RESEARCH ARTICLE



Synthetic hexaploid wheat as a source of variation for the traits specific to conservation agriculture

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Abstract

Breeding wheat for a better root system and compatibility under conservation agriculture (CA) practices have the potential to increase and sustain grain yield production under changing climate scenarios. Due to the practical challenges in evaluating the "hidden half" of crop plants under field conditions, screening of genotypes under hydroponic environments will help to get around these difficulties. The present study used hydroponic growing conditions to analyze the root traits of 55 Synthetic Hexaploid Wheat (SHW) genotypes and 19 bread wheat cultivars. These cultivars are developed over the past 100 years for India's North Western Plain Zone. For root architectural traits, a substantial diversity was found. Due to the positive correlation with dry root weight, root length (RLsc and RLrs), root volume, root surface area, average diameter, and dry shoot weight can be effectively combined. Further analysis of a subset of 34 SHW lines revealed that SHW lines like SYN2, SYN28, and SYN13 have greater root lengths, root volumes, root surface area, and number of root tips when compared to mega varieties like PBW 343, HD 2967, HD 3086, and other popular varieties. Many SHW lines displayed thicker and longer coleoptiles than released varieties. In CA, coleoptile thickness is equally important as coleoptile length because it helps the crop emerge well from the high residue load. The findings of this study clearly indicate that useful genetic variation for the traits important for CA exists in synthetic wheat and should be explored for bread wheat improvement.

Keywords: Conservation agriculture, synthetic hexaploid wheat, root traits, correlation, genetic variation

Introduction

Bread wheat (Triticum aestivum L., 2n=6x=42) is one of the world's most important staple food crops. Due to the limited gene flow caused by the few instances of natural hybridization that occurred during the evolution of wheat, the genetic diversity from the "D genome" is not completely represented in the current wheat cultivars (Li et al. 2018). Preferential inter-crossing of elite germplasm lines for the development of higher-yielding cultivars has further narrowed the genetic diversity by the depletion of a few alleles from a more diverse gene pool (Cox 1997). This narrow genetic diversity limits the availability of novel genes or QTLs useful for wheat improvement. To sustain wheat production under changing climatic conditions and emerging biotic and abiotic stresses, more useful genetic diversity needs to be incorporated into the wheat breeding programs. Consistent loss of genetic diversity has been observed when improved genomes are compared to domesticated landrace or non-commercial genomes and again when domesticated genomes are compared to the genomes of wild relatives (Moyers et al. 2018). This ranges up to a 77% loss of genetic diversity between wild and improved tomato populations (Lin et al. 2014). One approach to broadening the genetic base of present-day wheat cultivars is to introgress novel genes/QTLs from progenitor species such as *Ae. tauschii* through synthetic hexaploid wheat (SHW) production (<u>Dreisigacker</u> et al. 2008; <u>Ogbonnaya</u> et al. 2013).

The SHW, commonly designated as primary synthetic wheat, is created by crossing modern durum wheat and *A*. *tauschii*. The primary synthetics are of poor agronomic value, difficult to thresh, generally tall, low yielding, and frequently have poor quality, but they carry novel genetic diversity for a range of biotic and abiotic stresses (Trethowan and Mujeeb-

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Kazi 2008). Novel genes for economically important diseases like all three rust and Karnal bunt have been identified in SHW and efforts have been made to deploy them in elite genetic backgrounds (Thomas et al. 2010; Periyannan et al. 2013; Singh et al. 2000; Kazi et al. 2012). Many of the synthetics and synthetic-derived lines have shown good tolerance to important abiotic stresses like drought (Trethowan and Kazi 2008), heat (Ullah et al. 2021), and salinity (Nakayama et al. 2022). Thus, the introgression of novel and useful alleles for biotic and abiotic stress tolerance and improved end-use/ nutritional quality from SHWs into elite wheat germplasm is a major objective in many pre-breeding and germplasm development programs (Ogbonnaya et al. 2013).

The wider adaptation provided by increasing the genetic diversity of bread wheat via SHW provides a way to enhance productivity gains in the face of climate change scenarios. It is believed that SHWs may be an excellent donor for the traits important in conservation agriculture, such as better emergence due to longer coleoptiles, larger seeds, stronger early vigour for the best establishment, deeper and widespread root systems, and increased nutrient-use efficiency. One of the unexplored areas of research is the root system architectural traits in SHW and their potential to contribute to improved productivity via tolerance to moisture stress and lodging (Gaikwad et al. 2022). The conservation agriculture production environment is different from conventional tillage in many ways with respect to agronomy and varietal requirements. Therefore, the varieties developed through selection under conventional tillage may or may not necessarily perform equally well under CA (Yadav et al. 2017). Genetic variation for the traits important in CA could be explored from SHW lines. Therefore, in the present study, emphasis has been given to SHW lines to find genetic variation for the traits unique to CA and suggest ways to use them in the breeding programme.

