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Assessment of genetic variability in F_4 segregating generation for Nitrogen use efficiency and related traits in wheat (*Triticum aestivum* (L.))

Shikha Dubey, SA Desai and Suma Biradar

Abstract

An experiment was laid out during *Rabi* 2020-21 to evaluate the F_4 generations belonging to the crosses UAS BW 13356 × HD 2967 and WH 1022 × K 9107 in wheat for Nitrogen use efficiency and related traits. The F_4 progenies were derived from the parents with varied response for N use efficiency. The F_4 progenies showed significant genotypic variance for most of the characters studied. The Nitrogen use efficiency and related traits showed significant genotypic variance. The genotypic coefficient of variation (GCV), phenotypic coefficient of variation (PCV), heritability and genetic advance as percent of mean was found to be high for N use efficiency and related traits. Hence, existence of sufficient genetic variability shows that breeding for Nitrogen use efficiency is expected to be successful in wheat.

Keywords: Nitrogen use efficiency, F_4 generation, phenotypic coefficient of variation, genotypic coefficient of variation, wheat

Introduction

Wheat (*Triticum aestivum* L.) is a staple food crop of the world population, consumed by billions of people in the world, of which India is not an exception (Lumpkin, 2011). Wheat was one of the first domesticated food crops and for 8,000 years has been the basic staple food for major civilizations in Europe, Asia and North Africa. Today wheat is grown on more than 220 m. ha with over 750 m. tonnes of production. It continues to be counted among the 'big three' cereal crops as the most leading important food grain source for human beings (Anon., 2020) [1].

Nitrogen plays an important role in the biochemical processes of plants, including protein, DNA, RNA, enzymes and chlorophylls (Andrews and Lea, 2013) [2]. Considering the important role of nitrogen in plant life cycle, nitrogen application is essential in the form of chemical fertilizer. N fertilizer is immensely required for enhancing food production. Numerous studies indicated that N fertilization can increase both wheat grain yield and grain protein content (Gorjanovic and Balalic, 2008) [4]. It is absorbed from the soil mainly in the form of nitrate, ammonia/ammonium or urea (CONH2). Insufficient N severely affects the yield of crops, whereas oversupply has no effect on yield however contributes significantly to N pollution. Excess inorganic nitrogen in freshwater can cause algal blooms that in turn result in eutrophication of aquatic ecosystems (Vitouesk *et al*., 2009) [9].

In the present scenario, developing wheat lines which give high yields with minimum N inputs is therefore a priority. A major breakthrough is expected to improve upon selection for higher N use efficiency by plant breeding. The identification of potential donor genotypes with high nitrogen use efficiency to be used for further development of nitrogen use efficient wheat varieties will greatly facilitate the resource poor farmers to enhance the productivity of their fields. These varieties when incorporated into functional and sustainable farming systems will help close the gap between actual and economically realistic yields (Huggins and Pan, 2003) [6]. With this premises, the present study was carried out to study the genetic variability among F_4 progenies of two crosses for N use efficiency and related traits.

Material and Methods

Sixty-one F_4 progenies from the cross UAS BW 13356 × HD 2967 and one hundred seventy-three F_4 progenies from the cross WH 1022 × K 9107 were used for this experiment. The progenies were selected from superior F_3 progenies for N use efficiency and related traits. The experiment was laid out in augmented design using the parents and additional seven checks repeated in each block. Nitrogenous fertiliser (urea) was applied at the rate of 50 kg ha^{-1}. Twenty-five kg ha^{-1} of N was applied at the time of sowing and the rest 25 kg ha^{-1} was applied at the initiation of stem extension. The amount of N to be available per plot was calculated by subtracting the amount of N available from Diammonium Phosphate. Soil N content was estimated using the alkaline potassium permanganate method before sowing the crop and was found to be 72 kg ha^{-1} (Motsara, 2015) [8]. Observations were recorded on ten plants per progeny row for the following phenological and N use efficiency traits-Days to fifty percent flowering (DAF), Flag leaf length (FLL), Flag leaf width (FLW), Chlorophyll content at anthesis (SPAD2), Chlorophyll content at grain filling (SPAD3) plant height (PH), spike length (SL), spikelets per spike (SPS), thousand grain weight (TGW), grain yield plant^{-1} (GYP), number of productive tillers (NTP), above ground biomass^{-1} (AGBP), harvest index (HI), grain nitrogen content (GNC), straw N content (SNC), nitrogen harvest index (NHI), grain protein content (GPC), total N uptake (NUP), nitrogen uptake efficiency (NUpE), nitrogen utilisation efficiency (NUtE) and Nitrogen use efficiency (NUE).

