



Exploring phenotypic expression to augment quality of *Triticum aestivum* and improve selection efficiency

D. Mohan* and R. K. Gupta

ICAR-Indian Institute of Wheat and Barley Research, Karnal 132 001, Haryana

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Abstract

Contribution of height, heading days and grain ripening period was examined in quality parameters of 50 Indian bread wheat (*Triticum aestivum* L.) cultivars by regression analysis. Their collective contribution was highly significant in end-product quality. These traits also contributed in gluten and protein contents, extraction rate, test weight, grain appearance and yellow pigments but sedimentation volume, gluten index and grain hardness remained unfazed. Heading days was the most prominent factor as it contributed positively in flour recovery, protein and gluten contents, test weight, grain appearance, *chapati* score and negatively in biscuit spread factor and yellow pigments. Steady grain ripening enhanced physical grain properties whereas reverse was true for protein content and cookies. Height reduced bran content and improved *chapati* score but quality of the bread was hampered. Study demonstrated that phenotypic indicator could supplement quality of end-products and make the selection procedure simpler and improvised. In hard grain varieties, plant height and early flowering suited *chapati* quality. Reduced height enhanced benefits incurred from gluten content and its quality in bread making. Grain softness and reduced gluten content was more rewarding when supported by quick grain ripening.

Key words: Bread wheat cultivars, end-product quality, morphological trait, selection efficiency, wheat grain quality

Introduction

Environment plays a vital role in defining yield potential and grain quality of bread wheat (*Triticum aestivum* L.). Manifestation of such influences is seen through agronomic expressions i.e. height, days to flowering, crop duration, ripening period and grain weight. Accordingly, physiologically efficient genotypes possess agronomic traits suitable for grain formation, growth and development (De Vita et al. 2007). In wheat,

these parameters have been examined mainly in reference to grain yield. Few attempts had covered couple of quality parameters but such investigations were restricted to moisture/temperature stress or alterations in planting time (Guttieri et al. 2001; Gooding et al. 2003). Understanding influence of the environment, genotype and their interactions on end-use quality had also been attempted in India (Mohan and Gupta 2011) and other parts of the world (Zhang et al. 2004; Souza et al. 2004) but *per se* contribution of yield determinants, irrespective of environmental or genetic backgrounds, has yet to be properly understood. When agronomic and physiological attributes are known to make significant changes in grain yield, their effect on grain quality also needs to be addressed with same intensity as both aspects relate to grain development processes. Among influenced agronomic traits, rate and duration of grain filling had been touched in some studies (Wardlaw and Moncur 1995; Spiertz et al. 2006; Mladenove et al. 2012) focussing only protein accumulation in wheat grains. History of wheat research in India indicates that varieties known for good *chapati* were tall in pre-green revolution era but with advent of semi-dwarf Mexican varieties, product quality drifted as sink capacity and biomass partitioning was diverted to increase grain number (Khush 2001). Decline in bread quality during green revolution era and thereafter had been reported from Iran (Khodarahmi et al. 2010). In India also, varieties having edge to C 306 in *chapati* quality could not be developed after green revolution. Therefore while developing high yielding wheat varieties, there is need to revisit yield contributing attributes to simultaneously structure the end-use quality.

*Corresponding author's e-mail: devmohan@gmail.com

Wheat varieties in India are evaluated, released and recommended on the basis five mega zones carved according to the regional climatic variations, crop expression and disease spread. Besides yield, the national wheat research programme of India (AICW&BIP) examines test entries not only for yield related components like height, heading and crop duration and grain weight but also for quality parameters related with marketability, processing and end-usages. Wide differences had been reported between different zones in quality and yield determinants (Mohan et al. 2011). Parallelism between quality and yield components had also been reported amongst different climates in which test materials were evaluated under AICW&BIP (Mohan and Gupta 2015^a). Huge information generated in this national wheat research programme has been utilized as benchmark in this study to examine whether i) morphological parameters influence grain and end-use quality, ii) such expression can serve as handy tool in augmenting grain quality and iii) benefit can be accrued by involving phenotypic indicators in ventures targeted for end-usages. Quality improvement is a tricky business and requires lot of instrumentation. New paradigms are needed to make development of product specific varieties simple, easy and widely applicable. If phenotypic indicators could bridge this gap, it would be of immense interest to the wheat breeders.