Materials and methods

Plant materials and growth conditions

The study included 55 SHW lines and 19 bread wheat varieties, including some of the most widely grown mega cultivars released in the last 100 years for India's North Western Plain Zone and some newly developed varieties, to investigate genetic variation for traits important in CA projections. At the National Phytotron facility, ICAR-Indian Agricultural Research Institute, New Delhi, the experiment was set up in hydroponics using a Randomized Block Design (RBD) with two replicates over the 2019-20 and 2020-21 crop seasons. The cultivars that are used in the experiment are listed along with their parentage (Supplementary Table S1)

Hydroponic screening

The experimental material was grown under 10/14 hours

of light and dark timing using an automatic timer, having a 25/22°C day/night temperature with a light intensity of 300 µmol m⁻² s⁻¹ using cool fluorescent lamps and relative humidity of 65-70% was maintained (Ayalew et al. 2014). Each genotype's seeds were surface-sterilized for 2 minutes with 1% sodium hypochlorite, then thoroughly washed with distilled water and placed on towel paper in a seed incubator for germination. After a week, the hydroponic system received seedlings from each of the 55 SHW lines and 19 bread wheat varieties. A hydroponic system is made out of an 18 liter plastic tray with 8 mm diameter holes in an acrylic sheet at the top. Two seedlings were wrapped in cotton plugs and moved to each hole of the lid in such a way that the roots of the seedlings remained immersed in the plastic tray's hydroponic solutions. The experiment was duplicated twice more. 1M HCl or 1M KOH were used to keep the pH of the hydroponic solution around 6-6.5. The aquarium air pump was used to continuously aerate the solution. In the hydroponic system, the nutrient solution comprised 2 mM CaNO₃, 10 mM KNO₃, 0.4 mM NH₄NO₃, 0.1 mM KH₂PO₄, 2 mM MgSO₄, 0.1 mM Fe-EDTA, 1.5 mM Cacl₂, 2 mM MnCl₂, 3 mM ZnSO₄, 12.5 mM H3BO₃, 0.1 mM Na₂MoO₃, 0.5. The solution was changed weekly to preserve the nutrients' regular state. Plants and roots were carefully removed after 8 weeks of growth under hydroponic circumstances to record data on seedlings, including coleoptile length (CL), coleoptile thickness (CT), seedling root length (SRL), and seedling shoot length (SSL). Root length measured with a scale-RL(sc), root length measured in a root scanner- RL(rs), the surface area of the roots (SAR), root volume (VOL), the average diameter (AD), number of root tips (NTIPS), root forks (FORKS), and root cross (CROSS) were measured on 8-week-old plants using the root scanning system WinRHIZO (Regent Instrument Inc.). Data on dry shoot weight (DSW) and dry root weight (DRW) were also reordered on 8-week-old over-dried plants at 60°C for 5 days. The geometric parameters of the grains viz., grain length (GL), grain width (GW), grain thickness (GT), the surface area of the grain (SAG), length-breadth ratio (LBR), and thousand-grain weight (TGW) were recorded on field-grown genotypes.

On the basis of phenotypic acceptability and high value for the traits under study, a subset of 34 SHW lines was constituted and was evaluated along with 6 checks viz., HD 2967, HD 3086, HDCSW 18, HD 3117, C 306, and PBW 343 for all the traits following the same experimental procedure.

Statistical analysis

Analysis of variance (ANOVA) was analyzed as a randomized block design for hydroponic as well as for field experiments with STAR Software developed by IRRI. K mean Clustering analysis was done using Euclidean distance and Karl Pearson correlation coefficient analyses with the R software. Biplot of 'which-won-where' was plotted by PB Tools software developed by IRRI.

Results

Assessment of genetic variability

The ANOVA revealed statistically significant differences among the genotypes for all the traits studied (Supplementary Table S2). A quite noticeable range was observed for most of the traits, specifically the root traits viz., RL(rs) was 304.32 to 2129.17 cm, for SAR it was 42.06 to 307.04, VOL (0.4 to 3.68), NTIPS (1180 to 7623) and DRW (0.006 to 0.394 gm). The coleoptile length (2.5 to 6.5 cm) and grain-related parameters like SAG (51.29 to 122.10) and TGW (32.10 to 57.01) also showed a good a magnitude of variation. The data root traits generated by repeating the hydroponics experiment for the subset genotypes again showed highly significant differences among the genotypes (Supplementary Table 3). The range for the root and grainrelated traits also showed a similar pattern of variation.

Relationships among the traits

The correlation worked out among the different traits studied in the first year is presented in Figure 1. The results showed that RL(rs) has a highly significant correlation with the associated trait-like SAR (0.94***), VL (0.82***), NTIPS (0.79***), and FORKS (0.94***). It also correlates significantly with DRW (0.35***) (Fig. 1). The DRW showed a significant and positive correlation with SAR, VL, AD, GL, SAG, and DSW. The coefficient of determination (R²) was highly significant (R²- 0.5) for DRW and DSW for both SHW and released varieties. In the case of SHW, the regression of DRW with other root traits like RL(rs), VL, and SAR was highly significant. However, it was non-significant in the case of released varieties.

