C591 and H-68 had the plant height higher than 130cm, with internode length also higher than semi-dwarf cultivars. The short height cultivar Kohinoor possessed the lower values for 1st, 2nd and 3rd internode lengths. All the components of yield showed higher in cultivars Sarsabz which contributed significantly for increase in the grain yield of the variety (Table 2). The cultivar Kohinoor possessed the lowest value for all the characters and the differences were statistically significant.

There existed positive relationship of stomatal frequency with all the yield components viz., tillers per plant, number of spikelets, florets, grain and grain weight per spike, 1000 grain weight, grain yield kg per hectare and most of the 'r' value were found to be statistically significant (Table 3). Leaf stomatal frequency showed different response to height. It was negatively correlated in adaxial surface, whereas a strong positive correlation in abaxial surface. Higher stomatal frequency was observed in adaxial than abaxial surface in all the cultivars of bread wheat included in the present investigation as reported earlier by Cole & Dobrenz (1970) in alfalfa and Teare *et al.*, (1971) in wheat.

From the present studies it is obvious that stomatal frequency plays an important role in enhancing the yield and yield components. Teare (1971) also reported the relation between stomatal frequency and grain yield. Stomatal frequency at abaxial surface correlated positively ($p \leq 0.05$) with plant height but at adaxial surface was negative and non-significant. Third internode length showed non significant negative correlation with stomatal frequency at adaxial and abaxial surface as well as first internode length exhibited non significant positive correlation with stomatal frequency at adaxial surface, whereas there was negative correlation in abaxial surface. Second internode length showed non significant positive correlation with stomatal frequency at adaxial and abaxial surface. It has already been demonstrated that plant height has a significant positive correlation with stomatal frequency in *Secale cereale* (Rajendra *et al.*, 1977) and may be considered a new parameter of potential use in inheritance studies. In the present study, this relationship is confirmed in *T. aestivum* at abaxial surface. The character of

stomata appears to be useful in the elucidation of relationships at intergeneric level (Pack & Jun, 1995). Studies of structure, organographic distribution and taxonomic importance of stomata are still in progress in different parts of the world (Leela & Rao, 1996; Sahin, 1998; Yousufzai, 1999).

Table 1. Flag leaf stomatal frequency mm², plant height, internode length and tillers number in different wheat cultivars.

Cultivars	Stomatal frequency		Plant height (cm)	1 st Internode length (cm)	2 nd Internode length (cm)	3 rd Internode length (cm)	Tillers/plant (cm)
	Adaxial surface	Abaxial surface					
C-591	14.5c	12.0c	134.95a	44.42.ab	25.90ab	17.90b	11.20bc
H-68	13.0c	10.0d	130.95b	42.35ab	28.73ab	20.40a	12.25b
H-23-42	10.0d	7.0c	103.72d	30.85e	24.23c	18.20b	12.70b
Mehran	20.0b	15.0b	104.35d	41.20bc	19.92c	12.95c	13.30b
Pavon	15.5c	13.0c	102.10de	36.95d	19.75de	12.95c	10.35c
Pak-81	16.0c	13.0c	113.10c	39.17cd	21.05d	13.60c	11.75bc
Kohinoor	15.0c	12.0c	91.63f	32.53e	19.05e	12.80cd	11.66d
Sarsabz	25.0a	22.0a	99.75e	40.13bc	40.13bc	11.57d	15.50a

Values denoted by different letters are significantly different (p≤0.05) from each other.

Table 2. Morphological traits, yield and yield components of different bread wheat (*Triticum aestivum* L.) cultivars.

Cultivars	Spike length (cm)	Spikelets/spike	Florets/spike	Grain/spike	Grain weight/spike(g)	1000 grain weight (g)	Grain yield kg/ha
C-591	9.65d	22.25c	83.70c	58.45d	2.53de	44.50e	750b
H-68	9.40d	21.70d	67.90d	55.15e	2.47de	46.00c	830b
H-23-42	6.47e	22.05e	82.70c	57.90d	2.65cd	44.05d	650d
Mehran	12.65b	23.60bc	102.40b	73.35a	3.34a	47.70b	680d
Pavon	11.23bc	23.85b	100.00b	69.45b	3.15ab	46.10c	700c
Pak-81	13.88b	25.10a	107.95ab	67.33c	2.53de	44.30c	525e
Kohinoor	10.75c	18.95d	108.05ab	67.55bc	2.47de	40.70e	610d
Sarsabz	16.50a	25.60a	112.30a	69.80b	3.75a	50.60a	1330a

Value denoted by different letters are significantly (p≤ 0.05) different from each other.

Table 3. Correlation coefficients among stomatal frequency, morphological traits, yield and yield components.

