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Variety Development of Wheat in Turkey and the Impact of Regulations

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ABSTRACT

The main aims of this study were to compile the historical wheat variety list released in wheat breeding in Turkey, to compare the wheat variety release with some selected countries and evaluate the impact of regulations on wheat research and variety development. Wheat breeding started in 1925 soon after the foundation of Republic of Turkey. First variety was released in 1928, and 617 wheat varieties were released since then. National Variety List (NVL) was published every year, however only 484 of total released varieties are currently in the NVL. 485 varieties are of bread wheat and 128 of them are durum wheat, 2 two are of *Tr. monococcum*, 1 is of *Tr. turgidum* and 1 is of *Tr. spelta*. As Turkey has diverse growing conditions, spring, winter and facultative bread and durum wheat varieties are grown, and varieties are released for these conditions. 316, 191 and 110 varieties of winter, spring and facultative wheat have been released, respectively. The comparisons were made for variety release among Turkey, Iran, Hungary and the UK. The highest number of varieties released per ha was in UK followed by Hungary, Turkey, Iran in 5-year-period. New regulations stimulated releasing greater number of varieties and involvement of private sector in breeding. Surveys are made to find out the varieties planted in the farmers' field and their adoption rates. New molecular technologies, such as fingerprinting can be used to increase accuracy of the variety identification and adoption rate of any variety in such surveys.

Keywords: wheat, breeding, variety, National Variety List

Introduction

Considering the importance of wheat products in people's diet and wheat trade internally and internationally, wheat is a staple food and strategic crop in Turkey. Though, wheat has been grown in Turkey for about 10 000 years, modern wheat development activities started much later. During the Ottoman era, wheat was grown in high rainfall areas, mainly outside Modern Turkey's territory (Braun et al. 2001). Wheat breeding started in the country with the establishment of a research station in Eskisehir in 1925, soon after the foundation of the Republic of Turkey (Ozbek et al. 2016). The name of the station was "Islahı Buzur" meaning seed development. This was followed by the establishment of research stations in Adapazarı, Yeşilköy (Istanbul) and Ankara before 1930. By 1989, there were 14 public research institutes working on wheat, though some of them were closed later (such

as the Yeşilköy Research Institute). Wheat variety development studies aimed to develop Winter Wheat (WW) and Spring Wheat (SW), and Bread Wheat (BW) and Durum Wheat (DW) varieties, since Turkey has different regions with different climatical conditions. The most important outcome of the investment in agricultural research is the development of varieties for cultivation by farmers. It is noted that release of wheat varieties started soon after the establishment of first research stations (Keser et al. 2017). The first wheat variety released in Turkey was Karakılıçık1133 by Yeşilköy Agricultural Research Station, Istanbul (now closed) in 1928 (Altay, 2018).

The main aims of this study were to compile the wheat varieties developed and registered in the history of wheat breeding in Turkey and to give insight on the impact of regulations on wheat R&D development and variety registration. Though, the National Variety

List (NVL) is compiled, published and renewed yearly by Variety Registration and Seed Certification Center (VRSCC), not all released varieties in wheat breeding history are listed in NVL. In accordance with regulations, wheat varieties are registered for a period of 10 years and variety owners can apply for renewal after every 10 years. If the variety owner does not apply for renewal of it, the variety is automatically dropped from the NVL. This provides the basis and reason for this study. Previous studies have attempted to compile all the varieties released in Turkey; however, the numbers were increasing year by year and some of the old varieties and the ones dropped from the NVL were missing in those lists (Braun et al. 2001; Ozberk et al. 2016; Keser and Cakmak, 2021).

In this study, in order to look at the impact of regulations on wheat variety development, the variety development activities are divided into 3 periods: I. 1925-1963, II. 1964-2006 and III. 2007-2021. The main characteristics of the periods are as follows;

First period: 1925-1963

The first thing done in wheat breeding is the collection of Land Races (LR), mainly from the region where the Research Station is located. Pure lines were then developed by making the selections from the collected populations and good performers were registered, their seeds were multiplied and provided to the neighboring farmers first. Some LRs were also collected from different parts of Turkey and the same procedure was applied to them as well. The seeds of identified genotypes were produced in the fields of the Research Station and distributed to neighboring regions around the station, until State Farms (SF) were established in mid-1940s. There was no close coordination or concerted effort among the research stations; they were mainly serving the region where they were located. Crossing started in the late 1920s, and few varieties were developed from crosses in this period. The crosses were mainly made between LRs or between LRs and introduced germplasm, e.g. Mentana. Seed Law no 308, which set the rules for releasing a variety and seed production, was published in 1963 and is considered as the end of the first period (OG, 1963).

Second period: 1964-2006

After 1963, already established State Farms (SF) reframed their working according to Seed Law no 308 and these farms were becoming more formal and coordinating for seed production and distribution. While most new varieties were coming from crosses during this period, there were few LR-selected-varieties and introduced varieties. Seed was being produced by SFs, according to the rules of Seed Law no 308,

and distributed to the farmers during this period. There were also introduced/imported SW and WW varieties from Mexico and USA, respectively in mid- to late-1960's as part of a project. The Green Revolution was in progress in the 1960s in India and Pakistan. SW varieties developed by CIMMYT in Mexico were introduced and planted, and wheat production increased dramatically in those countries. As Turkey already had areas growing SW, Turkey also wanted to introduce new SW varieties to increase the wheat production. Turkey made an agreement with Oregon State University in the USA called the "Wheat Enhancement Program". This agreement aimed to increase wheat production in Turkey not only in SW areas, but also in areas growing WW. The project was implemented in two phases, starting in 1967 (Kronstad, 2000). The first phase was the introduction of SW varieties, developed in CIMMYT, Mexico and introduced into the coastal areas of Turkey. The second phase focused on enhancing wheat production on the Anatolian Plateau in collaboration with Oregon State University. Seventeen varieties (10 from USA and 7 from Mexico) were directly introduced from USA and they were registered in Turkey.

Third period: 2007-2021 (inclusive)

A new Seed Law no 5553 was published in the Official Gazette in 2006, replacing the 1963 Seed Law (OG, 2006). This is considered the end of the second period. Since 2007, all varieties were coming from crosses except a few primitive wheat varieties (*Tr. monococcum* and *Tr. turgidum* and *Tr. spelta*) selected from landraces. Before the new Seed Law was published, the Plant Breeders Right (PBR) Law no 5042 on new plant varieties was also published in 2004 (OG, 2004). These new regulations stimulated the variety development and registration not only in the public sector, but also in private sector.

The number of varieties registered has increased over time and these have been recorded in the NVL. This list is compiled and published by the VRSCC, in accordance with the rules in force. In 1963, seed was being produced for 31 wheat varieties (22 BW, 9 DW) (OG, 1969). By 2015, 135 wheat varieties were recorded in farmers' fields, according to a survey conducted across 27 provinces in Turkey (Bishaw et al. 2021). The number would have most probably been increased if the survey had covered all wheat growing areas of Turkey.

Materials and Methods

The sources used to compile the variety release/registration in Turkey from the beginning of wheat breeding to 2021 (inclusive) were the NVL and

the Registration Reports of the VRSCC, websites of variety owners, reports of the Research Institutes (especially for the varieties dropped from the NVL, which were also confirmed by breeders, when needed). Growth Habit (GH) and origin was assigned to each variety based on the registration report of the VRSCC, the website of the variety owner or the GH given in some publications and theses. In addition to Turkey, the number of varieties released in 5 years period were compiled from Hungary, the UK and Iran, and the comparisons have been made based on the number of released varieties in this period and area per variety in each country. In order to evaluate the impact of the regulations on variety release in Turkey, it has been studied in 3 periods (1925-1963; 1964-2006 and 2007-2021). The impact was evaluated based on the total number of varieties released and the number of variety release per year in each period.

Results and Discussions

The most important outcome of the investment in agricultural research is the development of varieties for cultivation by farmers. As the stages and timeline given in Table 1, it takes about 15 years from crossing to commercialize a variety in a conventional breeding scheme in Turkey (Keser et al. 2021). However, if any organization (public or private) introduces a genotype from outside the timeline becomes shorter; it may be reduced to 3-4 years depending on what stage the genotype is introduced at.

Since the first wheat variety was released in 1928, 617 wheat varieties have been released in Turkey (Annex 1). Only 484 of these are currently in the NVL (VRSCC, 2021); 133 wheat varieties are not currently listed. As both BW and DW are grown in Turkey, BW and DW varieties along with some primitive wheats have been registered; 485 varieties are BW and 128 of them are DW varieties, 2 are *Tr. monococcum*, 1 is *Tr. turgidum* and 1 is *Tr. spelta* (Table 2). As Turkey has diverse growing conditions, spring, winter and facultative bread and durum wheat varieties are grown, and varieties are released to be grown in those diverse conditions. 316 WW varieties have been released, and 191 and 110 for SW and FW, respectively.

While 15 public research institutes released 313 varieties, 7 universities released 23 varieties (Table 3), and 67 private companies were involved in releasing 281 varieties (Table 4) in total. Universities are considered as public sector in later analysis, as they were public universities. In public research institutes, the highest number of varieties were released by the Central Research Institute for Field Crops followed by the Transitional Zone Agricultural Research Institute,

with 49 and 46 varieties released respectively (Table 3). Though the list indicates 41 varieties were released by the Maize Research Institute, 17 of them were directly introduced from the USA (10) and Mexico (7) and registered with the Maize Research Institute within the wheat enhancement program.

Wheat breeding in Turkey has a long history with international relationships and intensive international cooperation on wheat development with countries and international organizations that made substantial impact on wheat breeding and production in Turkey (Lantican et al. 2005; Lantican 2016). Turkey has introduced wheat varieties from 23 different countries and two CGIAR Centers, CIMMYT and ICARDA. In total, 221 varieties originating from 23 countries have been released in Turkey; the highest number of varieties were from Italy with 60 varieties followed by France and Bulgaria with 28 and 20 varieties, respectively. 107 varieties originated from the CGIAR Centers, CIMMYT and ICARDA, with germplasm being mostly SW. Turkey, CIMMYT and ICARDA have been carrying out the International Winter Wheat Improvement Program (IWWIP) since 1986 (IWWIP, 2021). The IWWIP provides germplasm, advanced lines and potential variety candidates to breeding programs both in Turkey and around 50 other countries. 110 varieties originating from IWWIP germplasm have been released by both public institutions and private companies in Turkey.

Comparisons of wheat variety releases in selected countries

Wheat varieties are released by public institutions and private sector organizations in many countries. Their contribution to variety development and release is used to compare the public vs private sector and to understand the development stage of the private sector in a given country. In this study, several countries were selected to make comparisons among themselves but mainly with Turkey. When selecting countries, wheat area, similarity/diversity of wheat growing conditions and public/private/university roles and involvement in wheat variety development were considered. Hungary and the United Kingdom (UK) from Europe, and Iran (a neighboring country to Turkey where both, WW and SW are grown under irrigated and rainfed conditions) were selected. Wheat is grown in high rainfall and high yield potential conditions in Hungary and UK on around 1.0 million ha and 1.8 million ha respectively (based on a 5-year average of 2016-2020) (FAO, 2021). BW is the dominating species in both Hungary and UK, with few DW varieties and small acreage in Hungary. There is very little, if any, DW area in UK. Turkey, with 7.3 million ha on average of same period has the largest wheat area among the 4 countries, and

Iran follows Turkey with 6.8 million ha on average (FAO, 2021). DW covers about 25% of the wheat area in Turkey, and around 5-10% in Iran. While public institutions and universities and the private sector work on wheat variety development and release in Iran and Turkey, only public institutions and the private sector release wheat varieties in Hungary, and only the private sector releases wheat in UK. It is also necessary to note that any variety released in an EU country can be registered in Hungary or the UK and directly enter production.

Along with having the largest area of wheat planted, Turkey also released the highest number of varieties over the 2016-2020 period, followed by the UK, Hungary, and Iran (Table 5). There are frequent discussions among breeders, seed producers, millers, industry and decision-makers asking why so many varieties are released in each year and why there are so many varieties in the market, especially in Turkey. Without getting into the discussion deeper about whether the number of varieties are high or low, as this is not the scope of this study, we may evaluate the current situation in selected countries. The number of varieties released per year in each country may be misleading for making comparisons, without taking into consideration the wheat area, diversity of the wheat growing conditions and farm households, wheat species, wheat products, industry, and end-user demand, etc. However, the number of varieties released in 2016-2020 period and wheat area will be used to make some comparisons among the countries to provide initial insights to the readers of this study.

The average number of varieties released per year in the 2016-2020 period in Hungary (Personal com with Dr. Gyula, Budapest, Hungary 2021), Iran (Personal communication with Dr. Roustaii, Maragheh, Iran, 2021), Turkey and the UK (PVSG, 2021) were 13.2, 10.2, 43.8 and 34.2, respectively. The highest average number of varieties released per year was in Turkey followed by the UK, Hungary and Iran in the same period. One of the comparisons that can be made is how many varieties are released per area in the countries in the same period. Looking at the area per variety, the average area per variety released in 2016-2020 period is 76752 ha, 666075 ha, 167766 ha and 52478 ha in Hungary, Iran, Turkey and UK respectively (calculated as average total area divided by average number of varieties released in the period). While the highest number of varieties released per ha was in UK, lowest was in Iran in the same period, Hungary was the second and Turkey is in the third rank when area/variety is considered. The calculation and comparison based on

the area per variety released may not be completely reflecting varieties grown in the farmers' fields since every new variety may not go to production soon after release or in a given period that may stay in the list for years without contributing to the production. However, it may be an indication to show how productive the breeding programs in releasing variety in different countries are.

The impact of regulations on wheat variety release

Wheat releases were studied across the 3 periods in order to evaluate the impact of regulations on variety development in Turkey. During the first period, public institutes were developing varieties, producing the seed and distributing it to farmers within the framework of Ministry of Agriculture directives. There were not closely coordinated and concerted efforts among the institutes; mainly, each institute was acting separately. Nineteen varieties were released from the beginning of wheat research until the first regulation enacted in 1963, while 90 varieties were released in the second period, and 408 in the third (Table 6). While one variety was released in every 2 years in the first period, it was 4.4 varieties/year and 25.5 varieties/year in the second and third periods, respectively. The new 2004 PBR Law and the 2006 Seed Law stimulated variety release not only in the private sector but also in the public sector. Turkey, by changing the regulation, aimed to increase private sector involvement in variety registration, seed production and seed distribution in order to benefit from the yield advantages of newly developed wheat varieties in a short time as the newer varieties had higher yields both in irrigated and rainfed conditions mainly through genetic gain in yield (Gummadov et al. 2015; Keser et al. 2017). It was hoped that the newly developed varieties with high yield, good industrial quality, disease resistance, biotic and abiotic stress tolerance would go faster to farmers' fields and that production would be boosted and would be more stable without being influenced by weather fluctuations, both across different growing environments and years, as most of the wheat has been grown under rainfed conditions. Subsequently, the total number of released varieties increased drastically. In addition, due to the government's certified seed subsidies and the yield advantages of new varieties, it can be concluded that the increasing demand of farmers for certified seeds contributes to the introduction of a growing number of new varieties.

The private sector aggressively increased wheat variety registration in the 3rd period and released a greater number of varieties per year than public sector,

starting from 2011 onwards except in 2015 and 2019, when the numbers were very close (Figure 1). The private sector released 9 times more varieties than the public sector in 2017, releasing 45 varieties compared to the public sector's 5. In other years in this period, the private sector was releasing about two times as many varieties than the public sector, except 2015, 2019 and 2020.

Though the wheat seed production more than doubled from 210044 t in 2007 to 483951t in 2019 and distribution in the same years were 173045 t and 448882 t, respectively, it is not known what percent of the seed distributed was coming from new varieties. In addition, there has been no nation-wide study giving what is planted in farmers' fields in a given year though some studies provide areas covered by old and new varieties (Bishaw et al. 2021).

Conventionally, the calculation of which varieties are planted in farmers' fields and the rates of adoption of wheat varieties is determined through surveys, and the names of the varieties are determined based on the opinion of an expert or the farmer's declaration during the survey. However, some studies indicate that the accuracy of information collected during surveys and the identification of the variety when done by an expert or farmer's declaration may be as low as 30% (Jaleta et al. 2020), especially in countries where informal seed systems and seed exchanges among farmers are in high rates. Phenotypic identification and knowledge of the farmer about a particular variety can also vary due to environmental effects and the limited knowledge of the farmer. The application of molecular marker technologies to identify varieties grown in farmers' fields can overcome these challenges and increase the accuracy of varietal identification (Dreisigacker et al. 2019) and the adoption rate of any given variety. Molecular technologies including finger printing are becoming widely available and cheaper. These technologies may increase accuracy compared to phenotypic identification and farmers' declarations (Yirga et al. 2016). The fingerprinting technology has been used successfully in different countries and in different crops both for identification of the varieties and their adoption rates (in wheat Yirga et al. 2016; Dreisigacker et al. 2019; Hodson et al. 2020; Jaleta et al. 2020, Garapaty et al. 2021, in maize Yirga et al. 2016, in rice Kretzschmar et al. 2018). This technology can also be used to accurately identify varieties planted in farmers' fields and evaluate the adoption rates of the varieties in Turkey. This will help all stakeholders, breeders, seed producers, wheat industry, farmers and more importantly for decision makers to plan better for the future.

Conclusions

617 wheat varieties were developed in Turkey since the beginning of the wheat breeding in 1925. Many primary, secondary and tertiary regulations were developed to regulate the variety registration, seed production and distribution. New regulations stimulated to develop greater number of varieties and private sector involvement in variety registration seed production and distribution. New technologies, such as fingerprinting, can be used to evaluate what is planted in farmers' fields that would help all stakeholders in this business, especially for decision makers.

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Table 1. Conventional wheat variety development, evaluation, and release scheme in Turkey.

Steps	Variety Trial Stages	Timeline (year)	No of Locations
1	Crossing	0	1
2	Selecting segregating populations	1-5	1
3	Observation nursery	6	1
4	Preliminary yield trial	7	1-2
5	Yield trial	8	2-3
6	Advanced yield trial	9	2-3
7	Regional yield trial	10	4-6
8	Registration trials	11-12	6-10
9	Seed multiplication and demonstration	12-13	
10	Commercialization	~15	

Table 2. The number of wheat varieties released in different species and their growth habit.

Species ¹	No of Varieties	Growth Habit ²	No of Varieties
BW	485	WW	316
DW	128	SW	191
<i>Tr. monococcum</i>	2	FW	110
<i>Tr. turgidum</i>	1		
<i>Tr. spelta</i>	1		
Total	617		617

¹ Species; BW Bread Wheat; DW Durum Wheat² Growth Habit; WW: Winter Wheat, SW: Spring Wheat
FW: Facultative Wheat

Table 3. The number of wheat varieties released by public institutes and universities in Turkey.

No	Institutions and Universities	Number of Varieties
1	Central Research Institute for Field Crops, Ankara	49
2	Transitional Zone Agricultural Research Institute (ARI), Eskişehir	46
3	Maize Research Institute, Sakarya	41
4	East Mediterranean ARI, Adana	30
5	Bahri Dagdas International ARI, Konya	27
6	Aegean ARI, Izmir	25
7	GAP International ARI, Diyarbakır	23
8	Thrace ARI, Edirne	23
9	State Farms, Ankara	15
10	Eastern Anatolian ARI, Erzurum	11
11	GAP ARI, Şanlıurfa	7
12	Black Sea ARI, Samsun	6
13	Yeşilköy ARI, İstanbul	6
14	West Mediterranean ARI, Antalya	3
15	East Mediterranean Transitional Zone ARI, Kahramanmaraş	1
Total		313
1	Faculty of Agriculture, Ankara Uni., Ankara	6
2	Faculty of Agriculture, Çukurova Uni., Adana	6
3	Faculty of Agriculture, Namık Kemal Uni., Tekirdağ	5
4	Faculty of Agriculture, Harran Uni., Şanlıurfa	2
5	Faculty of Agriculture, Uludağ Uni., Bursa	2
6	Faculty of Agriculture, Dicle Uni., Diyarbakır	1
7	Faculty of Agriculture, Selçuk Uni., Konya	1
Total		23

Table 4. The number of wheat varieties registered by private companies in Turkey.