Classification of genotypes

To analyze the pattern in the data, mean Euclidean distance grouped 74 genotypes into five major clusters with cluster III accommodating maximum (28) genotypes followed by cluster I (21), cluster V (16), cluster II (5), and cluster IV (4), respectively (Fig. 2). Most of the mega varieties were part of cluster III which is the largest among the all cluster. This

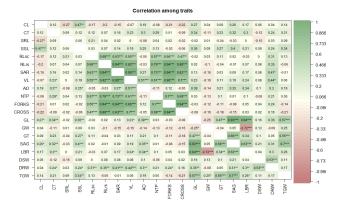


Fig. 1. Pearson correlation among the SHW and released varieties for the traits studied

cluster has the lowest cluster mean values for DRW and grain-related parameters like TGW, LBR, GL, and SAG (Table 1). Genotypes grouped under cluster V had lower cluster mean values for most of the trait. Four SHW lines viz., SYN 4, SYN 08, SYN 30, and SYN 15 in cluster IV have the highest trait values for grain-related traits. Genotypes in cluster II viz., SYN 2, SYN 28, SYN 29, HDCSW 18, and C 306 showed very high root length measured by root scanner (Figure 4) and associated traits (Table 1). SYN 2 recorded the highest root length of 2138.17 cm followed by popular drought-tolerant variety C 306 (1866.62 cm) and conservation agriculturespecific variety HDCSW 18 (1779.42 cm). SYN 28 and SYN 29 also recorded high root length. Genotypes in cluster I viz., SYN 6, SYN 13, SYN 27, SYN 33, and SYN 51 have shown higher root length as compared to mega varieties. The mega varieties like HD 2967 (745.99 cm), HD 3086 (900.63 cm), HD 2733 (897.02 cm), and PBW 343 (1092.41 cm) had root length lower than most of the SHW lines (Fig. 3).

The polygon view of the GGE biplot (Fig. 4) indicates the best genotype for the traits studied. The polygon is formed by connecting the markers of the genotypes that are farthest away from the biplot origin such that all other genotypes are contained in the polygon. For most of the root traits like RL(sc), RL (rs), SAR, NTIPS, FORKS genotype SYN 2 showed maximum trait value. Similarly, SYN 28 is better for DRW, and for most of the grain-related parameters, SYN 36 is the winning genotype. The released varieties like HD 2009 and HD 2329 are the poorest for the traits studied.

Assessment of sub-set genotypes

On the basis of phenotypic acceptability and high value for the traits under study, a subset of 34 SHW lines was constituted and was evaluated along with 6 checks for the traits studied earlier following the same experimental procedure (Table 2). For coleoptile length, eleven SHW lines

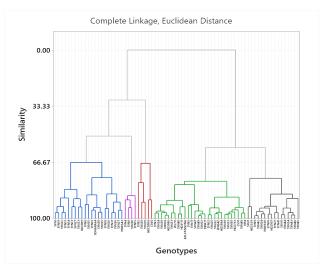


Fig. 4. Dendrogram showing grouping of genotypes based on euclidean distance

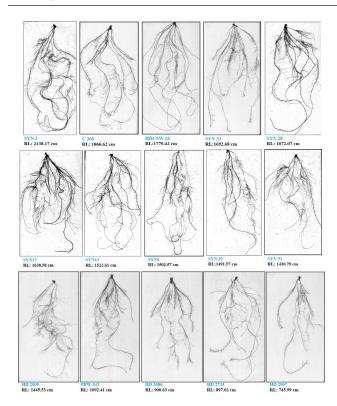
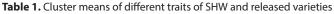


Fig. 3. Root length of a few SHW lines and released varieties measured by root scanner



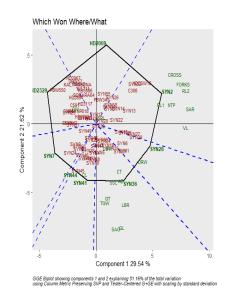


Fig. 4. Which won where biplot showing winning genotypes for the traits studied

have longer coleoptile than C 306 (4.40 cm). Out of eleven, six lines viz., SYN 8, SYN 27, SYN 35, SYN 45, and SYN 46 are of semi-dwarf to medium-tall plant height (range 92-107; data not shown). The coleoptile thickness (CT) is an important trait as it helps in proper and strengthful emergence under heavy residue load in CA. Ten SHW lines have significantly

Traits			Cluster me	ean	
	I	II	111	IV	V
No of the genotypes in eachcluster	21	5	28	4	16
CL	4.2	3.51	4.17	4.78	4.33
СТ	1.7	1.62	1.52	2.05	1.67
SRL	16.58	18.28	17.48	18.47	17.78
SSL	15.49	16.35	15.27	18.7	15.85
RL(sc)	43.5	52.33	36.03	45.34	31.2
RL(rs)	1300.65	1790.93	863.72	1001.34	576.03
SAR	200.91	276.89	123.78	141.39	90.61
VOL	2.46	3.42	1.68	1.56	1.06
AD	0.5	0.5	0.47	0.43	0.51
NTIPS	4503.52	6498.3	3073.75	6284.75	1970.72
FORKS	6787.93	9686.4	4382.71	4057.63	2782.84
CROSS	1683.98	2276.8	1109.38	992	606.63
GL	7.66	7.68	7.55	7.99	7.65
GW	3.29	3.39	3.44	3.25	3.45
GT	2.91	2.99	2.93	3.14	2.98
SAG	84.18	87.27	84.84	92.97	87.94
LBR	2.35	2.29	2.21	2.46	2.23
DSW	0.7	0.78	0.73	0.44	0.75
DRW	0.19	0.22	0.15	0.18	0.17
TGW	44.65	47.71	46.07	50.97	47.16