Materials and methods

Experimental material

Study material included released bread wheat (*Triticum aestivum* L.) cultivars used as checks in trials of AICW&BIP and covered five zones i.e. northern hills zone (NHZ), north-western plains zone (NWPZ), north-eastern plains zone (NEPZ), central zone (CZ) and peninsular zone (PZ). Environment for wheat growth is different in each zone. NHZ that covers hills and foothills of Himalayas has long winter and low temperature. NWPZ and NEPZ represent the Indo-Gangetic plains in which NWPZ, labelled as most productive wheat land of India, has the most soothing wheat growth environment whereas adjoined NEPZ has shorter winter and the climate is also humid. Crop in CZ often faces soil moisture stress as climate is hot and dry. Peninsula in down south i.e., PZ has similar temperature and soil moisture conditions but humidity is not that severe. Investigation included only those checks which were repeated in the advance varietal trials at least for years during the crop season 2010-15. Care was taken to include leading varieties

of all production condition as the genotypes under investigation included 17 timely-sown varieties, 18 late-sown varieties and 15 varieties meant for moisture stress conditions (Table 1). Study material involved 13 varieties from NWPZ and 10 each from NHZ and NEPZ. CZ and PZ region was represented by 9 and 8 varieties respectively.

Table 1. Bread wheat varieties examined in the study material

Zone	Name of the cultivars
NHZ	HS 277, HS 295, HS 490, HS 507, VL 804, VL 829, VL 892, VL 907, HPW 251 and HPW 349
NWPZ	PBW 175, PBW 373, PBW 590, PBW 644, WH 1021, WH 1080, WH 1105, DBW 17, DPW 621-50, C 306, HD 2967, HD 3043 and HD 3059
NEPZ	K 307, K 2087, HD 2733, HD 2985, HD 2888, NW 2036, HI 1563, DBW 14, DBW 39 and C 306
CZ	LOK 1, GW 322, HI 1500, HI 1544, MP 3336, MP 4010, MP 3288, HD 2932 and HD 2864
PZ	NIAW 34, NIAW 1415, NI 5439, MACS 6222, MACS 6478, GW 322 and RAJ 4083

Observations and statistical analysis

Attributes investigated among phenotypic traits were plant height, days to heading, grain filling period (GFP) and total maturity duration. Mean performance of a genotype was characterized on the basis of 5-8 trial conducting sites of concerning zone. Samples received from three locations of each zone were analysed at Karnal. Besides end-products (*chapati* quality score, bread loaf volume, bread quality score and biscuit spread factor), quality analysis included ten grain parameters i.e. grain appearance score, grain hardness index, sedimentation volume, grain protein at 14% grain moisture (GPC), wet gluten content, gluten index, test weight, extraction rate, yellow pigments content and GLU 1 score. AACC (2000) method was applied to examine processing and milling quality. Grain appearance score was a subjective test to collectively rate size, shape, soundness, colour and texture out total score 10. Conventional approach was adopted to derive GLU 1 score (Payne et al. 1981) and *chapati* quality (Rao et al. 1986). Single kernel characterization system 4100 was used to measure grain hardness whereas Quadrumat Senior mill was used for flour recovery.

Data is presented as mean performance of the varieties during the study period and range represents the varietal differences. Variability in each trait has been reported as coefficient of variation (CV %). For regression analysis, data was standardised and analysis tool provided in excel programme of the computer was used. Multiple regression analysis was applied to calculate the regression coefficients and beta values. Backward regression analysis was applied to determine key determinants and significance was tested at P 0.05.