No	Company Names	Number of Variety	No	Company Names	Number of Variety
1	ProGen Tohum A.Ş.	19	35	Altınbaşak Tohumculuk Ltd.Şti.	2
2	Ekiz Toh.Dan.Ür.Tic.Arş.Proj.Tar. ve Gıd.Ltd.Şti.	15	36	Büke Tarım ve Hay.İth.İhr. ve Tic.Ltd.Şti.	2
3	Tareks Tar.Ür.A.G.İth.İhr.Tic.A.Ş.	15	37	C.T.O Ekin Tarım Ürn.Tic.Ltd.Şti.	2
4	Trakya Tarım ve Vet.Tic.Ltd.Şti.	15	38	CYD Ağaoğlu San.Tic Paz.Ltd.Şti.	2
5	Tasaco Tarım San. ve Tic.Ltd.Şti.	13	39	Dobruca Tohumculuk San.Tic.Ltd.Şti.	2
6	Alfa Toh.Tar.Gıd.İnş.Hay.Paz.San.Tic.Ltd.Şti.	11	40	Kartaş Tohumculuk San. ve Tic.Ltd.Şti.	2
7	Tekfen Tarımsal Araş.Üre. ve Paz.A.Ş.	11	41	May-Agro Toh.San. ve Tic.A.Ş.	2
8	Maro Tarım İnş.Tic. ve San.A.Ş.	10	42	Orhas İç ve Dış Tic.Ltd.Şti.	2
9	Tekcan Toh.Gıda Tarım Ürün.San.Tic.Ltd.Şti.	9	43	OSM Tohumculuk San.Tic.Ltd.Şti.	2
10	Ata Tohumculuk İşl.San. ve Tic.A.Ş.	8	44	Prof. Dr. Turan TATLIOĞLU	2
11	Limagrain Tohum Islah ve Üret.San.Tic.A.Ş	8	45	RTS Tarım Ltd.Şti.	2
12	Tarar Un ve Gıda San.Tic.Ltd.Şti.	8	46	S.S. Akşehir İlgin Pancar Ekicileri Koop.	2
13	Yıldız Bitkisel Ürün.Tohum. ve Tar.San.A.Ş.	8	47	SAPEKSA	2
14	Akçakaya Tarım Tic. Ve San.Ltd. Şti.	7	48	Semillas Fito Tarım San.Tic.A.Ş.	2
15	Caso Tohum San. ve Tic.Ltd.Şti.	6	49	Taşpınar Tarım Tic. ve San.Ltd.Şti.	2
16	13 Yıldız Tohumculuk	5	50	Turtat Tohum Islah Ltd.Şti.	2
17	Agrova Tar. Üretim Paz.Ltd.Şti	5	51	Ankomer Tohum. ve Ziraat San.Tic.Ltd.Şti.	2
18	BC İnstitüte Tar.Ür.Oto.San. ve Tic.Ltd.Şti.	5	52	Ayazlar Toh.Tar.Ürün.İç Dış Tic.San.Ltd.Şti.	1
19	Semila Tohumculuk San. ve Tic.Ltd.Şti.	5	53	Bağlariçi Tohumculuk	1
20	Syngenta Tarım San. ve Tic.A.Ş.	5	54	Ekip Tarım Ltd.Şti.	1
21	Caussade Tohumculuk Tarım Ltd.Şti.	4	55	İsmailoğulları Tar.Ürün.İlaç.Nak.Tah.Ltd. Şti.	1
22	Kuran Tarım Dış.Tic.Ltd.Şti.	4	56	Kalender Gıda Tarım Ltd.Şti.	1
23	LT Tarım Üretim Paz.Tic.Ltd.Şti.	4	57	Kappadokia Tarım Ltd.Şti.	1
24	Ziya Organik Tarım A.Ş.	4	58	Marmara Tohum Geliştirme A.Ş.	1
25	Avesa Tarım Gıda ve Hay.Ltd.Şti.	3	59	Özbuğday Tar.İşl. ve Toh.A.Ş.	1
26	Çamlıca Tohumculuk Tic.Ltd.Şti.	3	60	Pankobirlik Kooperatifleri Birliği	1
27	İ.T.U. ve Tar.İlaç Toh.Paz.San. ve Tic.Ltd.Şti.	3	61	Rıfatoğlu Tar.Ür.Petr.Nak.İnş.Mad.San.Tic.Ltd.Şti.	1
28	Meya Tohum Dan.San.Tic.Ltd.Şti.	3	62	Safgen Tohumculuk San. Tic.Ltd. Şti.	1
29	NBC Tarım Ltd.Şti.	3	63	Sarı Tohumculuk San. ve Tic.Ltd.Şti.	1
30	Olgunlar Tur.Tar.Enerji Üret.Tic.Paz.Ltd.Şti.	3	64	SASEED Tar. Tic.Ltd.Şti.	1
31	Utek Tarım İnş. Gıda San. Tic.Ltd.Şti.	3	65	Silvan Nergiz San. Tic.Ltd.Şti.	1
32	Agro Teknik Zir.Ür.San.Tic.A.Ş.	2	66	Söke Tohum Ltd. Şti.	1
33	Aksoy Turizm ve Gıda San.Tic.Ltd.Şti.	2	67	Trakya Genetik Arge Dan.Ür.İth.İhr.Paz.Ltd.Şti.	1
34	Alkan İnş.Müh.Tur.Taş.San. ve Tic.Ltd. Şti.	2			

Table 5. The number of wheat varieties released by public institutions and private sector in selected countries in 2016-2021.

Year/ Institution	Hungary			Iran			Turkey		UK	
	Public	Private	Hungary Total	Public	Private	Iran Total	Public	Private	Turkey Total	Private Total
2016	4	11	15	4	0	4	10	26	36	10
2017	4	12	16	4	4	8	5	44	49	23
2018	6	5	11	9	3	12	12	20	32	54
2019	4	8	12	9	0	9	21	20	41	56
2020	6	6	12	8	10	18	27	34	61	28
2021	na	na	na	14	2	16	17	41	58	29
Grand Total	24	42	66	48	19	67	92	185	277	200

Table 6. The number of varieties released in different periods in Turkey.

Period	Number of Variety Released	Year	Var/Year
1928-1963	19	39	0.5
1964-2006	190	43	4.4
2007-2021	408	16	25.5
Total/Average	617	97	6.4

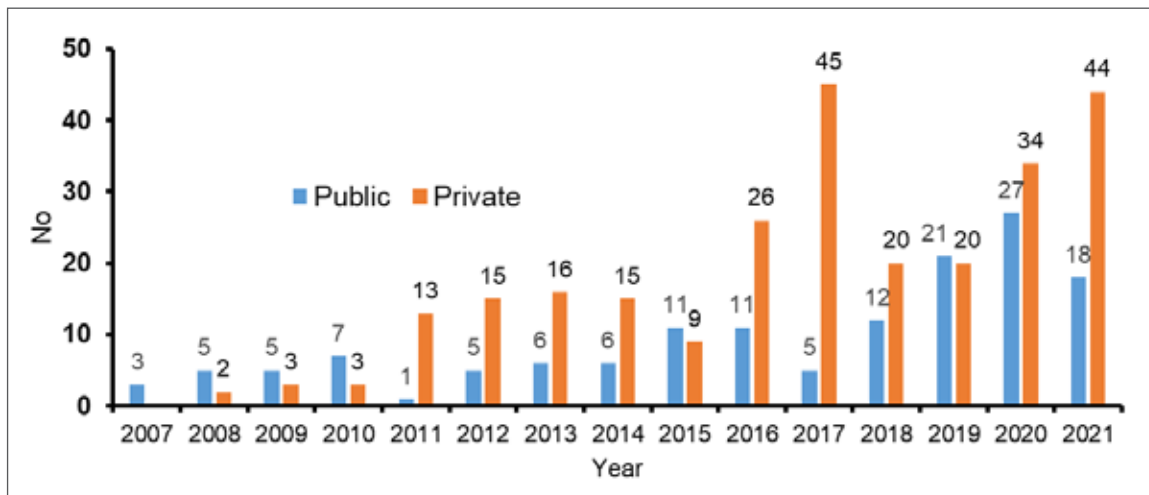


Figure 1. Wheat variety release by the public and private sectors between 2007 and 2021, after the Seed Law no 5043 was published in 2006.

Annex 1. Wheat varieties released in Turkey from the beginning of wheat breeding to 2021.

No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵	No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵
1	Karakılıçık1133	DW	1928	Public	S	TR	50	Bolal2973	BW	1970	Public	W	US
2	Sarı710	DW	1929	Public	F	TR	51	Kıraç 66	BW	1970	Public	W	TR
3	Ak702	BW	1931	Public	F	TR	52	Etoil De Choisy	BW	1975	Public	S	FR
4	Sivas111-33	BW	1933	Public	W	TR	53	Tosun144	BW	1975	Univ	W	TR
5	Cumhuriyet Buğdayı	BW	1936	Public	S	TR	54	Tosun21	BW	1975	Univ	W	TR
6	Kızılca6451	BW	1936	Public	S	TR	55	Tosun22	BW	1975	Univ	W	TR
7	Sertak52	BW	1936	Public	F	TR	56	Cumhuriyet75	BW	1976	Public	S	TR
8	Köse-220-39	BW	1939	Public	F	TR	57	Dicle74	DW	1976	Public	S	TR
9	Yayla305	BW	1939	Public	W	TR	58	Gediz75	DW	1976	Public	S	TR
10	Akbaşak7194	DW	1943	Public	W	TR	59	Porsuk2800	BW	1976	Public	W	US
11	Akbaşak073-44	DW	1944	Public	W	TR	60	Sakarya75	BW	1976	Public	S	TR
12	Ankara093/44	BW	1944	Public	W	TR	61	Lancer	BW	1977	Public	W	US
13	Kunduru414/44	DW	1944	Public	W	TR	62	Orso	BW	1977	Public	S	IT
14	Melez13	BW	1944	Public	W	TR	63	Çakmak79	DW	1979	Public	W	TR
15	Tunus Buğdayı	BW	1944	Public	W	TR	64	Gerek79	BW	1979	Public	W	TR
16	SürakM1593-51	BW	1951	Public	W	TR	65	Gököl79	DW	1979	Public	W	TR
17	Köse Melez1718	BW	1958	Public	W	TR	66	Haymana79	BW	1979	Public	W	US
18	Kırmızı5132	DW	1963	Univ	W	TR	67	Kırkpınar79	BW	1979	Public	F	TR
19	Sarıbursa7113	DW	1963	Univ	W	TR	68	Tunca79	DW	1979	Public	W	TR
20	185-1	DW	1964	Public	F	TR	69	Ata81	BW	1985	Public	S	TR
21	4-11	BW	1964	Public	W	TR	70	Atay85	BW	1985	Public	W	TR
22	Akova B-1	BW	1964	Public	S	TR	71	İzmir85	BW	1985	Public	S	TR
23	Mentana B-1	BW	1964	Public	S	IT	72	Çukurova86	BW	1986	Public	S	TR
24	4-22	BW	1966	Public	F	TR	73	Marmara86	BW	1986	Public	S	TR
25	Floransa N4-8	BW	1966	Public	S	AU	74	Diyarbakır81	DW	1987	Public	S	TR
26	P-8-6	BW	1966	Public	W	TR	75	Kop	BW	1987	Public	S	TR
27	P-8-8	BW	1966	Public	W	TR	76	Balçalı85	DW	1988	Univ	S	TR
28	Berkmen469	DW	1967	Public	W	TR	77	Ege88	DW	1988	Public	S	TR
29	Kunduru1149	DW	1967	Public	W	TR	78	Genç88	BW	1988	Univ	S	TR
30	Aköz 867	BW	1968	Public	S	TR	79	Kaklıç88	BW	1988	Public	S	TR
31	Bezostaja1	BW	1968	Public	W	RU	80	KateA-1	BW	1988	Public	W	BG
32	Burt	BW	1968	Public	W	US	81	Creso	DW	1989	Public	S	IT
33	Gaines	BW	1968	Public	W	US	82	Doğu88	BW	1990	Public	W	US
34	Inia66	BW	1968	Public	S	MX	83	Karasu90	BW	1990	Public	W	TR
35	Jaral66	BW	1968	Public	S	MX	84	Doğankent1	BW	1991	Public	S	TR
36	Lerma Rojo64	BW	1968	Public	S	MX	85	Gün91	BW	1991	Public	F	TR
37	Mayo64	BW	1968	Public	S	MX	86	Kızıltan91	DW	1991	Public	F	TR
38	Nadadores63	BW	1968	Public	S	MX	87	Murat1	BW	1991	Public	W	TR
39	Noroeste66	BW	1968	Public	S	MX	88	Seri82	BW	1991	Public	S	MX
40	Oviachic65	DW	1968	Public	S	MX	89	Sham1	DW	1991	Public	S	SYR
41	Penjamo62	BW	1968	Public	S	MX	90	Salihli92	DW	1992	Public	S	TR
42	Pitic62	BW	1968	Public	S	MX	91	Dağdaş94	BW	1994	Public	W	TR
43	Siete Cerros66	BW	1968	Public	S	MX	92	Kutluk94	BW	1994	Public	F	TR
44	Sonora63	BW	1968	Public	S	MX	93	Altıntaş95	DW	1995	Public	W	TR
45	SuperX	BW	1968	Public	S	MX	94	BasriBey95	BW	1995	Public	S	TR
46	Tevere	BW	1968	Public	W	IT	95	Başak95	BW	1995	Univ	W	TR
47	Tobari66	BW	1968	Public	S	MX	96	Ceylan95	DW	1995	Public	S	TR
48	Wanser	BW	1968	Public	W	US	97	Harran95	DW	1995	Public	S	TR
49	Yektay406	BW	1968	Public	W	IT	98	KaşifBey 95	BW	1995	Public	S	TR

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No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵	No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵
99	Kırgız95	BW	1995	Public	W	TR	148	Momtchill	BW	2000	Public	W	BG
100	Seyhan95	BW	1995	Public	S	TR	149	Tahirova2000	BW	2000	Public	S	TR
101	Sultan95	BW	1995	Public	W	TR	150	Yelken2000	DW	2000	Public	W	TR
102	İkizce96	BW	1996	Public	F	TR	151	Alparslan	BW	2001	Public	W	TR
103	Lirasa92	BW	1996	Private	S	MX	152	Alpu2001	BW	2001	Public	W	TR
104	Amanos97	DW	1997	Public	S	TR	153	Attila12	BW	2001	Public	W	HU
105	Bandırma97	BW	1997	Public	S	TR	154	Centauro	BW	2001	Private	S	IT
106	Karacabey97	BW	1997	Public	F	TR	155	İzgi2001	BW	2001	Public	F	TR
107	Kınacı97	BW	1997	Public	W	TR	156	Köksal2000	BW	2001	Univ	S	TR
108	Palandöken97	BW	1997	Public	W	TR	157	Martar	BW	2001	Private	W	DE
109	Pamukova97	BW	1997	Public	S	TR	158	Nenchatun	BW	2001	Public	W	TR
110	Selçuklu97	DW	1997	Public	F	TR	159	Nurkent	BW	2001	Public	S	TR
111	Süzen 97	BW	1997	Public	F	TR	160	Pandas	BW	2001	Public	S	IT
112	Altın40/98	DW	1998	Public	W	TR	161	Pınar2001	DW	2001	Univ	F	TR
113	Altıntoprak98	DW	1998	Public	S	TR	162	Sagittario	BW	2001	Private	S	IT
114	Ankara98	DW	1998	Public	F	TR	163	Saraybosna01	BW	2001	Public	W	SI
115	Aytın98	BW	1998	Public	W	TR	164	Sönmez2001	BW	2001	Public	W	TR
116	Gönen98	BW	1998	Public	S	TR	165	Svevo	DW	2001	Private	S	IT
117	Karacadağ98	BW	1998	Public	S	TR	166	Turan	BW	2001	Private	S	DE
118	Mızrak98	BW	1998	Public	F	TR	167	Zenit	DW	2001	Private	S	IT
119	Pehlivan	BW	1998	Public	W	TR	168	Akçakale2000	DW	2002	Public	S	TR
120	Sarıçanak98	DW	1998	Public	S	TR	169	Atlı2002	BW	2002	Public	F	TR
121	Türkmen	BW	1998	Public	F	TR	170	Aydın93	DW	2002	Public	S	TR
122	Uzunyayla	BW	1998	Public	W	TR	171	Bağcı2002	BW	2002	Public	W	TR
123	Yıldız98	BW	1998	Public	W	TR	172	Daphan	BW	2002	Public	W	TR
124	Yılmaz98	DW	1998	Public	W	TR	173	Dariel	BW	2002	Private	S	IL
125	Ziyabey98	BW	1998	Public	S	TR	174	Fırat93	DW	2002	Public	S	TR
126	Adana99	BW	1999	Public	S	TR	175	Galil	BW	2002	Private	S	IL
127	Ceyhan99	BW	1999	Public	S	TR	176	Konya2002	BW	2002	Public	W	TR
128	Çeşit1252	DW	1999	Public	W	TR	177	Meram2002	DW	2002	Public	F	TR
129	Flamura85	BW	1999	Public	W	RO	178	Meta2002	BW	2002	Public	S	TR
130	Genç99	BW	1999	Univ	S	TR	179	Negev	BW	2002	Private	S	IL
131	Golia	BW	1999	Public	F	IT	180	Sakin	BW	2002	Public	S	TR
132	Göksu99	BW	1999	Public	W	TR	181	Soyer02	BW	2002	Public	F	TR
133	Harmankaya99	BW	1999	Public	W	RO	182	Şölen2002	DW	2002	Public	S	TR
134	Karahan99	BW	1999	Public	W	TR	183	Tüten2002	DW	2002	Public	S	TR
135	Proştor	BW	1999	Public	W	BG	184	Yıldırım	BW	2002	Public	W	TR
136	Saroz95	BW	1999	Public	W	TR	185	Yüreğir89	BW	2002	Public	S	TR
137	Yakar99	BW	1999	Public	F	TR	186	Zencirci2002	BW	2002	Public	F	TR
138	Aksel2000	BW	2000	Public	F	TR	187	Canik2003	BW	2003	Public	F	TR
139	Altay 2000	BW	2000	Public	W	TR	188	Drophia	BW	2003	Private	W	RO
140	Balatilla	BW	2000	Univ	S	TR	189	Eser	BW	2003	Public	F	TR
141	Balçalı2000	DW	2000	Univ	S	TR	190	Özdemirbey97	BW	2003	Private	S	TR
142	Bayraktar2000	BW	2000	Public	W	TR	191	Ahmetağa	BW	2004	Public	F	TR
143	Çetinel2000	BW	2000	Public	W	TR	192	Alibey	BW	2004	Public	S	TR
144	Demir2000	BW	2000	Public	F	TR	193	Ekiz	BW	2004	Public	F	TR
145	Fuatbey2000	DW	2000	Public	S	TR	194	Gap	DW	2004	Public	S	TR
146	Kümbet2000	DW	2000	Public	W	TR	195	Menemen	BW	2004	Public	S	TR
147	Mirzabey2000	DW	2000	Public	F	TR	196	Özcan	BW	2004	Public	S	TR

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No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵	No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵
197	Seval	BW	2004	Public	W	TR	246	Geyal	BW	2011	Private	W	BG
198	Tosunbey	BW	2004	Public	F	TR	247	Levante	DW	2011	Private	S	IT
199	Turabi	DW	2004	Public	S	TR	248	Mane Nick	BW	2011	Private	S	ES
200	Gelibolu	BW	2005	Public	W	TR	249	OS Alka	BW	2011	Private	W	HR
201	Nina	BW	2005	Private	W	HR	250	Pinzon	BW	2011	Private	S	ES
202	Özberk	DW	2005	Univ	S	TR	251	Saragolla	DW	2011	Private	S	IT
203	Tekirdağ	BW	2005	Public	W	TR	252	Aglıka	BW	2012	Private	W	BG
204	Tina	BW	2005	Private	W	HR	253	Altındane	BW	2012	Public	S	TR
205	Urfa2005	DW	2005	Univ	S	TR	254	Andino	BW	2012	Private	W	FR
206	Dumlupınar	DW	2006	Public	W	TR	255	B 52	BW	2012	Private	na	na
207	Karatopak	BW	2006	Public	S	TR	256	Bisante	DW	2012	Private	F	TR
208	Müfitbey	BW	2006	Public	W	TR	257	Charly Nick	BW	2012	Private	na	na
209	Osmaniyem	BW	2006	Public	S	TR	258	Eraybey	BW	2012	Public	W	TR
210	Beşköprü	BW	2007	Public	F	TR	259	Forblanc	BW	2012	Private	W	IT
211	Guadalupe	BW	2007	Public	F	FR	260	Gündaş	DW	2012	Public	S	TR
212	Hanlı	BW	2007	Public	S	TR	261	Iridium	BW	2012	Private	W	FR
213	Artuklu	DW	2008	Public	S	TR	262	LG59	BW	2012	Private	W	ES
214	Cemre	BW	2008	Public	S	TR	263	May8059	BW	2012	Private	W	BG
215	Eyyubi	DW	2008	Public	S	TR	264	Quality	BW	2012	Private	W	IT
216	Krasunia odes'ka	BW	2008	Private	W	UA	265	Rumeli	BW	2012	Private	W	TR
217	Nacibey	BW	2008	Public	W	TR	266	Soylu	DW	2012	Univ	W	TR
218	Syrena odes'ka	BW	2008	Private	W	UA	267	Turkuaz	BW	2012	Private	W	TR
219	Şahinbey	DW	2008	Public	S	TR	268	Vasilina	BW	2012	Private	W	UA
220	Aldane	BW	2009	Public	F	TR	269	Vittorio	BW	2012	Private	F	IT
221	Eminbey	DW	2009	Public	W	TR	270	Yunus	BW	2012	Public	W	TR
222	Hakan	BW	2009	Private	W	TR	271	Zıtka	DW	2012	Private	F	RS
223	İmren	DW	2009	Public	F	TR	272	Adagio	BW	2013	Private	W	FR
224	Kaan	BW	2009	Private	W	TR	273	Adelaide	BW	2013	Private	F	IT
225	Kenanbey	BW	2009	Public	F	TR	274	Altın Başak	BW	2013	Public	S	TR
226	Selimiye	BW	2009	Public	W	TR	275	Antille	BW	2013	Private	W	IT
227	Yunak	BW	2009	Private	W	BG	276	Artico	BW	2013	Private	W	IT
228	Bereket	BW	2010	Public	W	TR	277	Avorio	BW	2013	Private	W	IT
229	Colfiorito	BW	2010	Private	W	IT	278	Bona dea	BW	2013	Private	W	SK
230	ES26	BW	2010	Public	W	TR	279	Casanova	DW	2013	Private	W	IT
231	Güney Yıldızı	DW	2010	Public	S	TR	280	Cesare	DW	2013	Private	F	IT
232	Kırık	BW	2010	Public	F	TR	281	Diñç	BW	2013	Public	S	TR
233	Lütfibey	BW	2010	Public	W	TR	282	Geronimo	BW	2013	Private	W	IT
234	May8462	BW	2010	Private	W	na	283	Gökkan	BW	2013	Public	S	TR
235	Özkan	BW	2010	Univ	S	TR	284	Kırkayak	BW	2013	Private	W	TR
236	TT601	BW	2010	Private	W	TR	285	Mesut	BW	2013	Public	F	TR
237	Zühre	DW	2010	Public	S	TR	286	Nota	BW	2013	Private	W	RU
238	Anapo	BW	2011	Private	F	IT	287	Pitagora	DW	2013	Private	S	IT
239	Ayyıldız	BW	2011	Public	W	TR	288	Renan	BW	2013	Private	W	FR
240	Botticelli	BW	2011	Private	W	IT	289	Sarı Başak	DW	2013	Public	S	TR
241	Burgos	DW	2011	Private	S	ES	290	Segor	BW	2013	Private	W	CH
242	Carisma	BW	2011	Private	W	IT	291	Seri2013	BW	2013	Public	S	TR
243	Claudio	DW	2011	Private	S	IT	292	Stendal	BW	2013	Private	W	IT
244	Cömert2	BW	2011	Private	W	TR	293	Yubileynaya100	BW	2013	Private	W	RU
245	Esperia	BW	2011	Private	W	IT	294	Anforeta	BW	2014	Private	W	IT