Morphological traits/yield & yield components	Stomatal frequency/mm ²	
	Adaxial surface	Abaxial surface
Plant height (cm)	-0. 3020	0.8706**
1st internode length (cm)	0.0384	-0.3506
2nd internode length(cm)	0.0467	0.4057
3rd internode length(cm)	-0.2587	-0.2078
Tiller/plant	0.6248**	0.6052**
Spike length (cm)	0.2008**	0.1919
Spikelets /spike	0.7283**	0.7047**
Florets /spike	0.6188**	0.8373**
Grain/spike	0.7286**	0.7699**
Grain weight (g)	0.6135**	0.8489**
1000 grain weight (g)	0.9820**	0.8489**
Grain yield kg/ha	0.9600**	0.7093**

*Significantly different at (p≤0.05), **Significantly different at (p≤0.01)

The present experimental results have led to the conclusion that wheat cultivars vary for stomatal frequency as they exist. The presence of positive relationships between stomatal frequency, yield and yield components is very useful for screening of higher

yielding genotypes of wheat, because counting stomata is much easier than selection of higher yielding varieties of wheat under field condition for longer periods. More over new cross combination can be established between low and high frequency stomata lines to obtained high frequency of flag leaf thus provides a new selection criterion to plant breeder for screening of higher yielding genotypes of bread wheat.

References

- Bhagwat, S.G. and C.R. Bhatia. 1993. Selection for flag leaf stomatal frequency in bread wheat. *Plant. Breed. J.*, 110(2): 129-136.
- Cole, D.F. and A.K. Dobrenz. 1970. Stomatal density in alfalfa (*Medicago sativa* L.). *Crop Sci.*, 10: 61-62.
- Dewey, D.R. and K.H. Lu. 1959. A correlation and path coefficient analysis components crested wheat grass seed production. *Agron. J.*, 51:515-518.
- Geok-Yong, T. and B. M. Dunn. 1975. Stomatal length, frequency and distribution in *Bromus inermis* Leyss. *Crop Sci.*, 15(3): 283-286.
- Heichel, G.H. 1971. Stomatal movements, frequency and resistance in two maize varieties differing in photosynthetic capacity. *J. Exp.Bot.*, 22: 644-649.
- Inamdar, J.A., R.C. Patel and J.S.S. Mohan. 1991. Structure and ontogeny of stomata in vegetative and floral organ of some *Apocynaceae*. *Feddes Report.*, 102(5-6): 335-344.
- Leela, M. and S.R.S. Rao. 1996. Structure, distribution, development and taxonomic importance of stomata in some *Ipomoea* L. (*Convolvulaceae*). *Beitrag zur Biologie der Pflanzen.*, 68(3): 329-342.
- Pack, D.Y. and E.S. Jun. 1995. Stomatal density, size and morphological characteristic in Orchids. *J. Korean. Soc. Hort. Sci.*, 36(6): 851-862.
- Rajendra, B.R., K.A. Mujeeb and L.S. Bates. 1977. A modified technique for stomatal study of the leaf epidermis in *Triticeae*. *Stain Tech.*, 52: 9-12.
- Rajendra, B.R., K.A. Mujeeb and L.S. Bates. 1978. On the correlation of height on stomatal frequency and size in two lines of *Hordeum vulgare* L. *Environ. Exp. Bot.*, 18: 117-119.
- Rajendra, B.R., K.A. Mujeeb and L.S. Bates. 1978. Relationship between 2x *Hordeum* spp., *Secale* spp., and 2x, 4x, 6x *Triticum* spp., for stomatal frequency and size distribution. *Environ. Exp. Bot.*, 18:33-37.
- Sahin, N.F. 1998. Morphological, anatomical and physiological studies of *Galanthus Ikariae* Baker and *G. rizehensis* Stern (*Amaryllidaceae* L.). *Pak. J.Bot.*, 30(1): 117-131.
- Shearman, R.C. and J.B. Beard. 1972. Stomatal density and distribution in *Agrostis* as influenced by species, cultivars and leaf blade surface and position. *Crop Sci.*, 12: 822-823.
- Steel, R.G. and J.H. Torrie. 1980. Analysis of variance. *Principles and Procedures of Statistics*. McGraw-Hill Book. Co, Tokyo Japan, 137-223.
- Teare, I.D. 1971. Stomatal frequency, distribution and guard cell length on 771 lines of *Triticum* and their relation to grain yield, *Wheat Newsl.*, 17: 82.
- Teare, I.D., C.J. Peterson and A.G. Law. 1971. The stomatal frequency of flag leaf in cultivars of *Triticum aestivum* and other *Triticum* species. *Crop Sci.*, 11: 496-491.
- Yoshida, T. 1976. On stomatal frequency in barley I. Relationship between stomatal frequency and photosynthesis. *Japan. J. Breed.*, 26(2): 130-136.
- Yousufzai, M.N. 1999. *Taxonomic variation for endomorphic traits in tribe Triticeae*. Ph.D. Thesis., University of Sindh, Jamshoro & Nuclear Institute of Agriculture TandoJam, Pakistan. pp. 231.

(Received for publication 4 April 2003)