Results and discussion

Varietal diversity

Range in the study material was large for all the field traits under study (Table 2). In phenology, differences

Table 2. Variability in yield and quality attributes

Parameter	Mean	Range	CV
Morphological traits			
Height (cm)	90	65-117	11.8
Heading days	87	54-147	29.0
Grain filling days	44	35-52	8.96
Total duration (days)	131	93-195	20.7
Grain characteristics			
Grain appearance score	6.2	5.4-7.0	5.92
Test weight (kg/hl)	79.1	74-83	2.52
Grain protein content (%)	11.6	9.6-13.5	8.26
Wet gluten content (%)	29	22-36	11.3
Sedimentation volume (ml)	43	34-55	13.4
Gluten index (%)	59	46-77	13.1
Grain hardness index	73	29-94	11.6
Flour extraction rate (%)	68.8	62-72	3.85
Yellow pigments (ppm)	3.28	2.3-4.6	16.1
GLU-1 score	-	4-10	20.0
End-product quality			
<i>Chapati</i> quality score	7.68	6.97-8.09	3.78
Bread loaf volume	552	493-586	4.80
Bread quality score	6.52	5.06-7.74	10.8
Biscuit spread factor	7.40	6.48-10.8	8.91

CV = Coefficient of variation

were large (CV >20%) in days to heading and total maturity duration and low (CV 9%) in grain filling duration. In comparison, variations in another prominent parameter of vegetative phase i.e. height

was 12%. Since cool and long winter prolongs flowering and promotes tillering (Ortiz et al. 2008); differences in heading days were highly prominent. It is well established that heading and GFP are highly variable yield components, especially when abiotic variations are large (Bordes et al. 2008). They not only put grain yield at risk through grain number and grain weight but GPC also gets affected in view of negative association between grain number and grain size (Oury et al. 2007). Diversity was of lower magnitude in flour recovery and physical grain characteristics like test weight and grain appearance score (CV: 2.5 to 5.9%). For all other grain attributes, CV was in the range 10-20%. Though range appeared good in quality of the end-products, CV was below 5% in *chapati* score and bread loaf volume. For bread quality score and biscuit spread factor, variations were little higher. GLU 1 score also expressed large differences as rang in the study material was 4 to 10.

Contribution in grain quality

Combined effect of height, heading days and GFP on grain properties was assessed through multiple regression analysis. Highly significant coefficients were observed in extraction rate, GPC, gluten content, test weight, grain appearance score and yellow pigment contents (Table 3). R² value suggested that more than 50% variation in these grain traits was influenced by phenotypic field expression of the plant. Influence of the morphological traits was insignificant on grain hardness, sedimentation value and gluten strength. It shows that phenotypic traits exert relevance in certain grain quality parameters like flour recovery, GPC, gluten content, test weight and appearance of the grain. Grain attribute related to hardness, gluten strength and gluten quality remains unaffected by the field expression of the crop.

Beta value of the component traits revealed that heading days was the most prominent field parameter as it affected flour recovery, protein and gluten contents, test weight, grain appearance and yellow pigment contents. Except yellow pigments, its contribution was negative in all other traits. It shows that early flowering enhances milling yield, GPC, gluten content and physical grain properties whereas yellow pigments content is enhanced when heading gets delayed. Contribution of the post-anthesis period was highly significant in GPC, test weight and grain appearance score. Long GFP enhanced physical properties of the grain whereas quick grain ripening benefitted GPC. Impact of height was positive and highly significant in extraction rate. It shows that height

Table 3. Contribution of significant morphological traits

Parameter	Regression Coefficient	R ² value	Beta value of individual traits		
			Plant height	Heading days	Grain ripening days
Grain characteristics					
Extraction rate	0.820***	0.673	0.34**	-1.00***	0.09
Wet gluten	0.818***	0.669	-0.06	-0.76***	-0.05
Grain protein	0.764***	0.583	-0.05	-0.53***	-0.34**
Test weight	0.730***	0.533	-0.07	-0.51***	0.77***
Grain appearance	0.727***	0.528	-0.01	-0.59***	0.72***
Yellow pigments	0.517**	0.267	0.22	0.44**	-0.27
Hardness index	0.327	0.107	0.22	-0.37	0.20
Sedimentation value	0.236	0.056	-0.12	-0.15	0.02
Gluten index	0.180	0.033	-0.10	0.22	-0.09
End-product quality					
<i>Chapati</i> quality	0.714***	0.510	0.34**	-0.90***	0.26*
Bread loaf volume	0.642***	0.413	-0.40**	-0.17	-0.21
Bread quality score	0.671***	0.450	-0.42**	-0.21	-0.18
Biscuit spread factor	0.574***	0.330	-0.05	0.64***	-0.36**