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No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵	No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵
295	Astet	BW	2014	Private	W	UA	344	GK Szala	BW	2016	Private	W	HU
296	Azul	BW	2014	Private	W	IT	345	Kaynarca	BW	2016	Public	S	TR
297	Biensur	DW	2014	Private	S	FR	346	Kayra	BW	2016	Public	F	TR
298	Bora	BW	2014	Private	W	IT	347	Leuta	BW	2016	Private	W	HR
299	Bozkır	BW	2014	Public	W	TR	348	Maden	BW	2016	Private	W	TR
300	Delabrad2	BW	2014	Private	W	RO	349	MassimoMeridio	DW	2016	Private	S	IT
301	Faur F	BW	2014	Private	W	RO	350	Miriana	BW	2016	Private	W	BG
302	Galateya	BW	2014	Private	W	BG	351	Natula	BW	2016	Private	W	PL
303	Genesi	BW	2014	Private	W	IT	352	NKU Ergene	BW	2016	Univ	W	TR
304	Glosa	BW	2014	Private	W	RO	353	NKU Lider	BW	2016	Univ	W	TR
305	Kharus	BW	2014	Private	W	UA	354	Nova	DW	2016	Private	F	TR
306	Masaccio	BW	2014	Private	F	IT	355	NS 40S	BW	2016	Private	W	RS
307	Metin	BW	2014	Public	F	TR	356	Oscar	BW	2016	Private	F	TR
308	Midas	BW	2014	Private	W	AT	357	Reis	BW	2016	Public	W	TR
309	Mihelca	BW	2014	Private	W	HR	358	Renata	BW	2016	Private	W	HR
310	Nevzatbey	BW	2014	Public	W	TR	359	Sobald	BW	2016	Private	W	FR
311	Prima	BW	2014	Private	W	HR	360	Sollario	BW	2016	Private	W	FR
312	Saban	BW	2014	Public	W	TR	361	Şanlı	BW	2016	Public	F	TR
313	Tekin	BW	2014	Public	S	TR	362	Tekir	BW	2016	Private	F	TR
314	Yakamoz	BW	2014	Public	S	TR	363	Toros1003	BW	2016	Private	S	TR
315	Alada	BW	2015	Public	F	TR	364	Trionfo	DW	2016	Private	S	IT
316	Alatay	DW	2015	Public	S	TR	365	Tripudio	DW	2016	Private	S	IT
317	Altınöz	BW	2015	Public	S	TR	366	Venka1	BW	2016	Private	W	BG
318	Ayzer	DW	2015	Public	S	TR	367	Yaren	DW	2016	Public	S	TR
319	Cendere	BW	2015	Private	S	TR	368	Yörük	BW	2016	Private	F	TR
320	Efe	BW	2015	Public	S	TR	369	Yüksel	BW	2016	Public	W	TR
321	Eker	DW	2015	Public	S	TR	370	Zümrüt	BW	2016	Private	F	TR
322	Hamza	BW	2015	Private	W	TR	371	Sena	DW	2016	Univ	S	TR
323	Hasanbey	DW	2015	Public	S	TR	372	Aliğa	BW	2017	Public	W	TR
324	Kale	BW	2015	Public	S	TR	373	Alp1	BW	2017	Private	F	TR
325	Köprü	BW	2015	Public	W	TR	374	Andalusia	BW	2017	Private	W	IT
326	Maestrale	DW	2015	Private	S	IT	375	Aslı	BW	2017	Private	W	AT
327	NKÜZiraat	DW	2015	Univ	F	TR	376	Candaş	BW	2017	Public	S	TR
328	Nusrat	BW	2015	Public	F	TR	377	Çeşmeli	BW	2017	Private	W	TR
329	Perre	DW	2015	Private	S	TR	378	Çıfçıklı	BW	2017	Private	S	TR
330	Sarımustafa	BW	2015	Private	W	TR	379	Dunaviya	BW	2017	Private	W	BG
331	Sertori	BW	2015	Private	W	CH	380	Duru17	BW	2017	Private	W	HR
332	Solveig	BW	2015	Private	F	FR	381	Ecem	DW	2017	Private	S	TR
333	Tigre	BW	2015	Private	S	FR	382	Edessa	DW	2017	Public	S	TR
334	Uniya	DW	2015	Private	W	RU	383	Ekinoks	BW	2017	Public	S	TR
335	Acar	BW	2016	Public	F	TR	384	Energo	BW	2017	Private	W	DE
336	Altuğ	BW	2016	Public	F	TR	385	Flamenko	BW	2017	Private	W	FR
337	Ant	BW	2016	Private	F	TR	386	Ghayta	BW	2017	Private	W	FR
338	Asuncion	BW	2016	Private	W	IT	387	Guappo vst	BW	2017	Private	W	IT
339	Beğendik	BW	2016	Private	W	TR	388	Güdük	BW	2017	Private	W	TR
340	Boema1	BW	2016	Private	W	RO	389	Günberi	DW	2017	Public	S	TR
341	Destan	BW	2016	Private	W	TR	390	Haciveli	BW	2017	Private	F	TR
342	Enola	BW	2016	Private	W	BG	391	Havabacı	BW	2017	Private	W	TR
343	Ganos	DW	2016	Private	W	TR	392	Hendrix	BW	2017	Private	W	FR

Continuing Annex 1

No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵	No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵
393	Hisar	BW	2017	Private	W	TR	442	Setan	BW	2018	Private	S	TR
394	Hüseyinbey	BW	2017	Private	W	TR	443	Severina	DW	2018	Private	W	BG
395	İkbal	BW	2017	Private	S	TR	444	Sultançayır	BW	2018	Private	F	TR
396	Lazarka	BW	2017	Private	W	BG	445	Sümerli	DW	2018	Public	S	TR
397	Lucilla	BW	2017	Private	F	IT	446	Şehzade1	BW	2018	Public	F	TR
398	Maria	BW	2017	Private	W	UA	447	Taner	BW	2018	Public	F	TR
399	Maya	BW	2017	Private	W	TR	448	Tekfen 1013	BW	2018	Private	S	TR
400	Merilin	BW	2017	Private	W	BG	449	Topkapı	BW	2018	Private	W	AT
401	Michelangelo	BW	2017	Private	W	IT	450	Türköz1	DW	2018	Public	W	TR
402	Mimmo	DW	2017	Private	F	IT	451	Viktoria	BW	2018	Private	W	HR
403	Misiia Odes'ka	BW	2017	Private	W	UA	452	Wafia	BW	2018	Private	S	FR
404	Musik	BW	2017	Private	W	FR	453	ZT Ziyade	BW	2018	Private	W	TR
405	Mv Taller	BW	2017	Private	W	HU	454	Abide	BW	2019	Public	W	TR
406	Nomade	BW	2017	Private	W	IT	455	Adonis	BW	2019	Private	S	TR
407	OS Jelena	BW	2017	Private	W	HR	456	Ahtska	BW	2019	Public	W	TR
408	Pannonia	BW	2017	Private	W	RS	457	Aleppo	BW	2019	Private	W	IT
409	Paşa	BW	2017	Private	W	TR	458	Almeria	BW	2019	Private	S	IT
410	Pelit	BW	2017	Private	F	TR	459	Anafarta	BW	2019	Public	W	TR
411	Rebelde	BW	2017	Private	W	IT	460	Annie	BW	2019	Private	W	CZ
412	Ronsard	BW	2017	Private	W	FR	461	Anzele	DW	2019	Private	S	TR
413	Sankerim	BW	2017	Private	F	TR	462	Arin	BW	2019	Private	S	TR
414	Skerzzo	BW	2017	Private	W	FR	463	Ayten Abla	BW	2019	Public	F	TR
415	Soledad	BW	2017	Private	W	FR	464	Başkurt	BW	2019	Public	W	TR
416	Tiziana	DW	2017	Private	F	IT	465	Beyazhan	BW	2019	Private	S	TR
417	Troubadur	DW	2017	Private	W	AT	466	Bilden	BW	2019	Public	W	TR
418	Yiğit	BW	2017	Private	W	TR	467	Bohemia	BW	2019	Private	W	CZ
419	Zlatoglava	BW	2017	Private	W	UA	468	Calumet	BW	2019	Public	W	FR
420	Zolotko	DW	2017	Private	W	RU	469	Çavuş	BW	2019	Public	W	TR
421	Perfekta	BW	2017	Private	W	RS	470	Demirhan	BW	2019	Public	F	TR
422	Adalı	BW	2018	Public	W	TR	471	Ginseng	DW	2019	Private	S	FR
423	Albachiara	BW	2018	Private	W	IT	472	Izvor	BW	2019	Private	W	RO
424	Argeles	DW	2018	Private	S	FR	473	İlkhan	DW	2019	Public	S	TR
425	Bc Anica	BW	2018	Private	W	HR	474	Karakalpak	BW	2019	Public	W	TR
426	Bojana	BW	2018	Private	W	BG	475	Kıpçak	BW	2019	Public	W	TR
427	Damla	BW	2018	Public	W	TR	476	Lavoisier	BW	2019	Public	W	FR
428	Dragana	BW	2018	Private	W	BG	477	Mario	DW	2019	Private	S	FR
429	FDL Miranda	BW	2018	Private	W	RO	478	Mirsa	BW	2019	Public	F	TR
430	Halis	BW	2018	Public	F	TR	479	Ovidio	DW	2019	Private	S	IT
431	Hamitbey	BW	2018	Public	W	TR	480	Pandiya	BW	2019	Private	W	UA
432	Iveta	BW	2018	Private	W	BG	481	Peçenek	BW	2019	Public	F	TR
433	Kışla	BW	2018	Public	F	TR	482	Polathan	BW	2019	Public	S	TR
434	Koç2015	BW	2018	Public	S	TR	483	Poyraz	DW	2019	Public	S	TR
435	Maja	BW	2018	Private	W	HR	484	Salgado	DW	2019	Private	F	IT
436	Meltem	BW	2018	Public	S	TR	485	Somuncuoğlu	BW	2019	Public	W	TR
437	Milandur	DW	2018	Private	W	AT	486	Stoyana	BW	2019	Private	W	BG
438	Muna	BW	2018	Private	S	TR	487	Tekfen1016	BW	2019	Private	S	TR
439	NKU Asiya	BW	2018	Univ	W	TR	488	Tekfen2038	BW	2019	Private	F	TR
440	Oktan	BW	2018	Private	S	TR	489	Teodorico	DW	2019	Private	S	IT
441	Otilia	BW	2018	Private	W	RO	490	Tigris	DW	2019	Private	S	TR

Continuing Annex 1

No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵	No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵
491	Tuğra	BW	2019	Public	W	TR	540	NKÜ Zirve	BW	2020	Univ	W	TR
492	Vehbibey	DW	2019	Public	W	TR	541	Payitaht	BW	2020	Private	W	TR
493	Waximum	BW	2019	Private	W	FR	542	Perge 07	BW	2020	Public	S	TR
494	Yavuz	BW	2019	Public	F	TR	543	Seki 07	BW	2020	Public	S	TR
495	Activus	BW	2020	Private	W	AT	544	Selamibey	BW	2020	Public	F	TR
496	Afra	BW	2020	Private	W	TR	545	Selçuklu	BW	2020	Public	W	TR
497	Akça	BW	2020	Private	F	TR	546	Sırçalı	DW	2020	Public	F	TR
498	Akmira	BW	2020	Private	W	TR	547	Simge	BW	2020	Public	S	TR
499	Aksungur	BW	2020	Public	W	TR	548	Şahika	BW	2020	Public	F	TR
500	Alturna	BW	2020	Public	W	TR	549	Teb0693	BW	2020	Private	W	TR
501	Arifbey	BW	2020	Public	W	TR	550	Tekfen1039	BW	2020	Private	F	TR
502	Aurelia	BW	2020	Private	W	HU	551	Tekfen2001	BW	2020	Private	W	TR
503	Ayaz	BW	2020	Public	F	TR	552	Tekfen2077	BW	2020	Private	W	TR
504	Babil	BW	2020	Private	S	TR	553	Tekfen2095	BW	2020	Private	F	TR
505	Bagira	BW	2020	Public	W	RU	554	Yusuf Bey	BW	2020	Private	W	TR
506	Bayındır	BW	2020	Public	W	TR	555	Zoria Ukrayna	<i>Tr. spelta</i>	2020	Private	W	UA
507	Bayram	BW	2020	Public	F	TR	556	Ağabey	DW	2021	Private	S	TR
508	Beyhan	BW	2020	Public	W	TR	557	Akçabey	BW	2021	Private	W	TR
509	Buhara	BW	2020	Public	W	TR	558	Albaşak	BW	2021	Public	W	TR
510	Cemile	BW	2020	Private	F	TR	559	Albayrak	BW	2021	Private	W	TR
511	Çağdaş	BW	2020	Private	W	TR	560	Armağan	BW	2021	Public	F	TR
512	Dekatlon	BW	2020	Private	F	TR	561	Atasiyez	<i>Tr. monoc.</i>	2021	Public	F	TR
513	Duronese	DW	2020	Private	S	DE	562	Avşar	BW	2021	Public	F	TR
514	Durusa	DW	2020	Public	S	TR	563	Badin	DW	2021	Private	S	TR
515	Ettore	DW	2020	Private	F	IT	564	Balkoni	DW	2021	Private	F	TR
516	Eylül	BW	2020	Public	W	TR	565	Bengisu	BW	2021	Public	W	TR
517	Ezgi	BW	2020	Private	W	TR	566	Beyaz1	BW	2021	Private	F	TR
518	Fazılbe	BW	2020	Public	W	TR	567	Bisanzio	BW	2021	Private	F	IT
519	Forcali	BW	2020	Private	W	FR	568	Boldane	BW	2021	Private	F	TR
520	Germanicia	BW	2020	Public	S	TR	569	Bolkar	BW	2021	Private	W	TR
521	GK Futar	BW	2020	Private	W	HU	570	Bozok	BW	2021	Public	W	TR
522	Güçlü	BW	2020	Private	W	TR	571	Cudi63	DW	2021	Public	S	TR
523	Hünkar	BW	2020	Private	W	TR	572	Cumakale	DW	2021	Private	S	TR
524	İkonya	BW	2020	Public	W	TR	573	Dar	BW	2021	Private	F	IL
525	Karmen	BW	2020	Public	F	TR	574	Enduro	DW	2021	Private	F	TR
526	Khersons'ka99	BW	2020	Private	W	UA	575	Enerji	BW	2021	Private	W	TR
527	Kirve	BW	2020	Public	S	TR	576	Erbaş	BW	2021	Public	F	TR
528	Kobra59	BW	2020	Private	W	TR	577	Esmeray	BW	2021	Private	W	TR
529	Kürşad	BW	2020	Public	F	TR	578	Eymenbey	BW	2021	Private	S	TR
530	Levent	DW	2020	Public	W	TR	579	Fadime Ana	BW	2021	Private	W	TR
531	Meke	BW	2020	Public	W	TR	580	Falado	BW	2021	Private	W	FR
532	Metropolis	BW	2020	Private	W	IT	581	Grador	DW	2021	Private	S	ES
533	Mv Kepe	BW	2020	Private	W	HU	582	Helina	BW	2021	Private	W	BG
534	Mv Kolo	BW	2020	Private	W	HU	583	Hilar	BW	2021	Public	S	TR
535	Mv Lucilla	BW	2020	Private	W	HU	584	İsmetbey	BW	2021	Private	W	TR
536	Mv Nemere	BW	2020	Private	W	HU	585	Kafkas	<i>Tr. turgidum</i>	2021	Public	F	TR
537	Mv Pantlika	BW	2020	Private	W	HU	586	Karaduman	BW	2021	Public	W	TR
538	Mv Toldi	BW	2020	Private	W	HU	587	Karolina5	BW	2021	Private	W	RU
539	Freeman	BW	2020	Private	W	US	588	Kılınç	BW	2021	Public	W	TR

Continuing Annex 1

No	Variety	Spp. ¹	Rel. Year ²	Rel. By ³	GH ⁴	Origin ⁵
589	Koç1453	BW	2021	Public	W	TR
590	Lancillotto	BW	2021	Private	W	FR
591	Mergüze	<i>Tr. monoc.</i>	2021	Public	F	TR
592	MesutÖzen	BW	2021	Private	W	TR
593	Meya1	DW	2021	Private	S	TR
594	Meya2	DW	2021	Private	S	TR
595	Meya3	BW	2021	Private	S	TR
596	Nizar	BW	2021	Private	S	TR
597	NS Mila	BW	2021	Private	W	RS
598	NS Obala	BW	2021	Private	W	RS
599	Pamira	BW	2021	Private	S	TR
600	Ramisbey	BW	2021	Public	W	TR
601	Refikbey	BW	2021	Private	W	TR
602	Saco16	BW	2021	Private	S	TR
603	Saco2018	DW	2021	Private	S	TR
604	Selenivka	BW	2021	Private	W	UA
605	Serince	BW	2021	Public	S	TR
606	Sy Atlante	DW	2021	Private	S	IT
607	Sy Leonardo	DW	2021	Private	S	IT
608	Tekfen2064	BW	2021	Private	W	TR
609	Tekfen2239	BW	2021	Private	W	TR
610	Tılsım	BW	2021	Private	F	TR
611	Tragen103	BW	2021	Private	W	TR
612	Tufan	BW	2021	Private	S	TR
613	Zorlu	BW	2021	Public	F	TR
614	Biryıldız	BW	2021	Private	W	TR
615	İkiyıldız	BW	2021	Private	W	TR
616	Tekfen2104	BW	2021	Private	W	TR
617	Altınsa	DW	2021	Public	S	TR

¹ Species; BW Bread Wheat; DW Durum Wheat

² Year of Release

³ Released by

⁴ Growth Habit; GH is assigned to any of the variety based on the registration report of Variety Registration and Seed Certification Center, Website of the variety owner or GH given in the some scientific studies and Thesis

⁵ Origin; is assigned to any of the variety based on the registration report of Variety Registration and Seed Certification Center, Website of the variety owner or from the some scientific studies and Thesis

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Pre-Breeding Evaluation of Germplasm for Hybridization and Screening of Resulting Transgressive Elite Genotypes Faba Bean for Yield and its Attributes for Semi-Arid Regions of India

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ABSTRACT

A long term research investigation was carried out for identification of elite genotypes from germplasm and their utilization in the development of high yielding variety through hybridization followed by pedigree method to obtain desirable transgressive segregants of Faba bean at MAP Section, Department of Genetics and Plant Breeding, CCS Haryana Agricultural University, Hisar during winter season from 2004-18. In the present investigation, germplasm was screened for seed yield, resistance to biotic stress and other traits during 2004-05. As a result, ten elite genotypes were identified. By using these elite genotypes, forty five F₁ hybrids were developed during 2005-06 and from F₂ up to F₆ generations were evaluated to identify the superior progenies during 2006-11. Later on, the superior transgressive segregants entries were evaluated under various station trials from 2011-15 which led to identification of HB11-12, HB11-15 and HB11-32 as promising genotypes. Subsequently, these genotypes were further evaluated at different locations having diverse agro-ecological conditions for seed yield, quality and resistance against insect pest and disease during 2015-18. On the basis of average yield over all the locations, HB11-12 exhibited 14.95% yield superiority and was also found free from insect pests and disease. Hence, HB11-12 may be recommended for commercial cultivation in plain zones of semi arid regions of India.

Keywords: Faba bean, pre-breeding, hybridization, transgressive segregants, vicine-convicine

Introduction

Vicia faba L. i.e. faba bean also known as broad beans or horse bean is the fourth most widely grown legume crop after pea, chickpea and lentil (FAOSTAT 2021; Maalouf et al. 2019). Grain legumes have been

recognized to have advantages over cereals, both in human diet and agriculture (Foyer et al. 2016; Cooper et al. 2017). Faba bean (*Vicia faba* L.) belongs to the family Fabaceae, however, it is an often cross pollinated crop (Bishnoi et al. 2012). It is mainly grown

as winter season crop and performs well under cool and moist conditions. It is important in crop rotation, as it fixes atmospheric nitrogen in the soil (Köpke and Nemecek, 2010) and is helpful in minimizing the requirement of chemical fertilizers resulting in improving the economics of the succeeding crops (Jensen et al. 2010; Arya, 2018; Arya et al. 2020), therefore, it reduces the environmental burden.