Statistical Analysis

Analysis of variance for the progenies of the two crosses was done in augmented design as per Federer (1961). Major descriptive indices including mean, standard error, and standard deviation, minimum and maximum were estimated for all the phenological and N use efficiency traits. The results of descriptive statistics of the two crosses are outlined in Tables 3 and 4 respectively. Genetic variability parameters including phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), broad sense heritability (h^2BS) and genetic advance as a percent of mean (GAM) were assessed for all the traits showing genotypic variance among the progenies in the F_4 generations of the crosses UAS BW 13356 × HD 2967 and WH 1022 × K 9107. The results for the same are summarised in Tables 5 and Table 6 respectively.

Results

Analysis of variance in F_4 generation of the three crosses

For the cross UAS BW 13356 × HD 2967, the differences among the F_4 progenies were significant for most traits except flag leaf length, chlorophyll content at anthesis, chlorophyll content at grain filling, harvest index, grain N content, grain protein content, total N uptake and NUpE as indicated in the ANOVA table. The differences among the checks were significant for all the traits evaluated. The differences among the progenies and checks were significant for all the traits except chlorophyll content at anthesis, plant height, harvest index, thousand grain weight, grain N content and grain protein content. For the WH 1022 × K 9107, the differences among progenies as well as between progenies and checks were significant for all the traits studied.

Descriptive statistics

The summary statistics estimated for the two crosses is shown in Table 3. The average number of days to fifty percent flowering was higher in WH 1022 X K 9107 (62.29 days). For all other phenological traits, WH 1022 X K 9107 showed higher mean value including, Flag leaf length, Flag leaf width, Chlorophyll content at anthesis, Chlorophyll content at grain filling, plant height, spike length, spikelets per spike, thousand grain weight, grain yield per plant, number of productive tillers, For N use efficiency and related traits, the two crosses displayed comparable mean values. However, range for most N use efficiency traits was greater in WH 1022 X K 9107. In UAS BW 13356 x HD 2967, the highest N use efficiency reported was 24.96 g g^{-1} N. In WH 1022 X K 9107, the maximum N use efficiency value was 22.34 g g^{-1} N.

Genetic variability parameters

The genetic variability parameters for the two crosses are shown in Table 4. Low GCV, PCV and low to moderate GAM was observed in UAS BW 13356 x HD 2967 for days to fifty percent flowering, SPAD 3, plant height, no. of spikelets per spike, thousand grain weight, grain N content, grain protein content and nitrogen harvest index. Moderate to high GCV, PCV and GAM was observed for flag leaf length, flag leaf width, spike length, harvest index, thousand grain weight, Straw N content, and NUtE. High GCV, PCV and GA was observed for number of productive tillers, AGB, GY, total N uptake, total N uptake and N use efficiency in this cross. The broad sense heritability was found to be high for all the traits except flag leaf width, total N uptake and total N uptake.

In the cross WH 1022 x K 9107, low GCV, PCV and low to moderate GAM was observed for days to fifty percent flowering, flag leaf width, plant height, thousand grain weight, grain N content, grain protein content and nitrogen harvest index. Moderate to high GCV, PCV and GAM was observed for flag leaf length, chlorophyll content at anthesis, chlorophyll content at grain filling, spike length, no. of spikelets per spike, harvest index and N utilisation efficiency. High GCV, PCV and high GA was recorded for number of productive tillers, above ground biomass per plant, grain yield per plant, Straw N content, total N uptake, total N uptake and N use efficiency. The broad sense heritability was found to be high for all the traits studied.