SYN1 SYN2			JUL	JUL		RL (rs)	SAR	VOL	AU	NTIPS	GL	R و	GT	DAC	LDN	NNC/1		MDI
SYN2	4.35	1.83	14.38	17.02	45.55	1150.64	242.47	3.67	0.62*	5147	8.09***	3.49	3.47***	105.91***	2.32***	0.84	0.14	52.37***
	2.80	1.30	21.23	22.73	74.10***	2178.07***	315.53**	3.72	0.46	7834***	7.19	3.66	3.05	82.79	1.97	1.17**	0.33***	51.14***
SYN4	4.10	2.20***	14.90	20.15	34.17	976.73	120.92	1.05	0.41	5807*	7.76***	3.40	3.37***	95.29***	2.28***	0.61	0.14	54.71***
SYN5	4.35	1.26	16.01	16.48	44.82	1083.80	163.97	2.14	0.48	2310	8.40***	3.50	3.38***	111.21***	2.40***	0.58	0.07	52.43***
SYN6	4.15	1.97*	20.01	20.30	42.62	1535.90	223.76	2.57	0.44	5408	8.10***	3.67	3.25*	105.23***	2.21**	1.01	0.25***	43.58
SYN8	4.90***	2.06***	16.00	21.79	38.14	805.20	118.76	1.21	0.45	6627***	8.26***	3.04	3.50***	101.16***	2.72***	0.49	0.07	48.09*
SYN10	5.25***	1.88	16.24	18.23	45.15	710.75	154.58	2.29	0.65*	2188	8.59***	3.30	3.06	104.80***	2.60***	0.75	0.23***	48.92***
SYN13	3.55	1.83	27.55	12.77	64.41***	1584.68	228.24	2.62	0.51	6148***	7.95***	3.57	2.93	94.07***	2.23***	0.47	0.11	50.88***
SYN15	5.65***	2.25***	21.31	19.36	66.60***	1390.71	215.05	2.72	0.55	6605***	8.24***	3.40	2.77	93.71***	2.42***	0.11	0.42***	50.37***
SYN17	3.95	1.83	23.35	13.02	43.68	992.42	148.27	1.58	0.44	3134	8.30***	2.95	2.82	87.30***	2.81***	0.52	0.09	38.38
SYN19	4.55	1.78	13.57	21.28	46.13	1357.74	241.25	3.17	0.56	5350	7.59*	2.92	3.04	77.66	2.60***	1.14*	0.29***	38.29
SYN21	4.95***	1.77	12.47	15.56	31.73	660.29	111.70	1.52	0.51	1928	7.29	3.55	3.20	85.46**	2.05	0.67	0.13	47.97*
SYN27	5.20***	1.70	17.07	17.02	57.47*	1678.90	215.58	3.49	0.50	5610	8.06***	3.68	2.92	98.02***	2.19**	0.67	0.25***	52.99***
SYN28	3.30	2.00**	12.90	13.42	55.37	1693.68	268.74	3.72	0.49	6577***	8.87***	3.00	3.45***	112.72***	2.96***	0.71	0.30***	50.64***
SYN29	4.10	2.11***	19.50	17.19	36.53	1466.80	239.76	3.00	0.50	7229***	8.06***	3.15	2.92	88.34***	2.56***	0.94	0.23***	49.52***
SYN30	4.40	1.97*	22.23	17.16	48.72	960.33	130.04	1.49	0.38	6791***	8.16***	3.34	2.75	90.72***	2.44***	0.63	0.10	49.76***
SYN31	3.40	1.93	21.67	18.81	41.90	1112.35	215.35	3.35	0.56	2891	7.29	3.25	2.57	70.27	2.25***	0.35	0.12	44.78
SYN32	3.70	1.62	23.03	15.35	47.07	971.26	156.07	1.63	0.46	2800	7.87***	3.15	2.76	82.02	2.50***	0.78	0.36***	43.88
SYN33	3.30	1.81	14.58	13.01	56.24	1727.30	262.15	2.70	0.43	4909	6.93	3.07	2.72	63.99	2.26***	0.99	0.41***	38.02
SYN35	4.80**	1.64	20.16	16.03	45.37	1234.74	191.20	2.10	0.53	4152	8.71***	3.24	3.20	108.95***	2.69***	0.66	0.17	49.21***
SYN36	5.15***	2.13***	23.73	20.79	42.59	1298.97	217.93	2.96	0.56	2927	9.57***	2.88	3.36***	123.92***	3.32***	0.55	0.15	50.72***
SYN37	3.80	1.56	21.14	15.57	47.25	1277.13	224.72	2.99	0.55	3524	8.40***	3.29	3.07	100.55***	2.55***	0.60	0.33***	53.79***
SYN38	3.85	1.46	19.87	15.64	27.95	885.42	116.67	1.07	0.43	2895	8.68***	2.80	2.97	94.70***	3.10***	1.14*	0.20	45.52
SYN39	4.50	1.72	13.91	17.53	24.61	1032.45	163.06	1.78	0.48	3456	7.68**	3.62	3.52***	100.44***	2.12	1.11	0.21**	45.91
SYN40	4.35	2.28***	17.59	21.69	40.31	872.81	131.34	1.32	0.47	3752	7.48	4.31***	* 2.85	96.70***	1.74	1.05	0.17	48.72**
SYN41	5.20***	1.53	20.23	20.26	28.01	627.03	116.84	1.50	09.0	1637	8.67***	3.18	3.32***	109.32***	2.73***	1.14*	0.25***	52.07***
SYN42	4.70*	1.46	29.08	22.90*	32.25	555.42	76.08	0.68	0.39	1734	7.05	3.39	2.81	72.15	2.08	1.01	0.24***	43.36
SYN43	4.35	1.69	25.11	13.02	55.47	765.88	108.78	1.03	0.52	2792	8.28***	3.62	2.89	101.20***	2.29***	0.93	0.21**	52.23***
SYN44	3.75	1.97*	23.49	14.07	15.89	674.73	95.13	0.89	0.53	1923	8.68***	3.67	3.25*	118.54***	2.36***	0.96	0.28***	52.12***
SYN45	5.38***	1.79	12.81	15.84	36.62	404.89	80.27	1.18	0.69**	1203	8.34***	3.46	3.00	101.27***	2.41***	1.09	0.21	46.17
SYN46	5.15***	1.67	11.86	17.54	33.61	824.35	109.71	1.35	0.46	2442	8.17***	3.50	3.09	100.33***	2.34***	1.73***	0.30***	50.37***
SYN48	4.35	1.64	13.63	19.02	44.26	1051.02	136.84	1.59	0.45	3260	8.00***	3.28	2.53	82.56	2.44***	1.28***	0.25***	47.00
SYN52	4.55	1.33	11.59	16.54	23.74	767.50	138.66	1.77	0.67**	2866	8.46***	3.51	3.16	108.21***	2.41***	1.05	0.19	54.03***