*, **, *** represent significance at P 0.05, 0.01 and <0.0001, respectively

helps in reducing bran content of wheat flour. Overall, it was evident that short pre-anthesis and long-post anthesis periods improve test weight and grain appearance. Early heading and quick grain ripening benefit GPC whereas early flowering is the key component for good gluten content. Early heading in tall plants is beneficial in recovering good flour yield.

Significant association of early anthesis, long GFP, high grain growth rate with high protein concentration had been reported in winter wheats by Mou et al. (1993). As reported by Asseng and Milroy (2006), GFP had highly with grain protein ($r = -0.57^{**}$) in this study also. Although gluten is derived from grain protein and the factors affecting them should have been the same but the key factors differed in this study. According to Mohan and Gupta (2015^b), protein-gluten ratio varies according to genotypes and the environments, therefore some deviations in the key determinants of protein and gluten was obvious. Effect of post-anthesis period on kernel mass and test weight is well recorded in literature (Stone and Nicolas 1995; Mladenove et al. 2012). Relevance of grain growth rate and GFP had been emphasized in bread wheat under irrigated as well as drought conditions (Sanjari et al. 2011). Mohan and Gupta (2015^a) examined relevance of physiological efficiency derived on the

basis of these agronomic traits in 503 released and pre-released genotypes tested by AICW&BIP during the period 2005-14 and reported their significant contribution in wheat grain quality. Significant contribution of heading and height in flour recovery (Mohan and Gupta, 2014) and strongly adverse relationship between iron and plant height (Morgounov et al. 2007) has been highlighted in wheat. Mohan and Gupta (2015^c) had also reported that short duration wheats draw more accumulation of micronutrients and same is true for grain protein as well.

Contribution in quality of the end-products

Since bran and gluten contents were influenced by the morphological traits, their impact on the end-products was obvious. Total contribution of three field traits was highly significant in each product i.e. bread, *chapati* and biscuit (Table 3). Beta values of the component traits underlined importance of all three morphological traits in *chapati* quality. Early heading, more plant height and steady grain ripening were helpful in improvement of *chapati* score. It was a pointer that short height and delayed heading is detrimental to quality of this domestic product. Height contributed negatively in bread quality. Long pre-anthesis and short post-anthesis growth phases were noted good for quality of the cookies. In contrast to *chapati* score,

biscuit quality needed delayed heading and quick grain ripening.

The study highlighted relevance of taller varieties in *chapati* score and short varieties for superior bread making. Traditionally, good *chapati* making Indian cultivars, whether it's the pre-green revolution era (Khush 2001) or thereafter especially under rainfed situation, have carried good height as well (Mohan et al. 2013). On the contrary, high tillering Mexican varieties are not very tall. Since shorter wheat varieties spearhead better yield advantage under irrigated conditions, it is opinionated that modern varieties have been put to disadvantage for good *chapati* standards. Screening for bread quality, might be better rewarding in dwarf high yielding genotypes.

Grain development during reproductive phase depends upon assimilates accumulated during vegetative period. In the study material, range in heading days was very large (54-147) in comparison to grain filling days (35-51). Heading-ripening ratio (HRR) in the test varieties indicated that for each grain filling day, certain varieties had support of 3.1 days whereas in few others it dropped down to 1.2 days. When HRR was plotted against quality score of the three products (Fig. 1), differential response was observed. Trend-line indicated sharp rise in biscuit quality with increase in HRR. It shows that pre-anthesis period should be longer to derive extra advantage in quality of the cookies. Declining trend was noted in case of bread and *chapati* and the relationship was stronger in *chapati* in comparison to bread. In indicates that pre-anthesis period should not be large when focus lie quality of the bread or *chapati*. Very strong negative association between HRR and flour yield was also observed in this study (Fig. 1). Flour recovery increased at faster rate when bigger pre-anthesis period

supported the grain ripening process (R^2 : 0.60). Relevance of pre-anthesis period in flour recovery had been highlighted earlier by Mohan and Gupta (2014).