It is mainly used as animal feed in developed countries and also as food for human beings in developing countries. It has high nutritive value due to presence of high lysine, protein, vitamins, minerals and carbohydrates in its seeds (Crépon et al. 2010). Faba bean is gaining importance as a plant source of protein for humans and animals to ensure food and nutritional security at global level (Bishnoi et al. 2015; Multari et al. 2015).

Along with its different uses, potential of high seed production, balanced and high nutritional quality and ability to grow over a broad range of climatic and soil conditions makes faba bean an appropriate crop for sustainable agriculture due to which it has gained greater global attention in recent years (Köpke and Nemecek, 2010). However, its full potential through hybrid breeding still remains unexploited largely due to its unique pollination biology and yield instability (Bishnoi et al. 2015; Arya, 2018; Maalouf et al. 2019).

The process for development of new varieties in faba bean includes introduction and germplasm collection, pure line selection, single plant selection, selection and hybridisation with appropriate modifications (Singh and Bhatt 2012). The faba bean breeding programs revolves around improving yield, increasing resistance/tolerance to biotic and abiotic stresses, adaptation to the intended environment, suitable phenology, and seed quality. Conventional breeding in faba bean is performed by crossing elite parents, followed by multi-stage testing of progeny, identifying superior progeny with respect to specific traits and finally releasing superior varieties.

Moreover, nutritionally rich high yielding varieties resistant/tolerant to insect pests would not only be helpful in enhancing farm profitability (Maalouf et al. 2019) but also elevate the nutritional security. ICARDA has been working in this direction and also developed new cultivars of faba bean for commercial cultivation (Subash and Priya 2012). Therefore, keeping in view the above facts, the identification and utilization of elite genotypes for hybridization to develop high yielding varieties through transgressive segregation was planned.

Materials and Methods

Germplasm screening: In present investigation, 100 germplasm lines including national check Vikrant

were screened for various agronomic traits under AICRN on Potential Crops in augmented block design with spacing of 45cm x 10 cm at Research Farm of Medicinal, Aromatic, and Potential Crops Section, Department of Genetics & Plant Breeding, CCS HAU, Hisar during winter 2004-05.

Hybridization and transgressive segregation:

The transgressive segregation is the outcome of pedigree method, in which genetically diverse genotypes are crossed to combine different genes in one plant, and in later generations' up to F_6 these genes segregate in all possible combinations. An outstanding plant having superior performance over both the parent is the result of transgressive segregation. In present study, only identified elite genotypes (HB180, HB123, EC329675, EC47755, HB430, HB204, EC248710, PRT-12, HB85 and Vikrant) were utilized in crossing programme and 45 F_1 crosses were made during winter 2005-06 and F_1 seed was produced in next season. During winter 2007-08, F_2 progenies of all the crosses were grown and individual superior plants identified from the segregating populations were harvested separately. The F_3 to F_5 generations were also evaluated to identify the superior transgressive segregants through the process of critical selection or rejection during 2008-09 to 2010-11. Eventually, in F_6 generation, superior progenies of 14 crosses (Table 1) were bulked and evaluated in PRT for yield performance against the national check, Vikrant during 2011-12. Likewise, these genotypes were also evaluated in SST, LST and FYT at Hisar during 2012-13, 2013-14 and 2014-15, respectively. RBD (Randomized Block Design) with three replications at spacing of 30 cm x 10 cm in four rows of 3m each and the agronomic management i.e. NPK (kg/ha) 40:40:20 with four irrigations was used in SST, LST and FYT.

Multi-location trial: The superior genotypes so identified were further evaluated in multi-location trials at seven different locations having diverse agroclimatic conditions in various states of India i.e. Ambikapur (21°14'19.3"N and 81°42'17.5"E, Chhattisgarh), Delhi (28°37'48.2" and N 77°09'02.9"E, National Capital), Faizabad (26°32'26.3"N and 81°50'06.9"E, Uttar Pradesh), Faridkot (30°40'32.4"N and 74°44'56.7"E, Punjab), Hisar (29°08'56.7"N and 75°41'35.9"E, Haryana), Ludhiana (30°54'06.9"N and 75°48'53.5"E, Punjab) and Ranchi (23°26'36.4"N and 85°18'53.4"E, Jharkhand) as shown in Figure 1 to identify the best performing high yielding varieties of faba bean for the plain zone of the country. Each genotype was planted in RBD with three replications at spacing of 30 cm x 10 cm in six rows of 3 m each and the same agronomic management i.e. NPK (kg/ha) 40:40:20 with four irrigations was used at all the locations.

Statistical analysis was carried out as per standard procedure (Sheoran et al. 1998).

Quality analysis: The seed samples were dried at 80°C for 8 hours and were ground to a fine powder. Crude protein content (%) was obtained by using micro-Kjeldahl method of AOAC (1984) (Hacıseferogulları et al. 2003). The vicine and convicine content were determined spectrophotometrically by using the method developed by Higazi and Read (1974).

Pathological study: All the faba bean genotypes were screened for their relative resistance/tolerance to *Alternaria* blight and root rot diseases under sick plot conditions in field at Research Area of Medicinal and Aromatic Plant Section, Department of Genetics and Plant Breeding, CCS HAU, Hisar during 2016-17 and 2017-18. Genotypes were sown in the second week of November in three replications of 4 m length and randomized block design was followed. Observation on per cent disease severity for *Alternaria* (Behairy et al. 2014) blight and per cent disease incidence for root rot (Habtegebriel and Boydom, 2018) were recorded at flowering stage. Disease incidence and severity were calculated by the following formula:

$$\text{Disease incidence (\%)} = \frac{\text{Number of diseased plant}}{\text{Total number of plant}} \times 100$$

$$\text{Disease severity (\%)} = \frac{\text{Sum of all numerical ratings}}{\text{Total no. of leaves observed} \times \text{Maximum disease rating}} \times 100$$

After recording the disease incidence, the genotypes were grouped under following reaction categories:

Alternaria blight disease			Root rot diseases		
Disease score	Disease severity (%)	Reaction	Disease score	Disease incidence (%)	Rating
1	0-5	HR	1	0-5	R
2	6-10	R	2	6-15	MR
3	11-20	MR	3	16-25	S
4	21-30	MS	4	>26	HS
5	31-50	S			
6	>51	HS			

Insect pest damage study: The plants of all genotypes were examined weekly for presence of insects at various times of the day (Nuessly et al. 2004). Observations of feeding associations of Aphids and Stink Bugs with faba bean leaves, stems, flowers, and pods were recorded. Number of aphids, *Aphis craccivora*, per five plants was also recorded. Stink bugs including

Lygus lineolaris, *L. heskerus* and *Nezara viridula* were counted per five plants. Percent pod borers were recorded by counting the number of damaged pods out of the total pods of five plants. Insects were identified to species where possible through the use of published systematic keys and direct comparisons with museum specimens of CCS HAU Hisar.

Results and Discussion

Germplasm evaluation: Before starting any breeding programme, generally, pre-breeding evaluation of germplasm lines is done to identify the desirable parents for different economic traits so that the enviable improvement could be achieved in shortest possible time. In the present investigation, 100 germplasm lines of faba bean were screened for yield and its contributing traits, biotic and abiotic stress, early maturing, shattering, and lodging against the national check, Vikrant during 2004-05 under AICRN on potential crops in augmented block design at Hisar. As a consequence, top 10 genotypes were identified with respect to above mentioned traits (Table 1).

Consequent upon the screening of 100 germplasm lines of faba bean, ten genotypes including check were identified for further hybridization program owing to their superior traits. The five genotypes HB 180, EC117755, HB430, HB204, and Vikrant were identified for higher seed yield. Similarly, genotype HB180 and HB85 had higher 100 seed weight. Likewise, HB123, EC117755, Vikrant were found to be highly tolerant to insect pests. Another yield contributing trait, number of pods per plant, was also considered and genotypes EC329675, HB204, EC248710, and PRT12 were found to be superior. Shattering remains a problem in faba bean resulting in direct losses in yield and contributes volunteer plants in the next cropping season. Therefore, the germplasm was screened for shattering tolerance and the genotypes HB180, and HB85 were identified. EC248710 was selected due to a unique characteristic of having short medium plant height and larger number of pods per plant.

Hybridization: In the present study, for integrating higher yield and resistance to biotic and abiotic stress, hybridization work was carried out among the identified superior genotypes/parents (HB180, HB123, EC329675, EC47755, HB430, HB204, EC248710, PRT12, HB85, and Vikrant) in all possible cross combinations through hand emasculation and pollination technique during 2004-05. However, success remained confined to 14 F₁ hybrids due to heavy dropping of crossed female flowers. The problem of abscission in buds, flowers and immature pods has been reported up to 89.5-92.5% by Rabie et al. (1991).

Generation advancement and selection of transgressive segregants: For advancement of generations, the plant progenies were evaluated for their growth, yield, and yield attributing characteristics as well as resistance to biotic and abiotic stress. In order to obtain homogenous and stable plant population, weak plants and progenies were discarded and only high yielding superior plant progenies were advanced. As the yield is affected by multiple environmental factors and management practices, therefore, these genotypes were evaluated for three consecutive years for yield and its contributing traits to realize their actual genetic potential under same management conditions. The resulting stabilized plant progenies were bulked and evaluated in Progeny Row Trial (PRT) for yield, its contributing traits and resistance to biotic and abiotic stress during winter 2011-12 (Table 2). The results revealed that the genotype HB11-2 has highest yield with seed yield 63.47 g/plant followed by HB11-15 (61.37 g/plant), HB11-12 (61.03 g/plant), HB11-32 (58.40 g/plant), HB11-3 (55.00 g/plant), HB11-4 (52.50 g/plant), HB11-5 (52.07 g/plant), HB11-18 (50.20 g/plant), HB11-1 (50.17 g/plant), HB11-9 (49.83 g/plant), HB11-13 (48.97 g/plant), HB11-20 (48.93 g/plant), Vikrant (45.55 g/plant), HB11-6 (45.27 g/plant) and HB11-14 (37.93 g/plant). The majority of genotypes were superior in yield performance against the national check Vikrant.

Performance in station trials: All the fourteen genotypes were promoted to station trials and evaluated for yield performance (Table 3) as well as its contributing traits for three years. In the small-scale trial (SST) during winter 2012-13, HB11-2 and HB11-12 produced maximum seed yield (4620 kg/ha) followed by HB11-15 (4520 kg/ha) and HB11-1 (4480 kg/ha). The large-scale trial (LST) was conducted during winter 2013-14 for second year evaluation of all the genotypes. The genotype HB11-12 produced highest seed yield (4639 kg/ha) followed by HB11-15 (4556 kg/ha), HB11-32 (4533 kg/ha), HB11-2 (4517 kg/ha) and HB11-18 (4394 kg/ha). In the next season, winter 2014-15, experiment was planned as final yield trial (FYT) to identify the promising genotype of faba bean against national check Vikrant. The highest yield was obtained from HB11-32 (4727 kg/ha) followed by HB11-12 (4667 kg/ha), HB11-15 (4556 kg/ha), HB11-2 (4400 kg/ha) and HB11-20 (4378 kg/ha). Based on the three years results of station trials (Table 3), only the top performing genotypes viz., HB11-12 (4642 kg/ha), HB11-15 (4544 kg/ha) and HB11-32 (4513 kg/ha) were proposed for further testing of their yield performance at multiple locations having various agro-ecological conditions in coordinated trials.

Performance in coordinated trials: Upon establishing the yield potential at one location, the desired genotype should be tested at multiple locations over the years before releasing for commercial cultivation (Arya, 2018). The top performing genotypes viz., HB11-12 (4642 kg/ha), HB11-15 (4544 kg/ha) and HB11-32 (4513 kg/ha) over three years in station trials were further evaluated for their yield performance at multi-locations in coordinated trials.

The newly developed and the best performing genotypes viz., HB11-12, HB11-15 and HB11-32 along with the national check (Vikrant) were evaluated in coordinated trials at seven different locations viz., Ambikapur, Delhi, Faizabad, Faridkot, Hisar, Ludhiana and Ranchi in initial varietal trial (IVT) during 2015-16, and advanced varietal trial-I (AVT-I) during 2016-17 and advanced varietal trial-II (AVT-II) during 2017-18 (Yadav et al. 2016; 2017; 2018). During this period, observations were recorded on yield and its contributing traits, quality parameters and tolerance to insect pests and diseases.

The results of present investigation (Table 4) revealed that the performance of different genotypes varied over the years and locations due to differences in soil as well as climatic conditions. On the basis of average of three years (2015-16, 2016-17 and 2017-18, Figure 2), average seed yield of HB 11-12 was recorded as 2234 kg/ha against national check Vikrant (1959 kg/ha) with 14.95 per cent yield superiority in plain zone at national level.

Quality analysis: The nutritive value of faba bean crop is due to its higher protein content, however, presence of antinutritional factors, Vicine and Convicine impair the nutritional benefits when included in the daily diet (Gupta, 1987). Vicine and convicine, upon hydrolysis by β -glucosidase is converted to the aglycones divicine (2,6-diamino-4,5-dihydroxypyrimidine) and isouramil (6-amino-2,4,5-trihydroxypyrimidine), respectively. According to McMillan et al. (2001), the individuals having low-activity variant of erythrocytic glucose 6-phosphate dehydrogenase (G6PD) are susceptible to favism, a life-threatening hemolytic crisis, upon consumption of faba bean because of divicine and isouramil. G6PD regenerates the reduced glutathione resulting from reaction of NADPH in red blood cell hexose monophosphate shunt. G6PD deficient individuals are unable to regenerate reduced glutathione and so they are prone to the oxidative stress and ultimately hemolytic anemia (Arese et al. 2012), however, content of the antinutritional factors can be reduced fermentation process resulting in improving the nutritional quality of faba bean (Rizzello et al. 2016;

Polanowska et al. 2020). Keeping in view the above factors, in the present investigation, protein (%), and vicine-convicine (%), contents were analysed for the three top performing genotypes *i.e.* HB11-12, HB11-32, HB11-15 against the check, Vikrant (Table 5). The average protein content over two years in HB11-12, HB11-32, and HB11-15 was recorded 23.54, 23.48, and 24.14, respectively. The mean vicine-convicine content (%) over two years was 0.73 in HB11-12 and followed by HB11-32 (0.74) and HB11-15 (0.76%).

Plant pathology: The pathology study revealed HB-11-12 showed moderately resistant reaction against *Alternaria* and root rot diseases (Table 6), however, it need to be tested for their authenticity with other inoculation techniques and thereafter it can be utilized in resistance breeding programme for at least development of tolerant variety of faba bean (Juroszek, and von Tiedemann, 2011). The genotypes, HB-11-32 and HB-11-15 showed moderately susceptible reaction against *Alternaria* blight and susceptible reaction against root rot diseases. However, the genotype, HB-11-12 showed moderately resistant reaction against *Alternaria* blight and root rot diseases and may be utilized for commercial cultivation. Among the tested genotypes, HB11-32 and HB11-12 showed minimum infestation of pods, moreover, the infestation and damage by insect pests was below economic threshold level. This indicates that insect pests *Aphis craccivora*, *Lygus lineolaris*, *L. heskerus* and *Nezara viridula* have lower preference towards these genotypes.

The new genotype, HB11-12 of faba bean has been developed as result of transgressive segregation of cross, EC117755 x HB180. It was evaluated over

the years at multiple locations and found promising in seed yield other attributing traits. It is also rich in protein content and comparable antinutritional, vicine-convicine content. HB11-12 was also found moderately resistant to *Alternaria* blight and root rot disease, and insect infestation was also below the economic threshold level. Therefore, it is concluded that HB11-12 may be recommended for commercial cultivation in plain zones of semi arid regions of India.

Insect pest damage study: Minimum population of Aphids, *Aphis craccivora*, appeared on the genotypes HB 11-32 (4.50 nymphs/5 plants) and it was statistically at par with HB 11-12 (4.75 nymphs/5 plants). The population of stink bugs was also recorded on the crop in the month of March 2017. Stink bugs includes *Lygus lineolaris*, *L. heskerus*, and *Nezara viridula*. Minimum population these bugs was recorded on HB-11-32, and HB 11-12. Percent pod borers were recorded by counting the number of damaged pods out of the total pods of five plants (Table 7). Minimum infestation of pods was recorded in the genotype, HB 11-32 (4.50%) and it was statistically at par with HB-11-12 (4.75%).

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Table 1. Performance of top 10 genotypes with respect to different traits.

No.	Parent	Distinct Characters Identified
1.	HB180	Higher seed yield, green pod yield, 100 seed weight, protein. Moderately tolerant to shattering
2.	HB123	Creamy white flower, light green seeds, highly tolerant to insect pests
3.	EC329675	Tall plant with more number of pods per plant
4.	EC117755	Higher seed yield, highly tolerant to insect pests and diseases
5.	HB430	Higher seed yield
6.	HB204	Higher seed yield and larger number of pods per plant
7.	EC248710	Short-medium plant height, larger number of pods per plant
8.	PRT12	Medium plant height, larger number of pods per plant
9.	HB85	Tolerant to shattering and salinity, higher in 100 seed weight
10.	Vikrant	Good yield, Tolerant to insect pests, diseases, lodging

Table 2. Performance of transgressive segregants of faba bean obtained from F₁ hybrids in Progeny Row Trial at Hisar during 2011-12.

No.	New Genotypes	Pedigree Details	Days to Maturity	Plant Height (cm)	Number of Pods/ Plant	Seed Yield/ Plant (g)
1	HB11-1	HB180 × HB 123	166.67	105.53	51.00	50.17
2	HB11-2	Vikrant × HB 123	158.00	98.57	66.33	63.47
3	HB11-3	EC329675 × Vikrant	155.33	96.60	65.67	55.00
4	HB11-4	EC117755 × Vikrant	170.00	115.97	64.67	52.50
5	HB11-5	EC117755 × HB123	153.33	94.17	54.67	52.07
6	HB11-6	EC117755 × HB430	151.00	91.40	48.67	45.27
7	HB11-9	HB204 × HB123	167.67	94.37	53.00	49.83
8	HB11-12	EC117755 × HB180	155.00	93.00	57.00	61.03
9	HB11-13	EC117755 × HB 204	169.00	114.33	52.00	48.97
10	HB11-14	HB430 × HB123	154.00	93.67	41.33	37.93
11	HB11-15	EC329675 × HB 180	170.00	114.00	58.67	61.37
12	HB11-18	EC248710 × Vikrant	169.67	103.50	53.00	50.20
13	HB11-20	EC117755 × PRT-12	168.33	112.60	53.67	48.93
14	HB11-32	HB85 × Vikrant	167.00	113.87	64.67	58.40
15	Vikrant	National check	165.00	106.90	37.67	45.55
	Mean		162.67	103.23	54.80	52.05
	CD (p = 0.05)		6.82	7.06	0.35	7.99
	CV (%)		2.56	4.17	3.91	10.60

Table 3. Seed yield performance of newly developed elite genotypes in station trials.

No.	New Genotypes	Seed Yield Performance (kg/ha)			
		SST, 2012-13	LST, 2013-14	FYT, 2014-15	Over All Mean
1	HB11-1	4480	4222	3461	4054
2	HB11-2	4620	4517	4406	4512
3	HB11-3	4380	4111	3611	4034
4	HB11-4	4280	4028	3611	3973
5	HB11-5	4210	3944	3628	3927
6	HB11-6	4030	2978	2889	3299
7	HB11-9	4340	4278	4217	4278
8	HB11-12	4620	4639	4667	4642
9	HB11-13	4380	4333	4239	4317
10	HB11-14	4100	3183	3250	3511
11	HB11-15	4520	4556	4556	4544
12	HB11-18	4380	4394	4306	4360
13	HB11-20	4200	4250	4378	4276
14	HB11-32	4280	4533	4727	4513
15	Vikrant (c)	3600	3556	3333	3496
	Mean	4295	4001	3952	4116
	CD (p = 0.05)	36.0	24.6	34.5	-
	CV (%)	55.0	35.8	52.5	--

Table 4. Seed yield performance of newly developed elite genotypes in coordinated trials.

No.	Genotypes	Seed Yield Performance (kg/ha)							
		Ambikapur	Delhi	Faizabad	Faridkot	Hisar	Ludhiana	Ranchi	Mean
IVT Winter 2015-16									
1.	HB11-12	1047	1375	2750	3368	4566	1733	1294	2305
2.	HB11-15	826	1265	2667	3542	4063	1733	1394	2213
3.	HB11-32	685	1253	2648	3316	4611	1833	1264	2229
4.	HB-11-38	647	970	2858	3385	3993	1967	1145	2138
5.	Vikrant (c)	1032	1581	2828	3507	3434	1800	1143	2189
	Mean	765	1138	2849	3432	3650	1720	1242	2012
	CD (p = 0.05)	218	436	598	355	247	206	119	-
	CV (%)	193	2184	1196	676	257	693	650	-
AVT-I Winter 2016-17									
1	HB11-12	1792	1384	2777	2785	4573	2083	1935	2476
2	HB11-15	1729	1523	2975	2423	3779	2222	2120	2396
3	HB11-32	1682	1464	2650	3183	4708	2118	1663	2498
4	Vikrant (c)	1728	1247	2858	3002	3590	1840	1390	2136
	Mean	1707	1326	2672	2826	4065	1987	1900	2240
	CD (p = 0.05)	120	225	472	695	181	253	94	-
	CV (%)	509	226	109	144	302	728	336	-
AVT-II Winter 2017-18									
1	HB11-12	871	762	2889	-	3864	1458	1678	1920
2	HB11-15	868	1358	2727	-	4089	1493	2140	2113
3	HB11-32	627	837	2481	-	3874	1354	1943	1853
4	Vikrant (c)	655	651	2130	-	2983	1510	1390	1553
	Mean	735	881	2500	-	3709	1516	1982	1889
	CD (p = 0.05)	178	267	415	-	309	169	120	-
	CV (%)	139	177	974	-	489	657	419	-

Table 5. Faba bean protein and vicine convicine content (%).