SYN54	4.55	1.54	14.98	12.40	28.16	655.36	103.47	1.16	0.59	2188	7.09	4.02	2.79	82.48	1.76	0.74	0.19	45.31
HD2967	3.30	1.80	16.30	13.08	46.66	786.44	109.15	1.31	0.43	2396	6.08	3.45	2.94	59.06	1.76	0.62	0.13	36.67
HD3086	3.30	1.55	17.52	13.26	32.47	898.85	129.24	1.57	0.47	3583	6.63	3.47	2.83	66.66	1.91	1.00	0.15	40.29
HDCSW18	3.30	1.57	21.80	13.42	52.96	1776.72	292.33	3.72	0.53	5409	7.42	3.57	2.69	79.68	2.08	0.86	0.15	41.49
HD3117	3.26	1.53	28.13	15.72	46.23	992.01	129.47	1.50	0.46	3679	6.93	3.87	2.90	79.01	1.79	0.55	0.11	46.62
C306	4.40	1.31	22.81	20.79	42.72	1863.45	271.05	2.98	0.47	5495	6.92	3.36	3.13	74.37	2.06	0.49	0.06	43.78
PBW343	3.30	1.68	16.02	14.39	43.96	1124.57	165.98	1.85	0.47	3819	7.07	3.55	2.94	77.05	1.99	1.01	0.19	46.26
CD 0.05	0.26	0.14	2.19	2.10	4.27	63.40	14.02	0.47	0.09	282.50	0.14	0.11	0.10	3.91	0.08	0.076	0.017	1.192
CD 0.01	0.34	0.18	2.93	2.81	5.73	85.06	18.81	0.63	0.12	379.00	0.18	0.15	0.13	5.25	0.11	0.102	0.023	1.600

high CT (1.97 to 2.28 mm) over HD 2967 (1.80 mm). For RL (rs) many SHW lines have a better root system and SAR when compared to mega variety HD 2967 (786 cm) and HD 3086 (899cm). However, over check C 306, only one SHW line SYN 2 (2178 cm) exhibited significantly higher root length and SAR (31553). Root tips are an important component of root system architecture, more root tips help in scavenging nutrients in a better way than others. Eight SHW lines exhibited significantly high NTIPS over the check C 306 with SYN 2 having a maximum number (7834) for this trait. SHW lines have a bigger grain length in general as compared to normal bread wheat varieties. In this study, except few, the rest of the SHW lines had significantly higher grain length than the check varieties (Table 2). Because of the bigger grain length and grain thickness, SHW lines exhibited high grain surface area than check varieties. Most of the SHW lines had significantly high SAG than HDCSW 18 and other checks therefore, they also had higher TGW (range: 47.97 to 54.71 gm). In the case of DRW, many of the SHW lines have significantly higher trait values as compared to check varieties (Table 2). It shows they have a longer root length than most mega varieties.

Discussion

It is crucial for developing countries to sustain wheat production at a greater level in order to meet the expanding population's needs for food security. It is possible to counteract the effects of climate change and decreasing natural resources like irrigation water and soil health on wheat productivity by adopting resource-conserving strategies like CA (Yadav et al. 2017). Also, proper assessment and introgression of novel variation (from SHW lines) for the traits specific to CA, like root system architectural traits in wheat breeding programmes (Gaikwd et al. 2022), will be crucial. The International Maize and Wheat Improvement Center (CIMMYT) has been instrumental in developing SHW lines since the late 1980s (Das et al. 2016). The agronomically undesirable characters, such as tenacious glumes that result in non-free threshing grains, prevent synthetic wheat from being used as a cultivar. Therefore, it is necessary to either eliminate these characters or transfer desired traits of synthetic wheat into common wheat varieties by creating synthetic derivative lines through crossing with superior common wheat cultivars (Mujeeb-Kazi et al. 2008). SHW has been recognized and verified in subsequent works and numerous synthetic derivative lines have been registered as cultivars (Li et al. 2018).