Discussion

Genetic variability is fundamental to the success of any breeding programme. Hence, any breeding programme is initiated with the assessment of genetic variability between genotypes. This is followed by making crosses in different combinations between selected genotypes. The best cross combinations are then selected to constitute a population to further obtain transgressive segregants. In the present study two F_4 populations obtained from F_3 progenies selected for Nitrogen use efficiency were evaluated for genetic variability and their response for N use efficiency and related traits. The descriptive statistics showed that UAS BW 13356 x HD 2967 was earlier flowering than WH 1022 X K 9107. The N use efficiency values observed for the two crosses were comparable. However, UAS BW 13356 x HD 2967 showed higher average value or total N uptake and WH 1022 X K 9107 showed higher average value for NUtE (Cormier *et al.*, 2013, Guttieri *et al.*, 2017) [3, 5] Significant genotypic variance was observed in both the crosses for all N use efficiency and related traits as observed from the analysis pf variance. However, the quantification of genetic variability showed that some phenological traits like days to fifty percent flowering, chlorophyll content at grain filling, plant height, no. of spikelets per spike, thousand grain weight, grain N content, grain protein content and nitrogen harvest index showed low

PCV and GCV as well as low heritability. Similar results were observed by Mahalaxmi *et al.* (2019) [7] in F_2 generation in wheat. Hence, it is advised that the plan for genetic improvement should consider the genetic variability and heritability of individual trait.

Table 1: Analysis of variance in Augmented design for F_4 generation of the cross UAS BW 13356 x HD 2967 for phenological and Nitrogen use efficiency traits at N_{50} during Rabi 2020-21

Source	Df	DAF	FL	FW	SPAD 2	SPAD 3	PH	SL	SPS	NTP	AGB
Block (ignoring Treatments)	1	1.44	15.86	0.05**	4.58	19.71	0.21	33.29**	5.46**	2.29*	67.97
Treatment (eliminating Blocks)	69	9.75**	9.28	0.02**	19.67	21.23	52.91*	1.37**	4.14**	2.54**	102.6**
Treatment: Check	8	29.76**	22.65*	0.01**	31.55*	49.54*	91.33**	1.13**	8.53**	5.63**	142.9**
Treatment: Test	60	4.34*	8.39	0.02**	18.37	17.86	47.91*	1.92**	3.58**	2.07**	48.74*
Treatment: Test vs. Check	1	175.98**	51.23*	0.05**	6.09	88.44*	21.17	1.42*	3.54*	7.96**	3052.33**
Residuals	8	1.18	4.75	0.0018	9.74	8.66	14.78	0.15	0.45	0.22	15.28

Source	Df	GY	HI	TW	GNC	SNC	GPC	NHI	NUP	NUP	NUtE	NUE
Block (ignoring Treatments)	1	9.42	0.00014	85.03**	0.005	0.01*	0.15	0.0012**	0.01	0.01	2.13	11.76
Treatment (eliminating Blocks)	69	13.22**	0.0039	24.3**	0.03	0.01**	1.07	0.0013**	0.01**	0.01**	15.68**	16.5**
Treatment: Check	8	17.02**	0.01*	49.68**	0.02*	0.01**	1.72*	0.00085**	0.02**	0.02**	23.68**	21.24**
Treatment: Test	60	26.35**	0.0036	4 22.32**	0.03	0.01**	1.06	0.0013**	0.01	0.01	14.53**	27.93**
Treatment: Test vs. Check	1	397.62**	0.000463	3.31	0.001	0.04**	0.05	0.0045**	0.37**	0.47**	22.41**	496.39**
Residuals	8	2.46	0.0015	4.35	0.01	0.0008	0.37	1.00E-04	0.0022	0.0027	1.85	3.07

Table 2: Analysis of variance in Augmented design for F_4 generation of the cross WH 1022 x K 9107 for phenological and Nitrogen use efficiency traits at N_{50} during Rabi 2020-21

Source	Df	DAF	FL	FW	SPAD 2	SPAD 3	PH	SL	SPS	NTP	AGB
Block (ignoring Treatments)	2	56.5**	46.4*	0.08**	386.67**	355.52**	217.02**	23.19**	8.77**	0.99	13.55**
Treatment (eliminating Blocks)	181	7.79**	97.5**	0.01**	34.75**	35.65**	58.69**	1.43**	5.66**	2.53**	7.69**
Treatment: Check	8	37.18**	252.72**	0.06**	56.36**	46.1*	179.91**	1.67**	8.79**	7.44**	24.18**
Treatment: Test	172	6.77**	57.92**	0.01**	35.85**	38.1**	53.99**	1.66**	5.43**	2.21**	4.11**
Treatment: Test vs. Check	1	59.86**	5665.73**	0.2**	443.79**	220.66**	327.85**	5.9**	29.01**	20.21**	17.49**
Residuals	16	1.48	11.01	0.002	2.45	11.91	8.38	0.09	0.88	0.53	1.07