No.	Genotypes	Protein Content (%)			Vicine-Convicine (%)		
		2015-16	2016-17	Mean	2015-16	2016-17	Mean
1.	HB 11-12	23.70	23.37	23.54	0.74	0.72	0.73
2.	HB 11-32	23.83	23.13	23.48	0.72	0.75	0.74
3.	HB 11-15	23.80	24.47	24.14	0.75	0.76	0.76
4.	Vikrant	25.10	24.80	24.95	0.79	0.80	0.80
	Mean	24.11	24.53	24.32	0.75	0.77	0.76

Table 6. Screening of faba bean against *Alternaria* leaf blight and root rot disease.

No.	Genotypes	Alternaria Blight (% Severity)			Disease Reaction	Root Rot Incidence (%)			Disease Reaction
		2016-17	2017-18	Mean		2016-17	2017-18	Mean	
1	HB-11-12	19.25	16.0	17.63	MR	13.25	11.2	12.23	MR
2	HB-11-32	24.75	26.5	25.63	MS	20.00	22.4	21.20	S
3	HB-11-15	18.25	21.2	19.73	MS	24.50	26.6	25.55	S
4	Vikrant	16.50	22.2	19.35	MS	24.25	14.6	19.43	MR

Table 7. Entomological data of promising genotypes in the advanced varietal trial 2016-17.

No.	Genotypes	Aphids/5plants	Stink Bugs / 5plants	% Pod Damage
1	HB11-12	4.75 (2.40)	4.75 (2.40)	4.25
2	HB11-32	4.50 (2.37)	4.50 (2.37)	4.00
3	HB11-15	7.75 (2.98)	7.75 (2.98)	8.75
4	Vikrant	11.00 (3.47)	9.50 (3.24)	9.25
	SE(m)	0.454	0.428	
	C.D. (p = 0.05)	1.343	1.267	

Note: Figures in parentheses are $\sqrt{n+1}$ value.

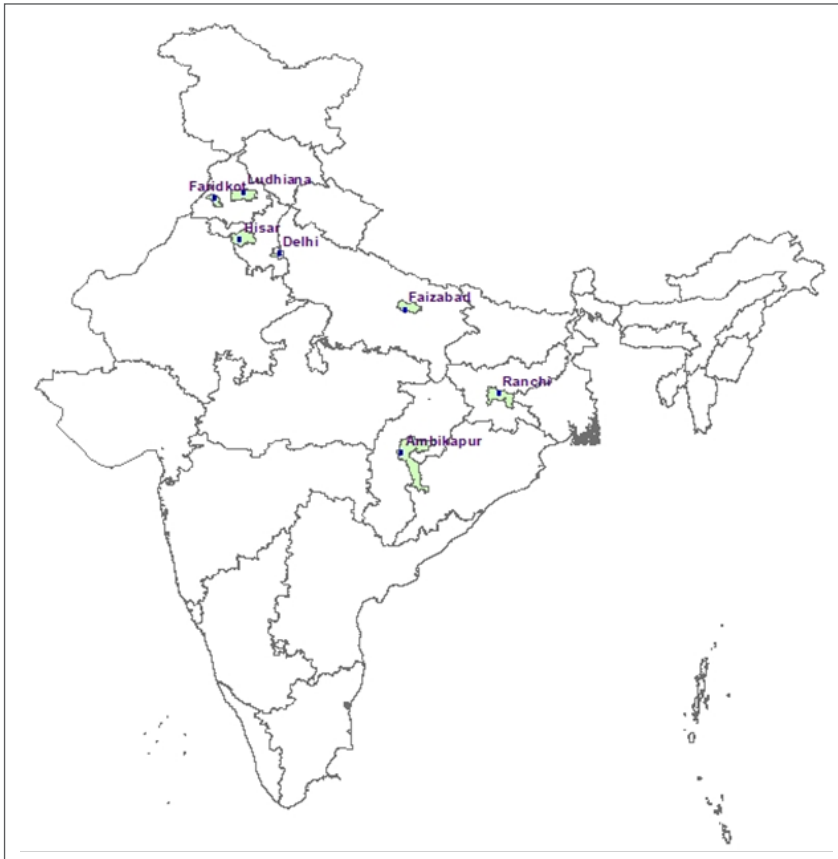


Figure 1. Map showing experimental locations of faba bean trials in plain zones having different agro-climatic conditions of India.

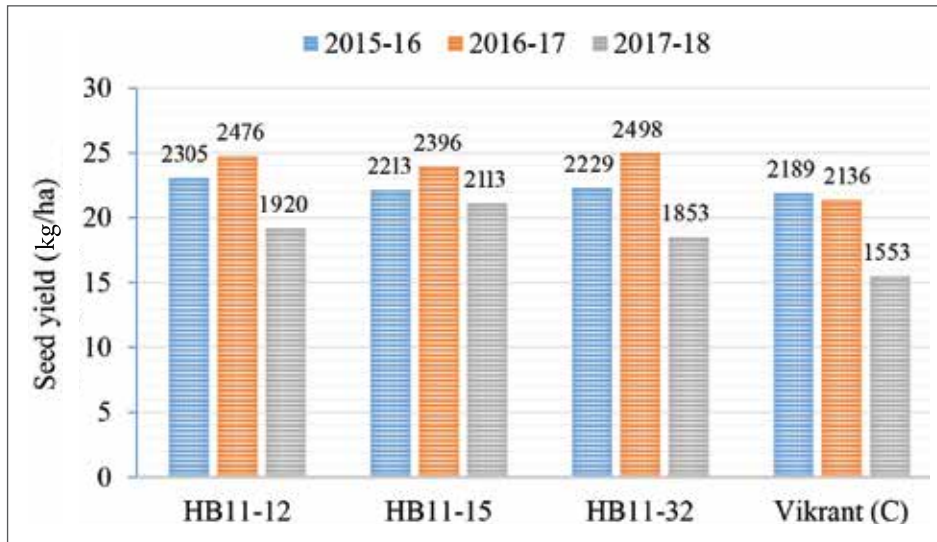


Figure 2. Seed yield performance of faba bean promising genotypes during 2015-16, 2016-17, 2017-18.

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Crossability Studies Among Various Species of Genus *Vigna*

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ABSTRACT

A total of 30 inter-specific crosses of *Vigna mungo* x *V. umbellata* (urdbean x ricebean) and *V. mungo* x *V. angularis* (urdbean x adzukibean) were attempted to study the crossability of urdbean with ricebean and adzukibean. Out of 30 cross combinations, 12 cross combinations of *V. mungo* x *V. umbellata* were successful. Day-to-day visual observations showed that most of the emasculated buds dropped the very next day after pollination and some aborted after about a week. Some of the crosses developed into pods without seeds and dropped. Pod formation involving *V. angularis* as one of the parents in inter-specific hybridization was not very successful as there was no pod formation and buds dropped after 1-3 days of pollination. Among the crosses of *V. mungo* x *V. umbellata*, the crossability of Palampur-93 x PRR-2 was highest (14.18%) followed by Him Mash-1 x VRB-3 (13.64%) and PDU-1 x PRR-2 (13.05%). Interspecific hybridization was performed with the objective to transfer disease resistance in urdbean. The study indicated that different kinds of pre and post fertilization barriers are responsible for complete sterility to low fertility. The genotypes showing a substantially high percent of crossability may be utilized for genetic improvement of urdbean.

Keywords: Inter-specific hybridization, crosses, urdbean ricebean and adzukibean

Introduction

Urdbean [*Vigna mungo* (L.) Hepper], $2n=2x=22$ popularly known as blackgram or mash, is the fourth most important food legume of India, belongs to family Leguminosae and subfamily Papilionaceae, with its wild progenitor, *V. mungo* var. *silverstris* (Bhareti et al. 2011). It has been reported to be originated in India with a secondary center of origin in central Asia (Pratap and Kumar 2011). It is a short-duration pulse crop and self-pollinated grain legume grown in many parts of India.

Food legumes are a vital source of protein, especially for the poor who often cannot afford animal products. Urdbean occupies an important position due to its high seed protein (25-26%), carbohydrates (60%), fat (1.5%), minerals (high amount of iron and phosphorus), amino acids and vitamins and ability to restore the soil fertility through symbiotic nitrogen fixation (Malik, 1994; Harmankaya et al. 2016). Despite

the huge benefits of urdbean, it is grown in 2.5 million hectares of area in India and produces about 1.5 million tonnes of urdbean annually with an average productivity of 400 kg per hectare (Anonymous, 2019). India is the largest producer as well as consumer of urdbean with major growing states, are Maharashtra, Andhra Pradesh, Madhya Pradesh, Uttar Pradesh, Rajasthan, Karnataka and Himachal Pradesh. In Himachal Pradesh, its cultivation is mainly confined to low and mid hills, and is popularly grown as an intercrop with maize as well as a monocrop. However, its yield is low compared to other grain legumes. The low productivity in this crop is attributable to its narrow genetic base due to common ancestry of various superior genotypes, poor plant type, vulnerability to abiotic and biotic stresses and their cultivation in marginal and harsh environments (Ali et al. 2006; Sert and Ceyhan, 2012). It is susceptible to various leaf spotting pathogens such as *Cercospora canescens*, *C. cruenta*, *Colletotrichum truncatum* and *Erysiphe polygoni* in high rainfall areas in the mid

hills of North-Western Himalayas resulting a 40-60% reduction in grain yield.

Extensive screening of the germplasm collections of this species has not yielded any source of resistance to these pathogens. Induced mutagenesis for the induction of resistance using *in vivo* and *in vitro* techniques has also not been successful. Thus under present circumstances, there is no other alternative, but to look for alien *Vigna* species that can provide effective sources of resistance for introgression and other desirable traits to *V. mungo*. Inter-specific hybridization forms the major part of crop improvement. But in many cases, it may be desirable (for useful traits), or even necessary (in cases where there is minimal variability), to cross individuals belonging to two different species (inter-specific hybridization) or genera (inter-generic hybridization). The related species i.e. *V. angularis* (adzuki bean) and *V. umbellata* (ricebean) have been found to be nutritive having high content of resistant starch, vitamins, amino acids, fibers, desirable fatty acids i.e. linoleic and linolenic acid and offers more protein and resistant to most of the fungal pathogen of urdbean. Therefore, the present study has been undertaken to study the crossability of *V. mungo* with various *Vigna* species.

Materials and Methods

For the present investigation, a total of 11 different varieties / genotypes i.e. 5 each of urdbean (Him Mash-1, HPBU-111, Palampur-93, UG-218 and PDU-1) were taken as female and 3 each of ricebean (PRR-1, PRR-2 and VRB-3) and adzuki bean (HPU-51, IC-341983 and IC-341948) taken as male were used to study the crossability relationship (Table 1). The Experimental Farm is situated at 32°8' N latitude and 76°3' E longitude and at 1290 m above mean sea level, representing mid-hill zone of Himachal Pradesh (Zone II) characterized by humid sub-temperate climate with high rainfall (2500 mm) having acidic soil with pH ranging between 5.0 to 5.6. During summer and *Kharif* 2017 and 2018, staggered sowing was done at interval of 10 days starting from 15th February to 31st July to have synchronized flowering in the glasshouse of Department of Genetics and Plant Breeding. The crossing was performed from 15th May to 15th October. Emasculation of female parent(s) at plump bud stage was done in the evening (3:00 to 6:00 P.M.) followed by pollination in the next day morning (7:00 to 9:00 A.M.). Three immuno-suppressants i.e. gibberellic acid (GA₃), indole acetic acid (IAA) and amino caproic acid were used at two concentrations (500 ppm and 1000 ppm) about half an hour after pollination to prevent premature flower abscission

(Fig.1). This was repeated for three consecutive days after pollination at an interval of 24 hours. A total of 30 interspecific crosses i.e. 15 each of *Vigna mungo* x *V. umbellata* (urdbean x ricebean) and *V. mungo* x *V. angularis* (urdbean x adzuki bean) were attempted. Observations were recorded on number of buds pollinated and number of pods harvested to calculate the crossability percentage.

Crossability percentage was calculated as follows:

$$\text{Crossability percentage (\%)} = \frac{\text{Number of crossed pod set}}{\text{Total number of urdbean buds pollinated}} \times 100$$

Statistical Analysis

Since the data were in percent and lying beyond the range of 0 to 30% or 30 to 70% or 70 to 100%, hence it was subjected to arc sine transformation (Gomez and Gomez 1984). The analysis of variance was based on transformed data and original mean values were used to compare the results. In Microsoft Excel, arc sine transformation of percentage data was done by using the following formula:

$$=\text{DEGREES}[\text{ASIN}\{\text{SQRT}(\text{cell}/100)\}]$$

Simple t-test

To test whether the mean difference of crossability, simple t-test was performed as:

$$\text{Student's t-test} = \frac{\bar{X}_d}{\text{SE}(X_d)} \text{ (at } n-1 \text{ df)}$$

where

\bar{X}_d = mean-difference between two sets of related samples

$\text{SE}(X_d)$ = Standard error of mean difference

n = Number of related samples

Results and Discussion

An interspecific hybridization is a promising tool to transfer the desirable traits and to widen the gene pool of any crop. However, wide crosses are not always successful because of the existence of pre and post fertilization barriers that are operative at various stages of development and also various incompatibility barriers limit the potential for recombining the important characters for improving production and adaptation. The present investigation was carried out with the objective to study the crossability relationship of urdbean with ricebean and adzuki bean.

A better understanding of the crossability relationship among the species had been helpful in opting methods for making successful crosses (Bhanu et al. 2018). Day-to-day visual observation showed that most of the emasculated buds dropped the very next day after pollination and some aborted after about a week. Pod formation involving *V. angularis* as one of the parents in inter-specific hybridization was not very successful. There was no pod formation and buds dropped after 1-3 days of pollination. The absence of seed set and abscission of crossed flowers within 72 hours from pollination demonstrate that the first barrier responsible for complete sterility is the delay in pollen tube entry into the ovules because of the difference in length of the style of species involved.

Varying degrees of success in interspecific hybridization was also reported by various workers *viz.* Ahn and Hartman (1977), Chen et al. (1983), Mittal et al. (2005, 2008, 2010) and Bhanu et al. (2018) owing to reproductive obstructions between the species involved in interspecific hybridization. The range of crossability percentage was observed to be 0-14.19% (Table 2). The analysis of results revealed that cross combinations Palampur-93 x PRR-2, Him Mash-1 x VRB-3, PDU-1 x PRR-2, Palampur-93 x PRR-1, Him Mash-1 x PRR-1 and Him Mash-1 x PRR-2 were found to be significantly superior over other cross combinations in *V. mungo* x *V. umbellata* hybridization in terms of crossability. Among the crosses of *V. mungo* x *V. umbellata*, the crossability of Palampur-93 x PRR-2 was highest (14.19%) followed by Him Mash-1 x VRB-3 (13.66%) and PDU-1 X PRR-2 (13.06%) (Fig.2). Crosses having a high crossability percentage were considered as successful crosses suggesting the parents of these cross combinations are ideal for the transfer of useful genes from one species to another species. Similar crossability success was also reported by Bhanu et al. (2018) 16.27% in *V. mungo* x *V. umbellata* and 37.50% in *V. mungo* x *V. radiata*; Lekhi et al. (2017) in *V. mungo* x *V. radiata* with 24.10%; Bharathi et al. (2006) in *V. radiata* x *V. umbellata* with 29.63 percent, *V. radiata* x *V. trilobata* with 8.48%, *V. radiata* x *V. aconitifolia* with 7.69%. The percent crossability among different cross combinations varied from species to species may be due to wide variation in the genetic architecture of the species involved in interspecific hybridization. Mittal et al. (2005) and Dhiman et al. (2013) also observed differential responses of the genotypes of urdbean and ricebean involved in interspecific hybridization. In the present study, cross combinations HPBU-111 x PRR-1 (5.37%), HPBU-111 x PRR-2 (4.96%), UG-218 x PRR-1 (2.65%), UG-218 x PRR-2 (4.70%) exhibited low crossability

rates and there was no pod set in cross combination HPBU-111 x VRB-3, UG-218 x VRB-3 in urdbean x ricebean hybridization. Present results are in agree with the findings of Thiyagu et al. (2008) who observed a low percentage of pod set (5.56%) in *V. mungo* x *V. umbellata* indicating the presence of reproductive barriers that renders the introgression difficult. They also found normal pollen grain germination on the stigmatic surface but slow pollen tube growth in addition to structural abnormalities in stigmatic and stylar regions. In some crosses, there is abscission of young fruits between 3 to 30 days after pollination, which might be due to failure of endosperm nuclei to divide or the delayed endosperm nuclear divisions leading to embryo abortion (Bhanu et al. 2018). So, crossability between the species is a prerequisite for gene transfer through interspecific hybridization. Some of the pods which were formed were without seed or had shrivelled seeds with a ruptured seed coat. Crosses, where HPBU-111 is used as one of the parents, had more number of empty pods. F₁ seeds developed were of two types (i) highly shriveled, minute, brown colored (ii) bold, comparatively brown colored but very weak as compared to selfed ones (Fig.3). The number of seeds per pod in the inter-specific hybrids varied from 1-4. Sehwat et al. (2016) also reported that the number of F₁ seeds per pod in interspecific crosses between urdbean and ricebean varied from 1 to 4. The F₁ seeds obtained from all cross combinations were small, wrinkled and shrunken. The F₁ seeds were small in size and shriveled because of the poor development of the endosperm and embryo which might be due to incompatibility between the two parental genomes or due to the failure of the embryo to reach maturity (Rashid et al. 1987).

Conclusions

The present study reveals the operation of pre fertilization barriers such as slow pollen tube development, no germination of pollen grains, delay in pollen tube entry into ovules and high abscission rate of crossed flowers within four days after pollination. Even though the fertilization barriers were predominant, some inter-specific hybrids were produced. The parents involved in interspecific hybridization showed differential genotypic response, which indicates the use of more genotypes and a large number of crosses should be attempted to get more F₁ plants. The crosses showing a substantially high percent of crossability can be utilized for genetic improvement of urdbean.

Table 1. Parentage/source of genotypes used in interspecific hybridization.

Species	Variety(s)	Source/Parentage
<i>Vigna mungo</i>	Palampur-93	Pureline selection from the local material of Himachal Pradesh collected by CSK HPKV, Palampur
	Him Mash-1	DPU 91-5 x Mash 338
	HPBU-111	Pureline selection from the local material of Himachal Pradesh collected by CSK HPKV, Palampur
	UG-218	IIPR Kanpur
	PDU-I	Selection from IC-8219
<i>V. umbellata</i>	PRR-1	Pureline selection from Jagdhar (Tehri) collection by GB Pant University by Pantnagar
	PRR-2	UUHF, Ranichauri
	VRB-3	Selection from heterogenous sample of accession IC538080
<i>V. angularis</i>	HPU-51	Pureline selection from the local material of Himachal Pradesh collected by CSK HPKV, Palampur
	IC-341983	Indigenous collection
	IC-341984	Indigenous collection

Table 2. Pod set and cross ability percentage in *V. mungo* and *V. umbellata* crosses.

No.	Cross Combination	Buds Emasculated and Pollinated	Crossed Pods Formed	F ₁ Seeds Obtained	Cross Ability Percentage
1	Palampur-93 x PRR-1	369	44	173	11.92**
2	Palampur-93 x PRR-2	444	63	248	14.19**
3	Palampur-93 x VRB-3	343	27	103	7.87
4	Him Mash-1 x PRR-1	400	49	193	12.25**
5	Him Mash-1 x PRR-2	391	47	183	12.02**
6	Him Mash-1 x VRB-3	432	59	235	13.66**
7	HPBU-111 x PRR-1	409	22	83	5.37
8	HPBU-111 x PRR-2	403	20	78	4.96
9	HPBU-111 x VRB-3	366	0	0	0.00
10	UG-218 x PRR-1	377	10	40	2.65
11	UG-218 x PRR-2	382	17	64	4.70
12	UG-218 x VRB-3	351	0	0	0.00
13	PDU-1 x PRR-1	356	0	0	0.00
14	PDU-1 x PRR-2	421	55	216	13.06**
15	PDU-1 x VRB-3	339	26	98	7.67
Mean ± SE					13.77 ± 2.09

** Significant at 1% level of significance



Figure 1. Interspecific hybridization. (Original)



Figure 2. Interspecific pods. (Original)



Figure 3. Interspecific seeds. (Original)

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Expansion of the Genetic Base by Interspecific Hybridization in *Capsicum annuum* and *Capsicum chinense*

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ABSTRACT

In Turkey, there is huge diversity in terms of pepper genotypes and varieties. However, in recent years, using standard and hybrid varieties in production caused the reduction of genetic diversity over time. While creating breeding programs for different purposes, the existing gene pool is needed to be well known and it should be enriched according to the breeding targets. In recent years, interspecific hybridization is intensively carried out to increase the existing genetic variation and to extend of genetic bases of varieties that have biotic and abiotic stresses tolerance in pepper breeding programs. The objective of the study was to broaden the existing genetic base by crossing *C. annuum* (253A and İnan3363) and *C. chinense* (PI 159236) varieties. The study involves the evaluation of 54 morphological characters. The first three eigen values showed 56% total variance in the F₂ population obtained from 253A x PI 159236 crossing (110 offspring), whereas in the F₂ population obtained from İnan3363 x PI 159236 crossing (150 offspring), it was 87%.