In this study, significant genetic variability and a wide range of traits in the SHW lines and released varieties revealed the absence of any purposeful selection for them, supporting previous findings (<u>Ranjan</u> et al. 2021). In the present study, GCV and PCV ranged from 7.30 to 62.32% and 7.97 to 63.45%, respectively, in the complete set of genotypes, and 8.54 to 45.87 % and 8.69 to 46.71 % in the subset of genotypes. The range was as per the earlier reports in wheat (Kumar et al. 2017; Ranjan et al. 2021). Higher values of PCV and GCV in the case of important roo traits like VOL, NTIPS, FORKS, SAR, and DRW indicated an opportunity for improvement in these traits through selection, but that would be difficult due to their below-ground presence.

In the past, direct visual selection for increased performance, which is nearly always focused on the assessment of the above-ground sections of the plant has been largely responsible for the steady incremental advances produced by current crop breeding. Due to the practical challenges in evaluating the "hidden half" of agricultural plants, direct selection of the root system is missed in this (Bishopp and 2015). Field-based root phenotyping of a large number of genotypes is frequently seen as being more difficult, labor-intensive, and timeconsuming than methods based on seedlings. It is common to see recommendations that specific root features be selected in breeding programmes in published studies on crop root systems, but rarely is there any mention of the mechanisms by which this would be achieved (Ober et al. 2021). Contrarily, a common misconception holds that breeding cannot optimally improve agricultural species' root systems since they are too expensive and/or complex. But the genetic basis for differences in root traits can be measured with high accuracy using low-cost methods like hydroponic screening, which are not affected by the environment (Petrarulo et al. 2015).

The root systems of SHW and their potential to boost productivity are untouched areas of research, which may be due to difficulty in the phenotyping of root architectural traits. In the present study, most of the released mega varieties showed a reduction in the studied traits in terms of root morphology (Fig. 4). In contrast, traits like RL, RSA, VOL, and NTIPS in the SHW lines, SYN2, SYN33, SYN28, SYN27 and SYN13 had higher values, indicating their appropriateness as donors for the enhancement of root morphology. This outcome is consistent with a recent study by Bektas et al. (2017), which found long, thin roots in the D genome progenitors of common wheat. Due to their direct impact on the acquisition of water and nutrients in the soil layers, the production of root branches (or forks) is a crucial determinant of the architecture of the root system as a whole (and Wojciechowski 2015). The total root length, surface area, and biomass are all increased by this single trait alone. Wide variations in these traits were found in our investigation. We have also found that root length has a highly significant correlation with the associated traitlike SAR (0.94***), VL (0.82***), NTIPS (0.79***), and FORKS (0.94***). The results are consistent with the findings of Sandu et al. (2021), who have reported a significant and positive correlation of total root length to RSA, RD, VOL,

Ntips, and FORKS in nested synthetic wheat introgression lines. Also in our study, many SHW lines had fork counts that were noticeably larger than those of released varieties.

The ability to extract more water from the soil was demonstrated to be aided by the maximum root depth, which also showed a higher heritability than other root traits (Lopes and Reynolds 2010). In the present study, a few SHW lines had longer root systems, and this trait also had a high GCA value. Among the released varieties used in this study, the well-known drought-tolerant variety C 306 and the CA-specific variety HDCSW 18 had longer root systems than many SHW lines. However, mega varieties like PBW 343, HD 2733, HD 2967, and HD 3086 showed shallow root systems. Although deeper root systems are linked to cooler canopy temperatures and yield increases in drought stress (Lopes and Reynolds 2011), shallow root systems may also be advantageous for catching rainfall that does not infiltrate deeper soil layers (Ehdaie et al. 2012). The mega varieties utilized in this study were mostly recommended for irrigated ecosystems, and hence their shallow root systems justify their selection history.

In a hydroponic study conducted in Australia under controlled glasshouse conditions, SHW lines showed larger initial aerial and root biomass than existing Australian check cultivars. The root systems of these SHW were noticeably thicker and, in some cases, even longer than those of outstanding common wheat cultivars (Dreccer et al. 2007). In our study, the average diameter of roots of a few SHW lines, viz., SYN1, SYN10, SYN45, and SYN68, was significantly higher than HDCSW 18, which has the highest average diameter in the case of released varieties (Table 2). Palta et al. (2011) visualised the root systems of three synthetic backcross lines (SBLs), and reported that a strong and large root system aids in adaptability to dry conditions and dry seasons.

Wheat's grain size, shape, and weight are highly important traits, and they are classic examples of traits with variations that emerged as a result of polyploidization and domestication (Yan et al. 2017). Wheat domestication transformed a long, narrow wild grain into a wider, shorter modern grain (Gegas et al. 2010). In the present study, SHW lines exhibited highly significant values for the traits associated with grain parameters like GL, GT, SAG, and LBR over released varieties (Table 2). In an extensive study on the synthetic derived lines (SDLs), Rattey et al. (2011) found that SDLs are most beneficial for increasing and maintaining the grain size across environments in northern Australia, even if the grain production is low. TGW showed a significantly positive correlation with GL, GW, GT, and SAR in this investigation. This confirms prior findings of earlier studies carried out by Morgounov et al. (2018) and Wang et al. (2021), who identified new SHW lines with larger TGWs exceeding 60 g. This implies that an increase in grain weight would follow an increase in the geometric parameters of the grains. The SHW lines (Table 2) should be crossed with high-yielding cultivars to transfer high-grain weight traits. This will generate new germplasm resources with large grain sizes and there is evidence that seed size may influence adaptation to zero-tillage as longer coleoptiles are associated with larger seeds (Botwright et al. 2001).