Opportunities in varietal development

Relevance of crop morphology in wheat grain quality prompted to explore whether these field indicators can be instrumental in selection of genotypes suited for good end-product quality. To workout added advantage of yield related traits, contributions of ten grain quality components was compared with the morphological parameters. Effort was also made to derive extra advantage when quality traits are combined with the yield parameters (Tables 4). Regression coefficient

Table 4. Regression statistics - combined effect of quality and morphological traits on end-products

Product	Contributors	Multiple R	R ² value
<i>Chapati</i> quality score	Quality traits	0.785	0.616
	Phenotypic traits	0.714	0.510
	Quality + phenotypic traits	0.801	0.642
Bread quality score	Quality traits	0.846	0.716
	Phenotypic traits	0.671	0.450
	Quality + phenotypic traits	0.874	0.763
Biscuit spread factor	Quality traits	0.892	0.795
	Phenotypic traits	0.604	0.365
	Quality + phenotypic traits	0.945	0.892

derived from the quality traits was higher than the morphological markers. It shows that morphological traits are no substitute for grain quality components as such. When quality and field traits were combined,

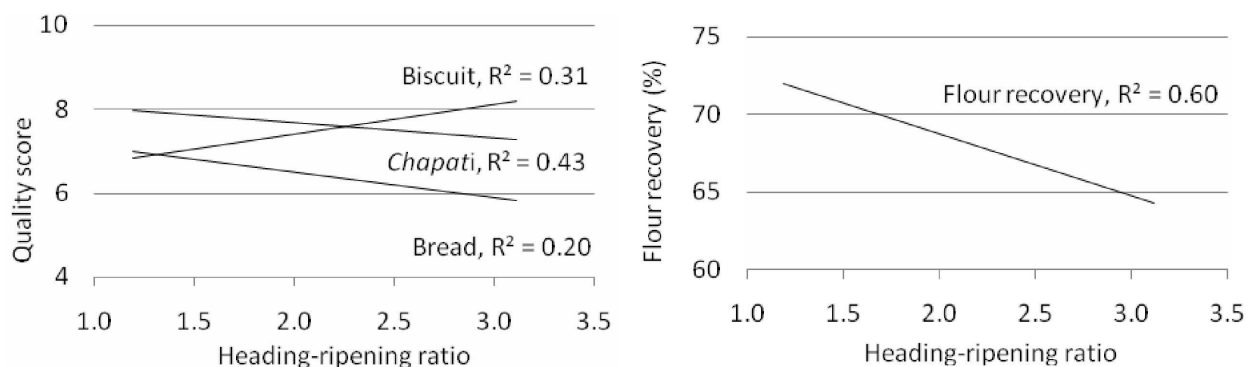


Fig. 1. Trend-line between product quality and heading-ripening ratio

the combined effect increased the R^2 value. This advantage varied in different products and was higher in biscuit spread factor (0.795 to 892) in comparison to *chapati* or bread quality. It was evident that scope exists to involve morphological traits in selection of varieties superior in end-product quality. It could make the selection procedure easy and better rewarding.