Keywords: Pepper, interspecific hybridization, genetic base, morphological characterization

Introduction

Pepper is grown in all regions of Turkey and is one of the vegetable species having high commercial potential. Turkey has been ranked third after China and Mexico with 7% of total world pepper (*Capsicum annuum*) production (FAOSTAT, 2021). Although Turkey is not the origin of pepper, it contains a great variety in terms of genetic resources (Bozokalfa et al. 2009). However, the use of standard and especially hybrid varieties in production in recent years leads to a decrease in the genetic diversity existing.

The use of wild species in breeding allows more widely and effectively use their valuable properties to improve cultivars by many characteristics, such as resistance to biotic and abiotic factors, cytoplasmic male sterility, and restoration of fertility (Shmykova et al. 2014). Plant breeding focuses on increasing crop yield to meet the needs of the developing world population, improving food quality to ensure a healthy

life, developing new biofuels and addressing global warming and environmental pollution issues. As creating breeding programs for different purposes, the available gene pool should be well known and enriched with the goals aiming. It is very important to design breeding programs by creating a large gene pool where wild and cultural forms are evaluated together.

With breeding studies, intraspecific or interspecific crossings are carried out between parents having desired characters to create variations in the population (Sunil and Rasheed, 1998). The use of wild species in breeding makes it possible to more widely and effectively use their valuable properties to improve cultivars by many characteristics, such as resistance to biotic and abiotic factors, cytoplasmic male sterility, and restoration of fertility (Shmykova et al. 2014).

In this study, the pepper genetic base has been expanded through interspecific crossings among a genotype (PI 159236) belonging *Capsicum chinense*

and a genotype (253A) and a variety (İnan3363) belonging *Capsicum annuum* obtained from Alata Horticultural Research Institute (Mersin, Turkey).

Materials and Methods

253A (a Charleston type inbred line with high heterosis strength) and İnan3363 (bell pepper type) belonging to the *Capsicum annuum* species and genotype PI 159236 (obtained from AVRDC-Taiwan and *Capsicum chinense* species) was used as parents. Seeds of pepper used as a parent were sown into trays containing peat: perlite (2 w/1 w). Seedlings having 6-7 leaves were planted in a plastic greenhouse at intervals of 40 x 100 cm as 5 plants from each genotype and necessary agricultural practices such as irrigation and fertilization were carried out. During the full anthesis phase, when stigma is active, crossings were performed between the parents reciprocally. To prevent embryo abortion due to interspecies crossing, fruits were harvested 28-40 days later from pollination and the embryo recovery method was applied *in vitro* conditions (Hossain et al. 2003; Yoon et al. 2006). To ensure embryo development; MS (Murashige and Skoog, 1962) nutrient medium including 0.5 mg L⁻¹ Gibberellic acid (GA₃), 0.05 mg L⁻¹ Naphthaleneacetic acid (NAA), 15 mg L⁻¹ silver nitrate (AgNO₃) and 0.25% activated charcoal was used (Pinar et al. 2011). The plants developed from embryos were acclimated and, 20 plants from each hybrid combination were planted into the greenhouse at 80 x 100 cm intervals and necessary agricultural practices such as irrigation and fertilization were performed. F₂ populations were obtained by selfing of F₁ plants at the full anthesis stage. In order to evaluate the morphological properties of pepper plants of F₂ populations, 62 criteria requested in the pepper descriptor list prepared on the basis of IPGRI feature documents were examined (IPBGRI, 1995). Trees showing the degree of relationship related to the morphological features of the pepper genotypes obtained were drawn by using the NTSYSpc 2.1 computer package program.

Results and Discussion

Embryo abortion, which is common in crosses between *Capsicum* species, occurs as a result of the effect of barriers existing before and after fertilization (Pickersgill, 1992). Endosperm development is much slower than normal after fertilization, which delays embryo development, and the embryonic sac collapses after about two weeks (Pickersgill, 1997). In the present study, plants were crossed reciprocally during the flowering period and embryo rescue was carried out 28-40 days later after pollination

to prevent embryo abortion. The plants developed from embryos were acclimated and transferred to greenhouse conditions (Figure 1).

The number of embryos rescued from the fruits opened varied in crossing combinations. When the İnan3363 and PI 159236 were used as female and male parents respectively, the highest viability rate of the embryos was recorded with 82%. On the contrary, the lowest rate was observed with 13%. It was the same in the hybrids of 253A and PI 159236. The viability rate was high (66%) and low (34%), when 253A and PI 159236 were used as the female parent (Table 1). In the combinations obtained by crossing two different *C. annuum* with PI 159236, it was observed that the viability rate was higher in the hybrids when *C. annuum* was used as a female parent. Casali (1970) reported that the germination rate of seeds varies between 0-6.6% and 0-47% when *C. chinense* and *C. annuum* are used as the female parent, respectively.

In this study, fruits were obtained from each combination in interspecific crossing and different rates of viability were observed in embryos rescued from seeds in these fruits. Similarly, Saccardo and Ramulu (1977) observed 70-76% fruits and 7-14% fertile seeds in *C. chinense* and *C. annuum* hybrids. Costa et al. (2009) performed crossings between twenty different *C. chinense* genotypes and one *C. annuum* variety and they could obtain fruits from all hybrid combinations. Fruit ratios and seed germination were found between 8.9-40.0% and 0-87.5%, respectively, regarded with combinations. It was reported that fruits and fertile seeds are observed in the *C. chinense* and *C. annuum* hybrids. Similar results were obtained in this study presented and less number of fruits and seeds were recorded from crossing between *C. chinense* and *C. annuum* compared to reciprocal hybrids.

Among the obstacles after fertilization in interspecific hybrids, hybrid weakness or necrosis can be mentioned. This anomaly is defined by a series of phenotypic features that are similar to those associated with a response to environmental stress, such as the attack of pathogens and viral diseases (Bomblies and Weigel, 2007). Martins et al. (2015) observed this type of expression in the hybrids of the *C. frutescens* × *C. baccatum* combination in the seedling and later stages and reported that some undeveloped seedlings die at a young period and those that develop stunted and have deformed cotyledons, plants do not produce flowers and therefore fruit cannot be obtained. In this study, a similar situation was observed in the hybrids of *C. chinense* × *C. annuum* combinations such as death at the seedling stage and deformed leaves, flowers and fruits with stunted development (Figure 2).

Although these plants were left open-pollinated, no seeds were formed in any fruit.

When F_1 plants obtained from $253A \times C. chinense$ and $Inan3363 \times C. chinense$ crossings were at full anthesis stage, F_2 populations were created by selfing. The plants were isolated with nets for obtaining F_2 populations and allowed to be pollinated with their pollens. In order to create back-cross populations, manual pollination was performed.

In the study, 110 plants from $253A \times PI 159236 F_2$ population and 150 plants from $Inan3363 \times PI 159236 F_2$ population represented the transgressive segregation from each F_2 population. Accordingly, the measurements and observations were carried out according to 54 morphological criteria and cluster analysis of the data obtained and trees showing the degree of relationship related to morphological features were drawn using NTSYS-PC 2.1 computer package program.

As a result of the analysis, when the dendrogram of $253A \times PI 159236 (C. chinense) F_2$ population was examined in Figure 3, it was consisted of 2 main groups. In general, it was determined that both main groups were divided into 4 side groups. In this F_2 population, the total number of plants was 110. The coefficient average of this dendrogram drawn according to the correlation matrix using UPGMA coefficient in SAHN was 0.48 and consisted of four groups according to this average. The value that the similarity index represents the dendrogram was $r = 0.99$. According to the first three eigen values, the cumulative variance was 87.7 (Table 2).

When the dendrogram of $Inan3363 \times PI 159236 (C. chinense) F_2$ population presented in Figure 4 was examined, it was consisted of 150 pepper plants in total. According to the correlation matrix, the cluster mean (similarity average) of this dendrogram drawn using the UPGMA coefficient in SAHN was 0.48 and when the dendrogram was examined, it was consisted of 2 main groups. It was determined that the first main group was divided into 2 side groups and the second main group was divided into 6 side groups. The value that the similarity index represents the dendrogram was $r = 0.89$. According to the first three eigen values, the cumulative variance was 56.46 (Table 3).

In the study, according to the dendrograms obtained from the morphological characterization of F_2 populations, it was seen that while the parents were in different groups, a separate group formed apart from the groups involved both parents. Based on these findings, the populations appeared to be segregated to represent transgressive segregation.

Genetic variations, which have an important role in the success of plant breeding, have been continued

to be reduced by modern plant breeding following cultivation (Tanksley and McCouch, 1997). It is known that interspecific crosses increase genetic diversity and heterosis. In the study carried out for this purpose, genetic variation due to interspecific crosses was calculated. While modern molecular methods are preferred to reveal genetic diversity among genotypes, agro-morphological characterization constitutes the basis and the first step of identification (Smith and Smith, 1989). The genetic diversity of *Capsicum* species can be evaluated as a whole, including agronomic, morphological and molecular features (Costa et al. 2016). Despite the accuracy in predicting the genetic deviation between accessions by molecular markers, phenotype knowledge by morphological and agronomic identifiers is still important. The collection of morphological data is practical and economical compared to the collection of quantitative and molecular data (Sudré et al. 2010).

Cluster analysis is widely used in comparing the plant and fruit characteristics of the varieties, and with the dendrograms created using this data, it is possible to determine groups of the cultivars (Panayotov et al. 2000). In the study, a total of 54 morphological features were evaluated and the genetic variation in the F_2 populations obtained for these features was found to be 58% in the F_2 population obtained from $253A \times PI 159236$ hybrids and 87% in the F_2 population obtained from $Inan3363 \times PI 159236$ hybrids. Keleş (2007) carried out the morphological characterization of 562 pepper genotypes of *C. annuum* and obtained a collection containing 96 genotypes by evaluating the dendrograms drawn as a result of the analyses. Total variance of three eigen value in terms of 53 morphological features in PCA was found to be 30%. When the study results are compared with this study, it is seen that interspecific crossing increases the genetic variation in *C. annuum* existed. Olszewska et al. (2011) analyzed biometrically F_1 hybrids (*C. annuum* \times *C. frutescens*, *C. frutescens* \times *C. annuum*, *C. frutescens* \times *C. chinense* ve *C. chinense* \times *C. frutescens*) obtained as a result of interspecific crossing. The characters evaluated in the hybrids tested were found to produce large variations. They described that the hybrids obtained from crossing as the most valuable material for future pepper breeding projects. They also reported that these materials contained valuable features that will be used as a parent in the genetic recovery of pepper.

Product development depends on the presence of genetic diversity and the degree of genetic variation that existed, and the degree of development also depends on the size of useful genetic variability (Uma Jyothi et al. 2011). Interspecific hybridization

enables transferring of genes between different species and allows them to develop genetically superior genotypes (Bosland and Votava, 1999). The characterization and evaluation of the cultivated *Capsicum* species are especially interesting for gene banks. Because, a wide variation, which is not yet fully known and used, exists in these species (İnce et al. 2009). Despite the accuracy in predicting genetic deviations between accessions by molecular markers, phenotype knowledge obtained from morphological and molecular identifiers is still important. Some studies have concluded that the relationship among morphological, agronomic and molecular data of *Capsicum* accessions is the most appropriate approach to estimate *Capsicum* genetic segregation (Costa et al. 2009; Moura et al. 2010) or that joint analysis of qualitative and quantitative data results in greater efficiency. Sudré et al. (2010) observed that only morphological descriptors can effectively distinguish between *Capsicum* species and botanical varieties.

Conclusions

Increasing the genetic diversity in the *Capsicum* genus provides parameters for identifying parents that produce more heterotic effects and increases the likelihood of obtaining superior genotypes in separated generations. In this study, genetic variation existing in *C. annuum* however decreasing gradually during the breeding process was increased by using populations obtaining through the crossing of *C. annuum* with *C. chinense*. Plants obtained from interspecific hybrids are very valuable materials for future breeding projects. As a result of this study, gene flow to the shrinking pepper genetic basis was provided and the variation was increased. Since the rich material basis needed for different pepper breeding projects (heterosis, biotic/abiotic stress resistance, QTL mapping, variety breeding having different biochemical content) was created, homozygous lines having genetically valuable properties can be produced and used as parents in breeding studies.

Table 1. Number of fruits opened, number of seeds, number of embryos recovered, % viability rate and number of alive plants in crossing combinations.

Crossing Combination	Opened Fruit Number	Seed Number	Recovered Embryo Number	Viability (%)	Alive Plant Number
253A × PI 159236	10	915	607	66	35
İnan3363 × PI 159236	8	1095	895	82	55
PI 159236 × 253A	10	558	195	34	58
PI 159236 × İnan3363	10	714	94	13	42

Table 2. First three eigen values of the correlation matrix in 253A × PI 159236 F₂ population.

	Eigen Value	Variance (%)	Cumulative Variance (%)
1	63.90	56.55	56.55
2	30.62	27.10	83.66
3	4.56	4.04	87.70

Table 3. First three eigen values of the correlation matrix in İnan3363 × PI 159236 F₂ population.

	Eigen Value	Variance (%)	Cumulative Variance (%)
1	29.79	19.60	19.60
2	23.71	15.60	35.20
3	18.46	12.14	56.46

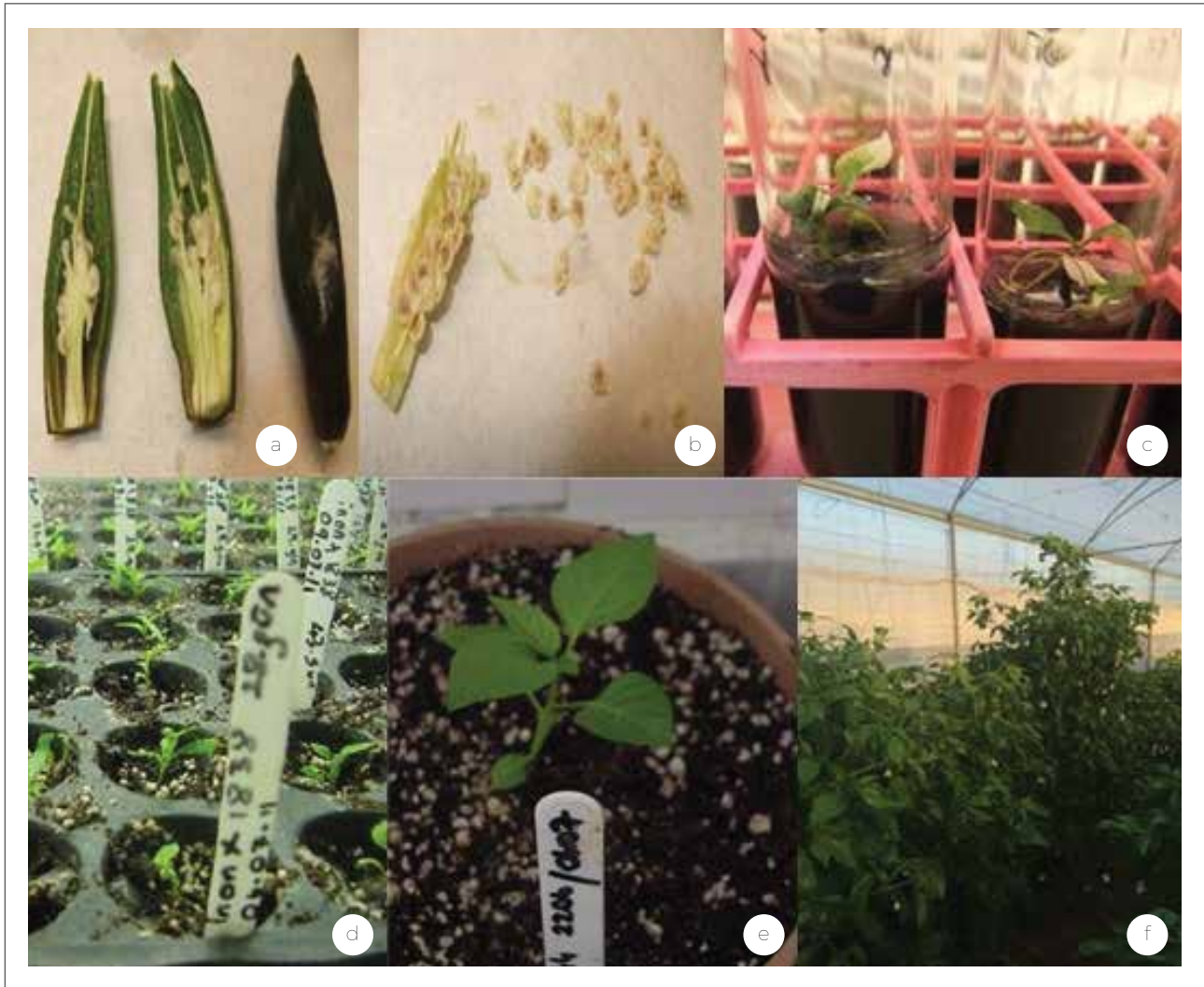


Figure 1. Embryo rescue processes and acclimation of plants (a: the fruits harvested, b: the seeds opened, c: the plants developed from embryos, d: the plants transferred to trays, e: the plants transferred to pots, f: the plants transferred to greenhouse). (Original)

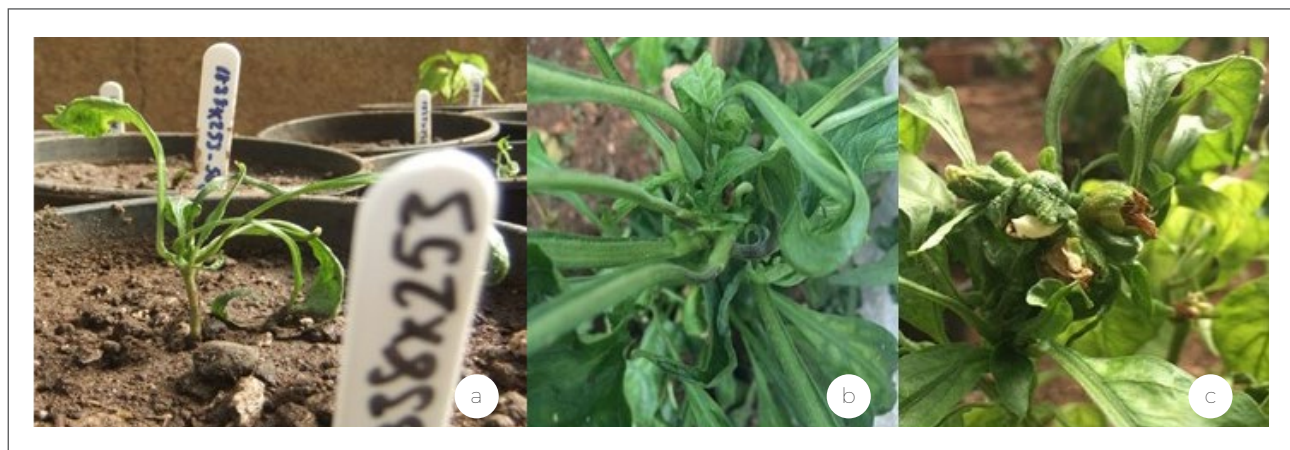


Figure 2. In the *C. chinense* × *C. annuum* hybrid combinations, the plants developed by the effects of hybrid necrosis due to post-zygotic barriers, however deformed (a: 4-5 seedling having leaves, b: the abnormal appearance of hybrid plants, c: deformed flowers). (Original)

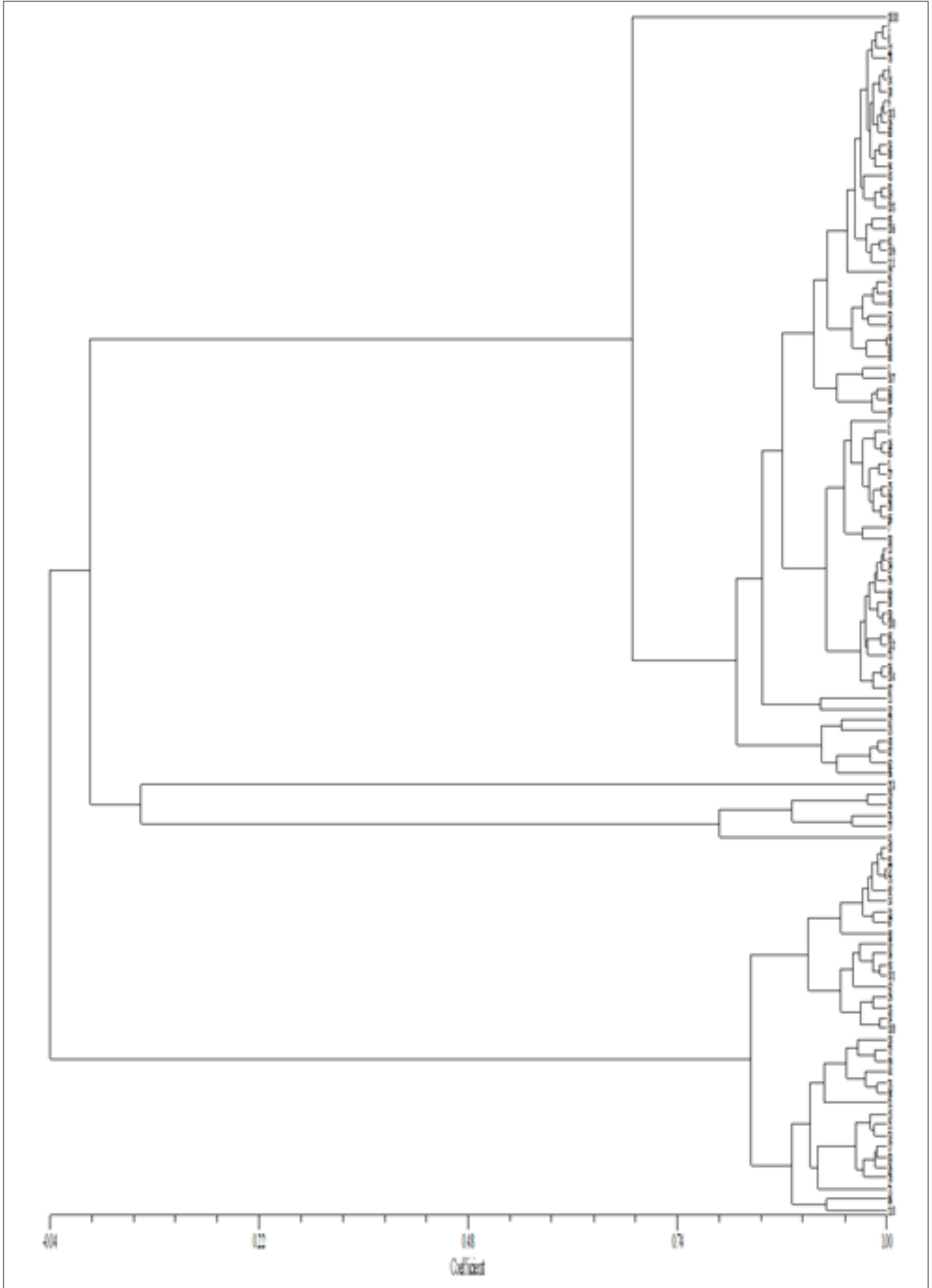


Figure 3. Dendrogram of pepper plants of 252A \times PI 159236 (*C. chinense*) F_2 population obtained according to UPGMA using the correlation matrix.