Before dwarfing wheat was introduced, deep planting was a common practice among Indian farmers to take advantage of the moisture present in the deeper layer of the soil profile. However, shallow seeding became more popular after the emergence of semi-dwarf varieties since they had shorter coleoptiles. Under CA conditions the high residue load, sometimes unevenness of the land, and the compactness of the top few centimeters of the soil, the seed drill must use more force to plant the seed in the deeper layer of the soil so as to reach the moisture (Yadav et al. 2018). This necessitates the use of a genotype with a longer coleoptile that can emerge even from this slightly deeper level of depth. Coleoptile thickness, in addition to length, is a crucial parameter in CA since it aids in the crop's proper emergence from a heavy residue load. In the present study, six SHW lines, viz., SYN 8, SYN 27, SYN 35, SYN 45, and SYN 46, exhibited longer coleoptile length with semi-dwarf to medium-tall plant height (range 92–107; data not shown) and 10 SHW showed significantly thicker CT over the checks. These findings clearly indicate that useful genetic variation for the traits important for CA does exist in synthetic wheat. These SHW lines, therefore, should be used in breeding programmes to develop SBLs or SDLs.

Supplementarial material

Supplementary Tables S1 to S3 presenting analysed data that can be accessed online www.isgpb.org

Authors' Contribution

Conceptualization of research (KBG, RY); Designing of the experiments (KBG, RY); Contribution of experimental materials (RY, KG); Execution of field/lab experiments and data collection (KBG, RP, AGS RA); Analysis of data and interpretation (KBG, NK, MK, PB); Preparation of the manuscript (KBG).

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Supplementary Table S1. A list of SHW lines and mega wheat varietiesused in the experiment

Genotypes	Parentage	Genotypes	Parentage	Genotypes	Parentage
SHW lines					
SYN1	DOY1/Ae. squarrosa (188)	SYN26	YAR/Ae. squarrosa (783)	SYN51	Not available
SYN2	ALTAR 84/Ae. squarrosa (193)	SYN27	68.111/RGB-U//WARD/3/FGO/4/ RABI/5/Ae. squarrosa (878)	SYN52	ALTAR 84/Ae. squarrosa (502)
SYN3	ALTAR 84/Ae. squarrosa (198)	SYN28	68.111/RGB-U//WARD/3/FGO/4/ RABI/5/Ae. squarrosa (878)	SYN53	ALTAR 84/Ae. squarrosa
SYN4	Not available	SYN29	68.111/RGB-U//WARD/3/FGO/4/ RABI/5/Ae. squarrosa (882)	SYN54	Not available
SYN5	Not available	SYN30	CROC 1/Ae. squarrosa (518)	SYN55	Not available
SYN6	ALTAR 84/Ae. squarrosa (211)	SYN31	PBW 114/Ae. squarrosa	Released varietie	es
SYN7	ROK/KML//Ae. squarrosa (214)	SYN32	ALTAR 84/Ae. squarrosa (JBANGOR)	NP4	SEL.HETEROZYGOUS LINE OF LOCAL MUNDIA
SYN8	YUK/Ae. squarrosa (217)	SYN33	YAY2/TEZ//Ae. squarrosa (249)	NP818	DO/E518//SPP/NP114/3/ WIS245'S'
SYN9	Not available	SYN34	GAN/Ae. squarrosa (180)	KALYANSONA	PJ SIB/GB55
SYN10	Not available	SYN35	D67.2/P66.270//Ae. squarrosa (257)	SONARA64	YT54/N10BII2*Y54 ORIGIN
SYN11	D67.2/P66.270//Ae. squarrosa (220)	SYN36	SRN/Ae. squarrosa (358)	HD2009	LR 64A/NAI 60
SYN12	DVERD 2/Ae. squarrosa (221)	SYN37	SCOOP1/Ae. squarrosa (358)	HD2329	HD 1962/E 4870/3/ K65/5/HD1553/4/UP262
SYN13	SORA/Ae. squarrosa (884)	SYN38	SCA/Ae. squarrosa (518)	HD2733	ATTILA/3/TUI/CARC// CHEN/CHTO/4/ATTILA
SYN14	68.111/RGB-U//WARD/3/Ae. squarrosa (326)	SYN39	YAR/Ae. squarrosa (518)	HD2894	HW 2002/WR 196
SYN15	ACO 69/Ae. squarrosa (309)	SYN40	BOTNO/Ae. squarrosa (620)	HD2967	ALD/CUC//URES/ HD2160M/HD2278
SYN16	GARZA/BOY//Ae. squarrosa (311)	SYN41	D67.2/P66.270//Ae. squarrosa (633)	HD3086	DBW14/HD2733// HUW468
SYN17	Not available	SYN42	Not available	HDCSW18	PBW 343/CL 1538
SYN18	YAV3/SCO//JO69/CRA/3/ YAV79/4/Ae. squarrosa (498)	SYN43	YAV2 , TEZ//Ae. squarrosa (895)	HD3117	HD2733/HD 2824// DW1278
SYN19	68.111/RGB-U//WARD/3/Ae. squarrosa (511)	SYN44	ARLIN/Ae. squarrosa (283)	C306	RGN/CSK3//2*C591/3/ C217/NI4//C281
SYN20	DOY 1/Ae. squarrosa (515)	SYN45	Not available	C591	Type9/8B
SYN21	68.111/RGB-U//WARD/3/ FGO/4/RABI/5/ <i>Ae. squarrosa</i> (629)	SYN46	CROC 1/Ae. squarrosa (879)	PBW343	ND/VG9144//KAL/ BB/3/Y ACO'S'/4/VEE#5 'S
SYN22	YUK/Ae. squarrosa (864)	SYN47	Not available	PBW550	WH 594/RAJ 3856//W 485
SYN23	CROC 1/Ae. squarrosa (725)	SYN48	DOY1/Ae. squarrosa (372)	DBW17	CMH79A.95/3*CNO 79// RAJ3777
SYN24	68.111/RGB-U//WARD/RESEL/3/ STIL/4/Ae. squarrosa (781)	SYN49	CPI/GEDIZ/3/GOO//J065/CRA/4/ Ae. squarrosa (409)	WL711	SKA/CHR//KAL
SYN25	68.111/RGB-U//WARD/RESEL/3/ STIL/4/Ae. squarrosa (783)	SYN50	ALTAR 84/Ae. squarrosa	WH542	JUP/BJY"S"//URES