In this investigation, backward regression analysis was applied to identify the key components as similar approach was used by Trethowan et al. (2001) to workout contribution of important quality components in wheat quality. Key components amongst ten grain quality parameters were similar to the earlier reports presented in Indian wheats (Mohan and Gupta 2013). In *chapati* quality, contribution of the key quality traits i.e. extraction rate and grain appearance score was similar as obtained with one grain parameter i.e. grain hardness, and three phenotypic indicators namely height, maturity period and days to heading (Table 5). Biggest contributor

chapati making. Just grain hardness is to be adjudged in the laboratory and rest of the screening can be exercised in the field. It's a big advantage when selection is based just on single grain characteristic as determining extraction rate is a tedious process and grain appearance score can also vary from person to person. Both grain properties can be replaced with grain hardness and a few field observations. In this study, heading had recorded very strong negative correlation with gluten content (-0.81^{**}) and extraction rate (-0.76^{**}). Early flowering therefore helped in enhancing gluten content and reducing bran. It is obvious therefore that additional grain hardness, plant height and reduced pre-anthesis period can be useful in enhancing *chapati* quality. Normally heading gets delayed in taller varieties as both are negatively correlated ($r: 0.60^{**}$), so a combination of tallness with early flowering is to be searched by the breeders to improvise *chapati* score.

In bread quality; gluten content, gluten index and

Table 5. Beta value of significant quality and morphological components

Parameters	Quality traits			Quality and morphological traits		
	<i>Chapati</i> quality	Bread quality	Biscuit quality	<i>Chapati</i> quality	Bread quality	Biscuit quality
R^2 value	0.589 ^{***}	0.654 ^{***}	0.760 ^{***}	0.583 ^{***}	0.706 ^{***}	0.821 ^{***}
Key quality traits						
Grain appearance	0.28 ^{**}	-	-	-	-	-
Extraction rate	0.63 ^{***}	-	-	-	-	-
Wet gluten content	-	0.66 ^{***}	-0.50^{***}	-	0.51 ^{***}	-0.62^{***}
Gluten index	-	0.24 ^{**}	-	-	0.22 ^{**}	-
GLU 1 score	-	0.36 ^{***}	-	-	0.32 ^{***}	-
Grain hardness	-	-	-0.61^{***}	0.29 ^{**}	-	-0.54^{***}
Key agronomic traits						
Plant height				0.28 [*]	-0.28^{**}	-
Heading days				-	-	-
Grain filling period				0.32 ^{**}	-	-0.28^{***}
Total maturity duration				-0.85^{***}	-	-

amongst four key factors was the short crop duration. In genotypes of short maturity duration, steady grain ripening, plant height and grain hardness enhanced *chapati* quality. If crop duration is ignored, there could be small decline in the contribution as R^2 value was reduced from 0.58 to 0.55 in this study. It paves way for a handy approach to select varieties suitable for

GLU 1 score were the prominent grain quality traits. As reported by Pena (2008) and Mohan and Gupta (2013), content and quality of gluten is paramount in quality of the bread. This investigation suggested that when plant height was added to these parameters, the R^2 value increased from 0.65 to 0.71. Contribution of height was negative in the study material. Height

was negatively correlated with GPC ($r: -0.51^{**}$) in the Indian varieties under study and protein is an important constituent of bread quality. Since gluten strength and gluten quality were not influenced by the field traits, there was no alternate for these grain attributes. It suggests that when selection for gluten content and gluten quality is exercised amongst the shorter genotypes, selection efficiency gets improvised. In biscuit spread factor, key grain quality factors are the grain softness and low gluten content. Contribution of these two traits was increased in this investigation when GFP was also included as R^2 increased from 0.76 to 0.82. Impact of GFP was negative which suggests that quick grain ripening can enhance the selection efficiency based on grain softness and gluten content.

The investigation has clearly spelled that even though yield and quality in wheat is influenced by genetic make-up, cultivation practices, surrounding environment and the magnitude of abiotic pressures (Zhang et al. 2004; Mohan and Gupta 2015^d); developments taking place during the vegetative and reproductive phases cannot be ignored as manifestation of such process is routed through phenotypic and physiological attributes (Mohan and Gupta 2015^a). Since quality and yield are two manifestations of the processes related to grain development, this study makes it clear that components affecting yield are also crucial in articulating quality of wheat grain and the end-products. Except gluten strength, gluten quality and grain hardness, all other grain quality parameters are affected by the morphological parameters. Expression of some non-grain parameters can also be utilized to make the selection procedure simple and improvised. Relevance of field related non-grain attributes like height and phenology opens new vistas in augmentation of wheat quality.

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