Source	Df	GY	HI	TW	GNC	SNC	GPC	NHI	NUP	NUP	NUtE	NUE
Block (ignoring Treatments)	2	7.86*	0.001	50.78**	0.02	0.01**	0.73	0.0014**	0.01**	0.01**	0.95	9.81*
Treatment (eliminating Blocks)	181	9.32**	0.01**	22.52**	0.04**	0.01**	1.48**	0.0012**	0.01**	0.01**	23.95**	11.63**
Treatment: Check	8	19.44**	0.01**	63.93**	0.05*	0.02**	1.55*	0.0022**	0.03**	0.03**	25.84*	24.27**
Treatment: Test	172	5.89**	0.01*	19.38**	0.04**	0.01**	1.45**	0.0011**	0.0047**	0.01**	23.73**	7.35**
Treatment: Test vs. Check	1	516.56**	0.01*	310.96**	0.23**	0.08**	7.85**	0.02**	0.57**	0.71**	35.26*	644.88**
Residuals	16	2.08	0.0019	4.58	0.01	0.00031	0.45	0.00019	0.001	0.0013	7.14	2.59

Table 3: Summary statistics for phenological and Nitrogen use efficiency traits in F_4 generation of the cross UAS BW 13356 x HD 2967

Trait	Mean	Std. Error	Std. deviation	Minimum	Maximum	Range
Days to 50 percent flowering	60.61	0.31	2.63	55.94	68.5	12.56
Flag leaf length (cm)	15.56	0.35	2.94	9.72	22.38	12.66
Flag leaf width (cm)	1.26	0.02	0.15	0.99	1.56	0.57
Chlorophyll content at anthesis	50.02	0.5	4.22	40.85	58.65	17.8
Chlorophyll content at grain filling	40.89	0.56	4.67	31.06	51.54	20.48
Plant height (cm)	76.19	0.84	7.01	48.58	92.07	43.49
Spike length (cm)	9.6	0.18	1.52	7.29	13.25	5.96
Number of spikelets per spike	17.78	0.23	1.91	13.1	22.4	9.3
Number of productive tillers plant^{-1}	5.12	0.18	1.51	2.84	10.16	7.32
Above ground biomass plant^{-1}	23.06	1.06	8.91	9.64	47.2	37.56
Grain yield plant^{-1}	8.64	0.39	3.25	3.49	18.24	14.75
Harvest Index	0.38	0.01	0.06	0.17	0.49	0.32
Thousand grain weight (g)	44.4	0.62	5.16	31.13	54	22.87
Grain nitrogen content (%)	2	0.02	0.18	1.61	2.33	0.72
Straw nitrogen content (%)	0.47	0.01	0.1	0.29	0.64	0.35
Grain protein content (%)	11.61	0.12	1.03	9.34	13.54	4.2
Nitrogen harvest index	0.81	0	0.03	0.74	0.87	0.13
Total Nitrogen uptake (g)	0.24	0.01	0.1	0.08	0.55	0.47
Nitrogen uptake efficiency (g g^{-1} N)	0.27	0.01	0.11	0.09	0.62	0.53
Nitrogen utilisation efficiency (g g^{-1} N)	32.51	0.45	3.78	21.88	41.26	19.38
Nitrogen use efficiency (g g^{-1} N)	9.65	0.43	3.63	3.9	20.38	16.48

Table 4: Summary statistics for phenological and Nitrogen use efficiency traits in F_4 generations of the cross WH 1022 x K 9107