Supplementary Table 2. Variability parameter of characters under the study of 55 SHW lines and 19 mega varieties

Character	MSS	CV	Range	Mean ± SE	GCV	PCV
CL	1.22*	5.06	2.54-6.5	4.20±0.091	18.26	18.94
СТ	0.1*	4.84	1.188-2.388	1.64±0.026	13.40	14.24
SRL	28.42*	14.71	10.18-27.14	17.40±0.438	19.01	24.03
SSL	12.37*	5.93	11.38-21.95	15.72±0.287	15.26	16.37
RL(sc)	221.88*	4.72	18.2-70.2	38.71±1.224	27.00	27.41
RL(rs)	281985*	1.21	304.32-2129.17	995.60±43.650	37.71	37.72
SAR	7219.24*	4.5	42.06-307.04	149.80±6.984	39.98	40.23
VOL	2.8*	10.86	0.4-3.68	1.88±0.138	62.52	63.45
AD	0.0102*	9.14	0.358-0.676	0.49±0.008	13.22	15.96
NTIPS	4539347*	1.53	1180-7623	3645.96±175.145	41.31	41.33
FORKS	8720882*	2.45	1075-11175	5060.14±242.745	41.23	41.30
CROSS	598148*	5.56	234-2722	1236.27±63.573	44.06	44.41
GL	1.108*	1.69	6.017-9.58	7.63±0.087	9.68	9.82
GW	0.138*	2.58	2.88-4.23	3.39±0.031	7.54	7.97
GT	0.1047*	3.71	2.387-3.497	2.95±0.027	7.30	8.18
SAG	497.03*	4.21	51.219-122.10	85.93±1.833	18.10	18.59
LBR	0.2091*	3.44	1.746-3.236	2.27±0.038	14.02	14.43
DSW	0.120*	3.61	0.107-1.427	0.71±0.029	34.19	34.38
DRW	0.0150*	7.88	0.006-0.394	0.17±0.010	50.22	50.83
TGW	58.23*	1.77	32.10-57.01	46.28±0.627	11.63	11.69

Supplementary Table 3. Variability parameter of characters under the study of selected 34 SHW lines and 6 mega varieties

Character	Mean Square	CV	Range	$Mean \pm SE$	GCV	PCV
CL	1.03*	3.01	2.80-5.65	4.23 ± 0.114	16.83	17.09
СТ	0.13*	3.86	1.26-2.28	1.75 ± 0.041	14.34	14.85
SRL	44.2*	5.78	11.59-29.08	18.74 ± 0.743	24.75	25.41
SSL	19.29*	6.11	12.40-22.90	17.00 ± 0.491	17.75	18.77
RL1	289.58*	4.94	15.89-74.10	42.78 ± 1.903	27.91	28.34
RL2	333996*	2.83	404.89-2178.07	1110.18 ± 64.614	36.76	36.86
SAR	8173.91*	4.03	76.08-315.53	172.00 ± 10.108	37.06	37.28
Volume	1.70*	11.01	0.68-3.72	2.10 ± 0.146	43.26	44.63
AD	0.1087*	8.8	0.38-0.69	0.50 ± 0.012	45.87	46.71
Ntips	6433526*	3.49	1202.84-7834.21	4010.63 ± 283.575	44.65	44.79
GL	1.03*	0.85	6.08-9.57	7.88 ± 0.114	9.09	9.13
GW	0.189*	1.59	2.80-4.31	3.40 ± 0.049	8.97	9.11
GT	0.136*	1.61	2.53-3.52	3.03 ± 0.041	8.54	8.69
SAG	478.81*	2.1	59.06-123.92	92.19 ± 2.447	16.72	16.85
LBR	0.258*	1.73	1.74-3.32	2.34 ± 0.057	15.28	15.38
DSW	0.182*	4.55	0.11-1.73	0.82 ± 0.048	36.43	36.71
DRW	0.0170*	4.05	0.06-0.42	0.21 ± 0.015	44.82	45.00
TGW	46.15*	1.24	36.67-54.71	47.46 ± 0.760	10.08	10.16