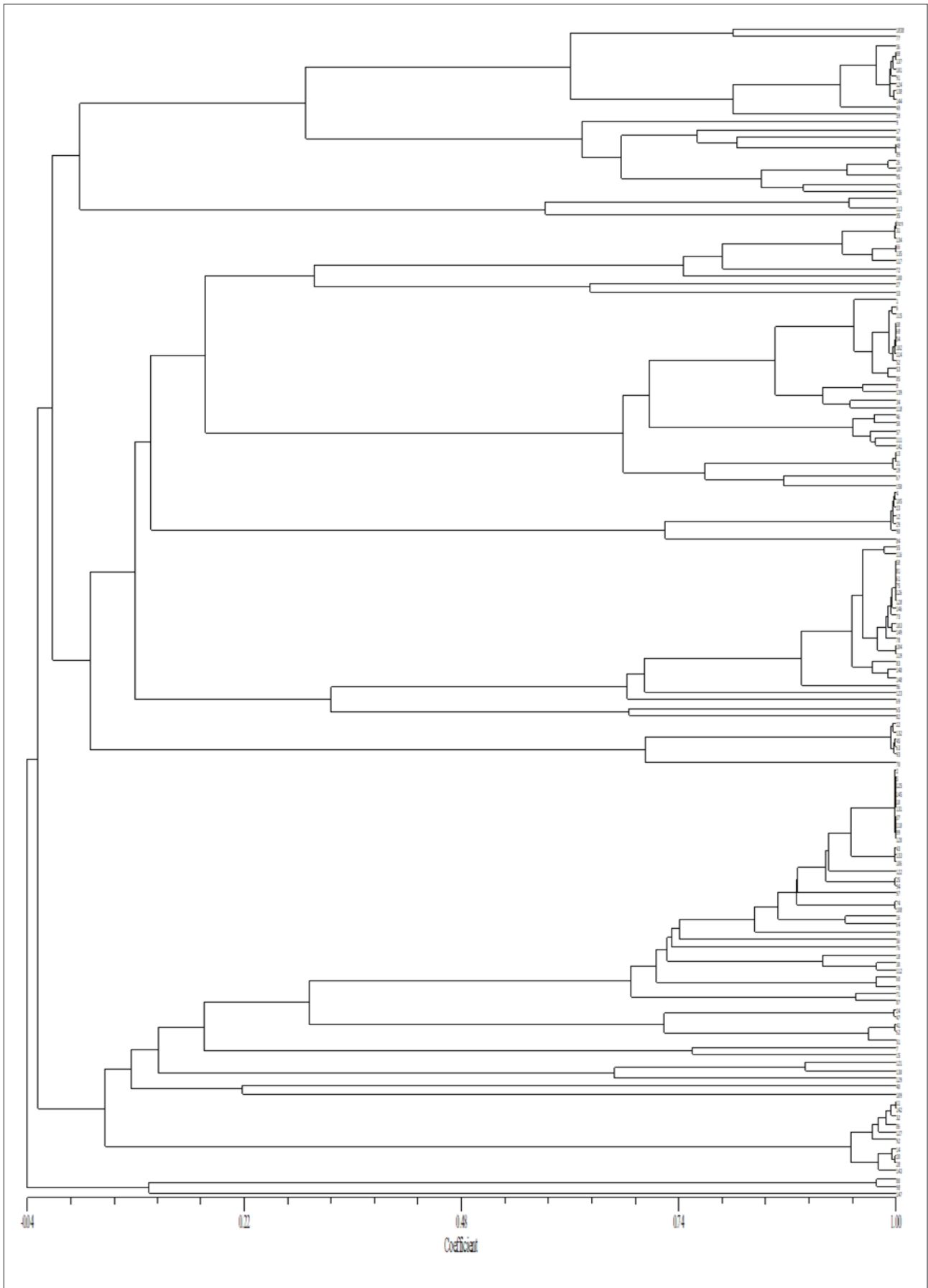


Figure 4. Dendrogram of pepper plants of Inan3363 \times PI 159236 (*C. chinense*) F_2 population obtained according to UPGMA using the correlation matrix.

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Deciphering Genotype×Environment Interaction by AMMI and GGE Biplot Analysis Among Elite Wheat (*Triticum aestivum* L.) Genotypes of Himalayan Region

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ABSTRACT

To determine yield stability and the effects of the interaction between the genotype and the environment, 101 wheat genotypes were assessed over 2 years (2018-2020). The experiments were performed at different diverse locations in Kashmir traversing a significant altitudinal range viz. Khudwani (34.38°N of latitude and 77.0°E of longitude) and Wadura (34.52°N of latitude and 74.52°E of longitude) following recommended agronomical practices. Analysis of the main additive effect and multiplicative interaction (AMMI) of the seed yield variance revealed a significant genotype, environmental and genotype × environment interaction effect at $p < 0.01\%$ probability level. Three main principal components based on AMMI explained most of the variation due to genotype × environment interaction at $p < 0.01\%$ probability level. The GGE biplot indicated that two mega-environments were present. The first section (large environment) contains the KH18 and WA18 test environments with genotypes G2, G38, and G16 with the highest yield (winner), the second section (large environment) contains KH19 and genotypes G28, G19 and G3, KH19 and WA18 WA19 environment included. as a winner. Plots of mean versus stability show that the genotypes exhibiting both high mean yield and stability scores at the test sites are G2, G50, G26, G80, and G1. Hence, the above identified genotypes with superior yield and other desirable attributes can be recommended as generally adapted or niche specific genotypes for broad and specific areas, respectively.

Keywords: Wheat germplasm, biplot, wheat yield stability, principal component, wheat screening

Introduction

Bread-making wheat (*Triticum aestivum*, L.) is a major cereal grown throughout the world as a basic food. In India, wheat is the second largest grain crop and the primary food crop in the north and northwest of the country. The crop is grown on 30.5 million hectares and yields an average of 3.51 tons per hectare (FAO 2021).

The most important wheat-producing states are Punjab, Haryana, Uttar Pradesh, Rajasthan and Madhya Pradesh. Global population in general and Indian population in specific has been increasing at an exponential rate which proportionally increases demand for food supply on daily basis. New benchmarks for food requirement are set as challenges for plant breeders across the globe to

enhance the sustainable food production for nutritional security. It is mandatory to explore interventions of vertical expansion rather than horizontal under scarce availability of land and other resources. Therefore, it is imperative to improve upon the productivity potential of the existing wheat germplasm using tools of crop improvement. The existence of genetic variability is pre-requisite for any successful breeding program (Kant et al. 2011). Therefore, before planning of any breeding program the variability parameters like coefficient of variation (COV), genotypic variation, critical difference, heritability and correlation is important to get efficient results (Abebe et al. 2017). There is significant level of genetic variability among various ecotypes of wheat in the existing regional and global wheat biodiversity and act as an important source of elite ness and disease resistance for breeding novel wheat varieties. It has been ascertained that novelty of eliteness or superior performance either gets masked up or performs poorly across diverse niche environments. This differentiated behavior of crop varieties in ecoregions is due to the interaction of genotype \times environment, as reported for environmental-induced yields and other phenotypic traits (Ajay et al. 2018) and other biotic and abiotic factors. It is mainly due to gene \times environment interaction that complicates the selection process for targeted trait due to change in response under varied environmental conditions affecting selection accuracy. The improvement is also limited due to the complex nature of trait (yield) being regulated by many genes (Sallam et al. 2019). For such conditions, stability analysis facilitates the best possible solution to assess the relative performance of genotypes with respect to specific and broad environmental evaluation (Kant et al. 2014). Large scale testing across multiple sites helps in delineating major mega-environments. Various statistical parameters were developed and used for efficiency in estimating the stability index of genotypes across environments, such as, coefficient of determination (R_i^2), regression coefficient (b_i), coefficient of variability, deviation due to regression (S_{di}^2) linear regression, and pooled analysis of variance across the environments. Additive Main Effect and Multiplicative Interaction (AMMI) model and two-step GGE analysis have been observed and reported to accurately capture the majority of sum-of-squares interactions, isolate major and interacting components, and facilitate visualization of genotypic fitness in various environments (Shashikumara et al. 2020). The AMMI model mainly consists of the additive main effects of genotype and environment and the multiplier effect of the genotype \times environment interaction, so it can obtain more information than other methods. It

can also be viewed as a combination of ANOVA and principal component analysis (Ebdon and Gauch, 2002) and describes the genotype \times environment interaction in more than one dimension (Yan and Kang, 2003). The AMMI stability parameters allow examining yield stability after reduction of the noise from GE interaction effect (Ajay et al. 2020). This model interprets genotype \times environmental interactions in terms of external environmental factors and genotypic variables in common wheat. Here, multi-media testing (MET) data can be used to predict phenotypic responses in an uncontrolled environment using explicit environmental information (Mohammadi et al. 2020). AMMI method has been used in several studies to select stable bread wheat cultivars (Katsenios et al. 2021; Ljubičić et al. 2021; Verma and Singh 2021a, 2021b). The present investigation was undertaken to explore available wheat diversity for genetic variability for yield superiority and other important attributing factors. To evaluate the stability of genotypes in different environments, experimental experiments were performed at multiple sites to identify a wide range of specifically suitable wheat germplasm. Good and stable genotypes were identified by evaluating yield and variation due to genotype, environment, and GE interactions for yield determinants.

Materials and Methods

Experimental wheat trials were conducted during the year 2018-19 and repeated in 2019-20 i.e. two years at two locations constituting four (4) environments. The evaluation of these experimental wheat genotypes was carried out during the Rabi season of these respective year at Mountain Research Centre for Field Crops (MRCFC) Khudwani (Anantnag- South Kashmir) and Research field of Division of Genetics and Plant Breeding, Faculty of Agriculture (FOA), Wadura, Sopore (North Kashmir). The experimental material comprised of 101 genotypes collected from CIMMYT, ICAR-IIWBR Karnal exotic nurseries and four check varieties Shalimar Wheat1, Shalimar Wheat 2, HS-562, VL 907. (Table 1.) Field trials were presented in a randomized block design (RCBD), each repeated twice over two years. Each genotype was represented by a plot size of 1 \times 1 m with six rows. The row-to-row spacing kept at each block is 20 cm. Data was recorded from all the genotypes in each replication for yield and associated morphological traits. Observe to determine characteristics such as plant height (cm), days to maturity, number of tillers per meter, spike length (cm), number of spikelet's per spike, grains per spike, thousand grain weight, and yield per plot.

Statistical analyses

We calculated $G \times E$ interactions using the AMMI method. An integrated analysis of variance was performed and the mean was used as the basis for the AMMI analysis. The basis of the AMMI mathematical formula was as follows:

$$y_{ij} = \mu + g_i + e_j + \sum_{k=1}^N \lambda_k Y_{ik} \alpha_{jk} + \varepsilon_{ij}$$

where y_{ij} is the yield of the i -th genotype in the j -th environment, N is the number of major components of the AMMI model, μ is the total mean value of the genotype, g_i and e_j are the overall mean value of the deviation between the genotype and the environment, λ_k is the k -axis eigenvalue of the PCA, Y_{ik} and α_{jk} are the estimates of principal components of the genotype and environment on the k -axis, and ε_{ij} is the remainder.

AMMI analysis results were interpreted based on two AMMI analysis plots. The first type of diagram was constructed based on the values of the first principal component, genotype, and mean overall fields for the environment, while the second type of diagram was constructed based on the values of the first and second principal components.

ANOVA pooled analysis and AMMI stability analyzes were performed using R package ‘ammistability’ (Ajay et al. 2018). The ranking of genotypes was based on the co-selection index for yield and stability (SSI).

The graphical analysis was carried out using the GGE biplot methodology (Yan, 2001; Yan and Kang, 2003) according to Equation below:

$$Y_{ij} - \bar{Y}_j = \lambda_1 \xi_{i1} \eta_{j1} + \lambda_2 \xi_{i2} \eta_{j2} + \varepsilon_{ij}$$

Y_{ij} : Mean yield of genotype i in medium j , \bar{Y}_j : Average yield of all genotypes in medium j , λ_1 and λ_2 : Characteristic value for PC1 and PC2 (equivalent respectively), ξ_{i1} and ξ_{i2} : scores PC1 and PC2 (respectively) for genotype i , η_{j1} and η_{j2} : scores PC1 and PC2 (respectively) for environment j , ε_{ij} are the remaining phenotypes of genotype i in environment j . GGE biplot analysis was carried out by using R package ‘GGEbiplotGUI’ (Frutos et al. 2014).

Results and Discussion

Combined Variance Analysis

The observed data for all the phenotypic traits recorded at different locations and over the years was processed for combined analysis of variance (ANOVA) and it was revealed that there were significant variations due to the interaction of environment, genotype and genotype \times environment ($p < 0.01$) for plant height, spike length, spikelets per spike, grains per spike and grain yield (Table 2.)

The genotypes exhibited significant level of genotype by environment interaction (GEI), which is attributed to differential adaptation of diverse genotypes across the locations and years. The total variation exhibited by the genotypes with respect to measured traits was partitioned into variation due to genotypes (G), environment (E) and genotype \times environment (GxE) interaction. The major proportion of variation was explained by genotypes for different traits as depicted in Table 2. Spikelet's per spike explained 54.92% of total sum of squares. Likewise, grains per spike and plant height also explained 52.11% and 51.11% of total sum of squares, respectively. This implicates that maximum variation in studied traits is due to genotypic difference. However, significant quantum of phenotypic variation was observed from environments and GEI. This also signifies the role of both genotype and environmental factors. The observed data was also analyzed for AMMI effects and also visualized using GGE biplot analysis with an objective to identify desirable genotypes over environments based on their stability and higher mean yield. The current findings of this investigation are consistent with previous findings (Mwadingeni et al. 2017).

Correlation

Genotypic and phenotypic correlation for 8 studied traits

Improvement of grain yield is a major objective of major wheat breeding programmes and also in other crops as reported in multiple studies. However, to improve yield its direct selection for yield trait is not an effective means (Kumar et al. 2016 and Nagar et al. 2018). Indirect selection for highly contributing traits to yield is more effective in improving novelty of the developed/identified wheat genotypes. Correlated response of yield for indirect selection of effective attributing trait is mainly driven by the level of significant correlation between yield and yield component traits. The critical assessment of results demonstrated a positive and very significant correlation of yield (g/plot), with plant height, tillers per meter, spikelet's per spike, grains per spike and thousand grain weight. (Fig 1). Yield revealed a negative and highly significant correlation with maturity, these findings validate the earlier observations reported from other diverse studies (Baranwal et al. 2012, Nagar. et al. 2018 and Mansouri et al. 2018).

Yield AMMI analysis

Additive analysis of main effects and multiplicative interactions (AMMI) is an effective statistical method for analyzing proportional variation due to genotype-environmental interactions (GEI) as a

major component of the interaction between genotype-dependent variation in grain yield and environmental factors (Rad et al. 2013). This GEI significantly affects the attainment of genetic advance from phenotypic selection due to differential response of genotypes under the target test or productive environments (Mwadzingeni et al. 2017). AMMI analysis revealed that the grain yield is significantly affected by the genotype, environment, and GE interactions and explained 46.65%, 7.83% and 23.58% of variation, respectively. AMMI principal component I and II cumulatively explained 83.8% of genotype x environment variation (Table 3).

All the three interaction principal components (IPCA's) were significant at ($p < 0.01$), among which first IPCA captured 52.8% of interaction sum of square, second 31% and third one contributed 16.2% of interaction sum of squares.

The significant proportion of GEI assures the estimation of phenotypic stability of genotypes over environment (Ajay et al. 2020). A significant proportion of variation (sum of squares) was exhibited due to genotypes diverse nature of genotypes, with respect contrasting features among yield and yield attributing traits. GEI was identified as another significant factor along with environmental variation that attributes differential performance of genotypes for grain and other related traits across the environments.

Based on mean performance in grain yield and AMMI analysis, 10 best selections in terms of their relative performance across the environments were compared and few genotypes were identified that performed better in more than one tested environment as depicted in Table 4. G80 and G33 were best performers across 3 environments (except one) followed by G7, G20, G23, G2, G28, G31, G-19 and G34. The two-way data on grain yield from top performing genotypes and other wheat varieties was used to perform stability analysis using GGE biplot visualization method to precisely identify specific and broadly adapted stable wheat genotypes across different environment.

GGE biplot analysis for grain yield of wheat

Yan et al. (2001) proposed a procedure known as GGE-biplot to graph GE models of interactions with test data in different environments (MET) with different advantages. Two GGE chart analyses consider the influence of genotype (G) and GE interactions and graphically display GE interactions in a two-sided table (Yan et al. 2000). It permits graphical scrutiny of the relationships among the test environments, genotypes and the GE interactions.

Due to the different conditions of the experimental environment, GGE biplot graphic method was used to

study the cultivars and obtain more information about their reaction in these environments. Based on the results of this method, the sum of the first and second main components (PC1 = 92.53 and PC2 = 3.06) explained 95.59% of the variation, which means that these two components were able to 95.59% of the variation. Explain the variation related to grain yield, which indicates the high validity of the biplot diagram obtained from this study in explaining the changes in G + GE.

When the bipolar diagram explains at least 60% of the variance of the data, it can be used to determine large environments (Yang et al. 2009). The details of this method are explained below.

Polygon view (Which-Won-Where Pattern) of GGE biplot analysis for grain yield of MET data

Polygonal 2D plots are the best way to display the presence or absence of GE crosstalk by expressing the interaction patterns between genotype and environment (Yan and Kang, 2003). It provides an efficient and elegant visualization tool for which-won-where patterns in MET datasets useful for evaluating the existence of various mega-tools (Gauch and Zobel, 1997; Yan and Rajcan, 2002; Yan and Tinker, 2006). Fig. 2 represents the polygon view of wheat genotypes for grain yield data in this investigation. In this biplot, a polygon was drawn by joining the genotypes that are located away from the biplot origin, so that all other genotypes are enclosed within the polygon. A genotype located at the edge of a polygon is called a vertex genotype. Separating the GE interactions by GGE biplot analysis revealed that PC1 and PC2 accounted for 92.53% and 3.06% of the GGE sum of squares, respectively, explaining a total variance of 95.59%. The vertex genotypes are G-2, G-38, G-16, G-28, G-19, G-3, G-56, G-57 and G-67. They have the longest vector in each direction, which is a measure of their response to the environment. Therefore, the upper genotype belongs to the most sensitive breeds. All other genotypes are less sensitive in their respective directions. These genotypes were the best or worst genotypes in some or all environments because they were the most distant from the origin of the biplot (Yan and Kang, 2003). The genotypes located at the beginning of the biplane have the same grade in all environments and do not respond at all.

The perpendicular lines are drawn to each side of the polygon, these lines are called equality lines. These lines divide the genotypes and the environments into sections. The polygon view of biplot shows that the genotypes fell in seven sections and the test environments fell in two sections. The first section contains test environments KH18 and WA18 with genotypes G-2, G-38 and G-16 as the best yielder.

And the second section contains environments KH19 and WA19 with genotypes G-28, G-19 and G-3 as the winner. This cross over GE suggests that the target environments may be divided in to two mega-environments. No environments fell in the sections with G-56, G-57 and G-67 as vertex genotypes. This specifies that these genotypes were not the best in any of the test environments, reflecting the fact that they yielded poorly at each environment (Rahmatollah et al. 2013).

Mean and stability performances of genotypes

The productivity and stability of genotypes were evaluated by the average coordinates of the environment (AEC) method (Yang, 2001; Yang, Hunt, 2002; Yang, 2002). In this method, the average environment is determined by the average PC1 and PC2 scores for all environments indicated by the small circles (Figure 3). The line passing through the origin of the two graphs and the average environment is the axis of the average environment. This is the abscissa AEC. The projection of the genotype markers on this axis roughly coincides with the average yield of the genotype. Thus, genotypes are ranked on the AEC abscissa, with arrows indicating higher average performance. Genotype G-2 was clearly the highest yielding genotype, on average, followed by G-28 and G-80, followed by G-21, G-50, G-19, G-1, G-26, G-38, G-42, G-58, G-20, G-10, G-41, G-4, G-23, etc.