Trait	Mean	Std. Error	Std. deviation	Minimum	Maximum	Range
Days to 50 percent flowering	62.29	0.2	2.72	54.81	67.33	12.52
Flag leaf length (cm)	13.8	0.17	2.28	9.85	22.04	12.19
Flag leaf width (cm)	1.25	0.01	0.11	1.01	1.54	0.53
Chlorophyll content at anthesis	49.1	0.45	6.01	36.28	67.43	31.15
Chlorophyll content at grain filling	38.99	0.47	6.3	24.3	56.02	31.72
Plant height (cm)	71.07	0.55	7.43	48.81	93.02	44.21
Spike length (cm)	9.35	0.1	1.3	5.57	12.52	6.95
Number of spikelets per spike	16.86	0.18	2.45	8.65	22.7	14.05
Number of productive tillers plant^{-1}	4.94	0.11	1.53	1.75	11.29	9.54
Above ground biomass plant^{-1}	24.75	0.63	8.53	10.06	51	40.94
Grain yield plant^{-1}	8.55	0.21	2.81	3.13	19.99	16.86
Harvest Index	0.36	0.01	0.07	0.14	0.49	0.35
Thousand grain weight (g)	38.88	0.33	4.47	28.37	52.55	24.18
Grain nitrogen content (%)	2.1	0.02	0.21	1.71	2.52	0.81
Straw nitrogen content (%)	0.47	0.01	0.1	0.28	0.63	0.35
Grain protein content (%)	12.22	0.09	1.2	9.94	14.62	4.68
Nitrogen harvest index	0.82	0	0.03	0.75	0.89	0.14
Total Nitrogen uptake (g)	0.25	0.01	0.08	0.1	0.61	0.51
Nitrogen uptake efficiency (g g^{-1} N)	0.28	0.01	0.09	0.11	0.68	0.57
Nitrogen utilisation efficiency (g g^{-1} N)	33.89	0.36	4.85	20.72	45.5	24.78
Nitrogen use efficiency (g g^{-1} N)	9.56	0.23	3.13	3.5	22.34	18.84

Table 5: Estimates of genetic variability parameters for phenological and Nitrogen use efficiency traits F_4 generation of the crosses UAS BW 13356 x HD 2967 and WH 1022 x K 9107

Cross	UAS BW 13356 x HD 2967				WH 1022 x K 9107			
Trait	GCV	PCV	H	GAM	GCV	PCV	H	GAM
Days to 50 percent flowering	2.93	3.44	72.79	5.16	3.69	4.18	78.13	6.73
Flag leaf length (cm)	12.26	18.61	43.39	16.66	12.62	14.68	73.89	22.38
Flag leaf width (cm)	12.07	12.54	92.74	23.98	7.97	8.74	83.22	15
Chlorophyll content at anthesis	5.87	8.57	46.97	8.3	11.77	12.19	93.17	23.44
Chlorophyll content at grain filling	7.42	10.33	51.49	10.98	13.13	15.83	68.73	22.45
Plant height (cm)	7.55	9.08	69.15	12.96	9.5	10.34	84.48	18.02
Spike length (cm)	13.89	14.46	92.33	27.54	13.36	13.76	94.32	26.77
Number of spikelets per spike	9.95	10.65	87.38	19.19	12.65	13.82	83.84	23.9
Number of productive tillers plant^{-1}	26.49	28.06	89.12	51.6	26.23	30.11	75.89	47.14
Above ground biomass plant^{-1}	25.08	30.27	68.66	42.88	27.67	30.75	80.99	51.38
Grain yield plant^{-1}	22.85	29.18	61.3	36.9	22.83	28.37	64.72	37.88
Harvest Index	12.02	15.79	57.93	18.88	16.22	20.23	64.29	26.83
Thousand grain weight (g)	9.55	10.64	80.52	17.67	9.9	11.32	76.39	17.84
Grain nitrogen content (%)	7.13	8.85	64.79	11.83	8.15	9.83	68.61	13.92
Straw nitrogen content (%)	20.03	20.92	91.69	39.58	20.06	20.42	96.54	40.66
Grain protein content (%)	7.13	8.85	64.79	11.83	8.15	9.83	68.61	13.92
Nitrogen harvest index	4.27	4.44	92.27	8.45	3.73	4.11	82.73	7.01
Total Nitrogen uptake (g)	22.63	29.88	57.32	35.34	23.92	26.98	78.61	43.75
Nitrogen uptake efficiency (g g^{-1} N)	22.63	29.88	57.32	35.34	23.92	26.98	78.61	43.75
Nitrogen utilisation efficiency (g g^{-1}N)	10.95	11.72	87.24	21.1	12.02	14.37	69.89	20.72
Nitrogen use efficiency (g g^{-1} N)	22.85	29.18	61.3	36.9	22.83	28.37	64.72	37.88

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