The AEC ordinate is a line passing through the origin of the two plots and perpendicular to the AEC abscissa (Figure 3). The transverse AEC estimates G and the longitudinal AEC approximates the GEI associated with each genotype, which measures the variability or instability of the genotype. This means that large projections on the AEC ordinate, regardless of orientation, exhibit significant instability. Therefore, G16 at the top and G3 at the bottom of the 2D plot are more variable and less stable than the other genotypes. Other genotypes with above-average yields include: G-2, G-28, G-80 etc and the genotypes with yield less than mean yield include G-57, G-56, G-67 etc. The ideal genotype for breeding is a genotype with high average yield and high stability. It is close to the origin on the 2D plot and has the shortest vector in ATC. Genotypes with high yield and stability are G2, G50, G26, G80, and G1. In addition, genotypes with high yield but low stability are G28, G7, and G19, which are similar to genotypes with low yield and stability. Low stability was G57 and G56. Genotypes G2 (relatively high yield) and G60 (lowest yield) were parallel in the GE interaction.

Yan and Kang (2003) reported that based on the grain yield and stability performance, genotypes can be

classified into three categories: (1) generally adapted, genotypes with high yield and stability performance (G-2, followed by, G-50, G-26, G-80, G-1 etc.) (2) specifically adapted, genotypes with high mean yield but low stability performance (G-28, G-7 and G-19) and (3) adapted nowhere, genotypes with low grain yield and low stability performance (G-57 and G-56).

Scientists can also use mean vs stability to select the genotypes with best response to specific environments. The genotype G-28 had the highest yielding performance in environment WA18, genotypes G-2 and G-1 performed well in the environments KH-18 and the genotypes G-7 and G-19 performed better in WA19, whereas G-28 was poor in environment WA19 and genotypes G-2 and G-1 had low yield performance in KH19.

Evaluation of genotypes relative to an ideal genotype

The ranking of genotypes according to “ideal” genotypes is shown in the graph (Figure 4). The small circle on the AEC abscissa where the arrow points indicate the ideal class. It is defined by two criteria. 1) It has the highest income among the entire data set. 2) Absolutely stable as indicated by the AEC abscissa. Since such an ideal genotype hardly exists in reality, it can be used as a reference for genotype evaluation (Mitrovic et al. 2012). The closer the genotype is to the “ideal” genotype, the more desirable (Kaya et al. 2006 and Mitrovic et al. 2012). The genotype closer to the “ideal” genotype on this graph is G2. Rank other genotypes by ideal genotype: G50> G1> G80> G26> G21> G42> G38> G23, etc. That is, the low-yield genotypes (G56, G57) were bad because they were far from the ideal genotype.

Evaluation of environments relative to an ideal environment

An ideal environment can be defined based on the projection of the ideal environment on the same horizontal ATC axis as the longest vector of all environments (Figure 5). Environments closer to the hypothetical ideal environment showed that the environment was suitable for genotyping testing. Therefore, the WA18 environment is more suitable, followed by KH19, WA19 and KH18.

Relationship among test environments

Fig. 6 is referred to as a vector view of a 2D GGE plot in which the environment is associated with the origin of the binary plot by a line called a vector. Looking at the 2D graph in this way helps us understand the relationships between the environments. One interesting interpretation is that the angular cosine between the vectors in the two media is approximately equal to the correlation coefficient between the two

media. Acute angles indicate positive correlation, obtuse angles indicate negative correlations, and right angles indicate no correlation (Yan and Kang, 2003). Short vectors can indicate that the test environment is not connected to other environments. The cosine of the angle is not accurately converted to a correlation coefficient because the 2D plot does not account for all the variations in the data set. However, angles provide enough information to provide a complete picture of the relationship between the test environments. Based on the angle between the environment vectors, the environments KH19 and WA19 form an acute angle with each other, so the two environments show a strong positive correlation. Further, KH19 and WA18 and also, KH18 and WA18 made acute angles with each other, therefore these environments are also positively correlated. No negative correlation was found between any of the environments. The 2D vector images were also used to define environments that could be used for indirect selection. It also helps to identify matches between natural and artificial conditions for indirect selection.

Discriminating ability and representativeness of the test environments

Discriminating ability is an important criterion for a test environment. A test environment without discrimination is useless because it does not provide information on genotype (Yan and Kang, 2003). Another equally important indicator of a test environment is its representativeness to the target environment. If the test environment is not representative of the target environment, it is not only useless but also deceptive as it can provide biased information about the genotype tested (Yan and Kang, 2003). In the GGE biplot, genotype distinctness and representativeness of the target environment are important measures of the test environment. Concentric circles in the 2D plot as shown in Figure 1. 7 helps to visualize the length of the media vector, which is proportional to the standard deviation of each media and measures the distinctness of the media. Therefore, among the four test environments, KH19, WA18, and WA19 were the most discriminating (informative) and KH18 was least discriminated. A test environment that is not always indistinguishable (uninformed) should not be used as a test environment as it provides little information about genotype (Yan and Tinker, 2006). The average environment (indicated by the small circle at the end of the arrow) has the average coordinates of all test environments, and the Average Environment (AEA) axis is the average environment and two plots (Yan, 2002). A test environment with a smaller angle with the AEA better represents other test environments. Therefore, after WA18, KH19 and WA19

were the most representative media, and KH18 was the least representative (Figure 7). A differential and representative test environment (location) is generally a good test environment for selecting an adaptable genotype (Yan and Tinker, 2006). Therefore, KH19, WA18 and WA19 were good test environments for selecting widely adapted genotypes. According to Yan and Tinker (2006), when the target environment can be subdivided into mega-environments, a differential but non-representative test environment is useful for selecting especially adaptive genotypes, and when the target environment is a single mega-environment, it is useful to select unstable genotypes. Useful for sorting. environment. On the other hand, indiscriminate and representative environments are meaningless.

Conclusions

The most important goal in all crop breeding programs is to increase yield, and yield improvement requires the use of efficient statistical methods to identify superior genotypes. In determining the superiority of genotype, in addition to high yield, yield stability in different environments must also be considered. AMMI and biplot analyses are good tools for selecting superior genotypes and to increase efficiency in selection. It has been concluded that the combined analysis of variance for yield and yield-associated traits is the best tool for displaying the significance of components of variance among the studied traits. The correlation between the traits depicts the importance of traits related to target trait. The tools like AMMI Analysis and GGE biplot are very effective in order to study the GEI in multi-environments similarly GGE biplot facilitates the graphical representation of GEI pattern of multi-environment traits (MET). It also permits the graphical inspection of the relationship among the test environments and GE interactions. Based on the AMMI analysis and GGE biplot, many genotypes were identified (G-1, G-2, G-3, G-7, G-8, G-16, G-19, G-20, G-21, G-26, G-28, G-31, G-33, G-34, G-38, G-58, G-50, G-80,). Which are suggested to send for further field trails across the country, in order to understand and scrutinize their stability of these genotypes throughout the environments thereafter can be used under breeding programs aimed high yield as well as can be recommended as generally adapted varieties or varieties for specific areas.

Table 1. Experimental material of bread wheat (*Triticum aestivum* L.) used for present study.

Code	Pedigree	Code	Pedigree	Code	Pedigree
G1	CIM-KW-17-1	G35	CIM-KW-17-95	G69	CIM-KW-142-17-163
G2	CIM-KW-17-2	G36	CIM-KW-17-96	G70	CIM-KW-142-17-164
G3	CIM-KW-17-3	G37	CIM-KW-17-97	G71	CIM-KW-142-17-165
G4	CIM-KW-17-4	G38	CIM-KW-17-98	G72	CIM-KW-142-17-166
G5	CIM-KW-17-5	G39	CIM-KW-17-99	G73	CIM-KW-142-17-167
G6	CIM-KW-17-6	G40	CIM-KW-17-100	G74	CIM-KW-142-17-168
G7	CIM-KW-17-7	G41	CIM-KW-17-141	G75	CIM-KW-142-17-169
G8	CIM-KW-17-8	G42	CIM-KW-142-17-142	G76	CIM-KW-142-17-170
G9	CIM-KW-17-9	G43	CIM-KW-142-17-143	G77	CIM-KW-142-17-171
G10	CIM-KW-17-10	G44	CIM-KW-142-17-144	G78	CIM-KW-142-17-172
G11	CIM-KW-17-11	G45	CIM-KW-142-17-145	G79	CIM-KW-142-17-173
G12	CIM-KW-17-12	G46	CIM-KW-142-17-146	G80	CIM-KW-142-17-174
G13	CIM-KW-17-13	G47	CIM-KW-142-17-147	G81	CIM-KW-142-17-175
G14	CIM-KW-17-14	G48	CIM-KW-142-17-148	G82	CIM-KW-142-17-176
G15	CIM-KW-17-15	G49	CIM-KW-142-17-149	G83	CIM-KW-142-17-177
G16	CIM-KW-17-16	G50	CIM-KW-142-17-150	G84	CIM-KW-142-17-178
G17	CIM-KW-17-17	G51	KW-17-12	G85	CIM-KW-142-17-179
G18	CIM-KW-17-18	G52	KW-17-7	G86	CIM-KW-142-17-180
G19	CIM-KW-17-19	G53	KW-17-3	G87	CIM-KW-142-17-181
G20	CIM-KW-17-20	G54	KW-17-8	G88	CIM-KW-142-17-182
G21	CIM-KW-17-81	G55	KW-17-6	G89	CIM-KW-142-17-183
G22	CIM-KW-17-82	G56	KW-17-2	G90	CIM-KW-142-17-184
G23	CIM-KW-17-83	G57	CIM-KW-142-17-151	G91	CIM-KW-142-17-185
G24	CIM-KW-17-84	G58	CIM-KW-142-17-152	G92	CIM-KW-142-17-186
G25	CIM-KW-17-85	G59	CIM-KW-142-17-153	G93	CIM-KW-142-17-187
G26	CIM-KW-17-86	G60	CIM-KW-142-17-154	G94	CIM-KW-142-17-188
G27	CIM-KW-17-87	G61	CIM-KW-142-17-155	G95	CIM-KW-142-17-189
G28	CIM-KW-17-88	G62	CIM-KW-142-17-156	G96	CIM-KW-142-17-190
G29	CIM-KW-17-89	G63	CIM-KW-142-17-157	G97	CIM-KW-142-17-191
G30	CIM-KW-17-90	G64	CIM-KW-142-17-158	G98	Shalimar Wheat-1
G31	CIM-KW-17-91	G65	CIM-KW-142-17-159	G99	Shalimar Wheat-2
G32	CIM-KW-17-92	G66	CIM-KW-142-17-160	G100	HS 562
G33	CIM-KW-17-93	G67	CIM-KW-142-17-161	G101	VL 907
G34	CIM-KW-17-94	G68	CIM-KW-142-17-162		

Table 2. Combined analysis of variance for 8 yield contributing traits at 2 locations under 4 environments.

SV	Df	DM	%	PH	%	T/M	%	SL	%
E	3	180.4	3.61	88.80	5.77	1100	10.14	4.74	11.02
REP (ENV)	4	13.9	0.37	0.504	0.05	181.6	2.23	0.271	0.84
G	100	39.8	26.53	23.56	51.11	156.6	48.13	0.467	36.26
G×E	300	16.2	32.41	3.41	22.39	1.8	16.82	0.119	27.80
Residual	400	13.9	37.06	2.381	20.65	18.4	22.66	0.077	24.14
COV%		1.62		1.65		4.77		2.54	

SV	S/S	%	G/S	%	TGW	%	YIELD	%
E	40.44	18.50	25.96	19.97	35.06	11.57	20440	7.83
REP (ENV)	0.158	0.09	1.365	1.39	0.381	0.16	502	0.25
G	3.6	54.92	2.034	52.11	4.415	48.59	3634	46.45
G×E	0.479	21.93	0.275	21.11	0.566	18.67	615	23.58
Residual	0.072	4.42	0.512	5.35	0.477	20.98	428	21.87
COV%	1.92		2.02		2.33		4.87	

SV= Source of Variation, G= Genotype, E= Environment, Rep (Env) = Replications within Environments, G×E, Genotype × Environment Interaction, CoV= Coefficient of Variation, df= Degree of Freedom, DM= Days to Maturity, PH= Plant Height, T/M= Tillers per Meter, SL= Spike Length, S/E= Spikelet's per Spike, G/S= Grains per Spike and TW= test weight

Table 3. Seed yield variance analysis of wheat promising lines by AMMI analysis.

Source	df	SS	M.S	Percentage Variance
Total	1107	966896	3778	
Environments	3	61320	20440**	7.83
Genotypes	100	363440	3634**	46.65
Rep (Env.)	4	2009	502	0.25
Interactions	300	184503	615**	23.58
IPCA1	102	97365	955**	52.8
IPCA2	100	57163	572**	31.0
IPCA3	98	29975	306**	16.2
Residuals	400	171121	428	21.87

** Significant at 0.01

Table 4. Mean of seed yield and amount of first four interaction principal component analysis of AMMI model in wheat promising lines.

No	KH 18-19		WA 18-19		KH 19-20		WA 19-20	
	G	Y(g/p)	G	Y(g/p)	G	Y(g/p)	G	Y(g/p)
1	G-80	546.17	G-33	540.68	G-31	540.76	G-7	557.22
2	G-20	542.65	G-41	538.92	G-28	540.58	G-1	538.53
3	G-23	541.12	G-34	538.65	G-8	534.81	G-19	536.42
4	G-2	538.44	G-42	536.19	G-80	534.6	G-3	533.6
5	G-50	537.32	G-3	535.61	G-58	530.79	G-21	533.07
6	G-26	531.65	G-26	534.85	G-7	529.79	G-2	531.93
7	G-38	530.95	G-28	534.73	G-21	527.41	G-50	523.74
8	G-31	528.51	G-23	534.72	G-22	525.91	G-28	517.86
9	G-33	528.49	G-20	532.32	G-20	520.72	G-33	517.73
10	G-1	528.12	G-38	526.06	G-34	520.37	G-58	517.72
Mean of Selected Individuals		535.34		535.273		530.57		532.78
Mean of All Individuals		420.99		433.79		446.81		427.71
Selection Differential		114.35		101.48		83.76		105.07
Mean GY (SW-1)		425.07		418.52		457.43		417.09
Mean GY (SW-2)		423.5		436.05		415.89		448.7
Mean HS-562		424.88		432.24		445.46		433.06
Mean VL-907		454.8		430.93		428.06		437.72
Percent Mean of Selected Individuals Over Checks		23.9		24.6		21.5		22.7

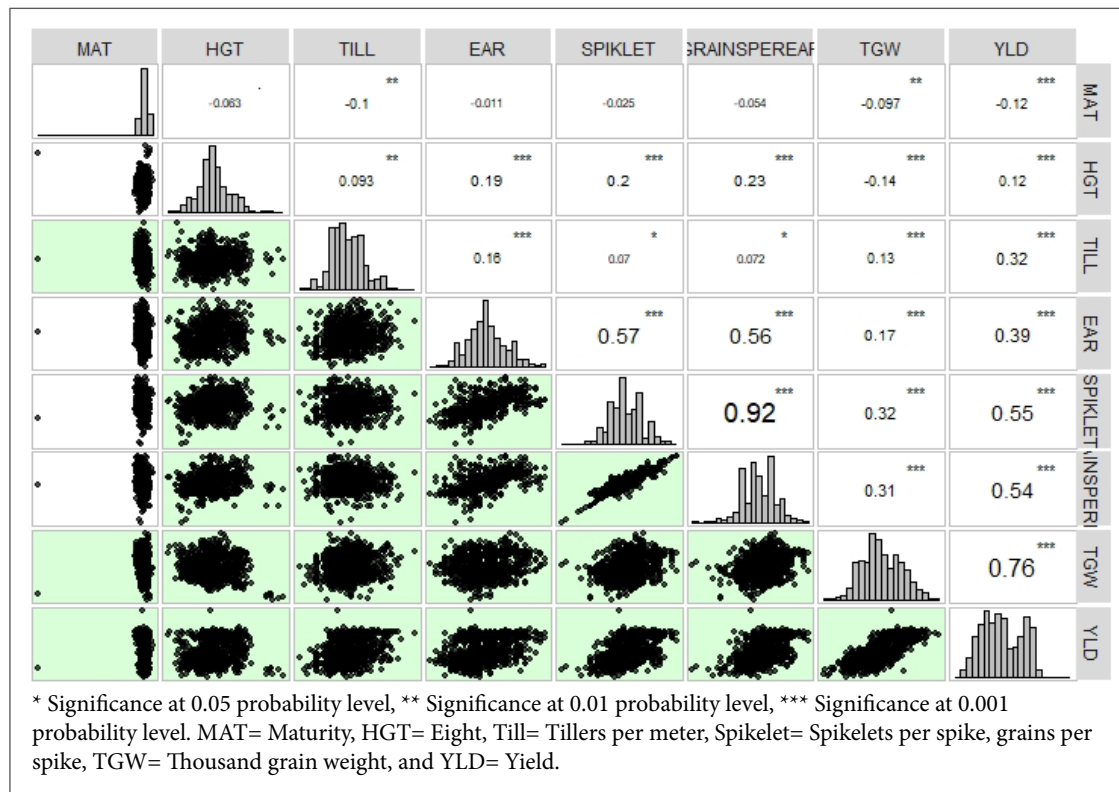


Figure 1. Correlation among yield and yield associated traits in wheat under four environments across two locations. Bigger size of the number is indicator of strong correlation while smaller number size depicts weak correlation between the traits.

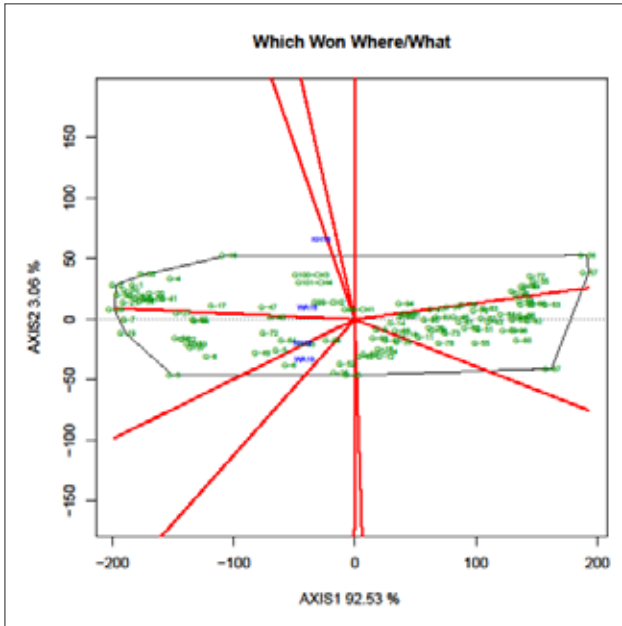


Figure 2. Polygon view of GGE biplot (which-won-where model) showing view of wheat genotypes and environments. Black and blue numbers represent genotypes and environments, respectively.

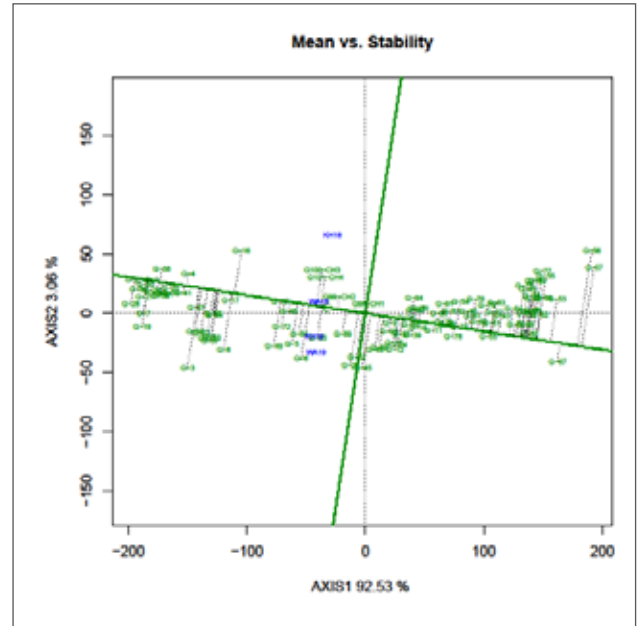


Figure 3. Average environment coordination (AEC) views of the GGE-biplot based on environment-focused scaling for the mean performance and stability of genotypes.

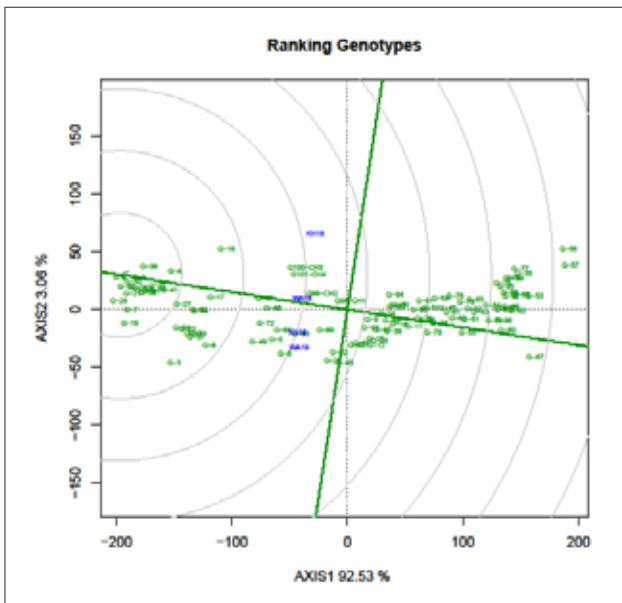


Figure 4. GGE-biplot based on genotype-focused scaling for comparing the genotypes with the ideal genotype.

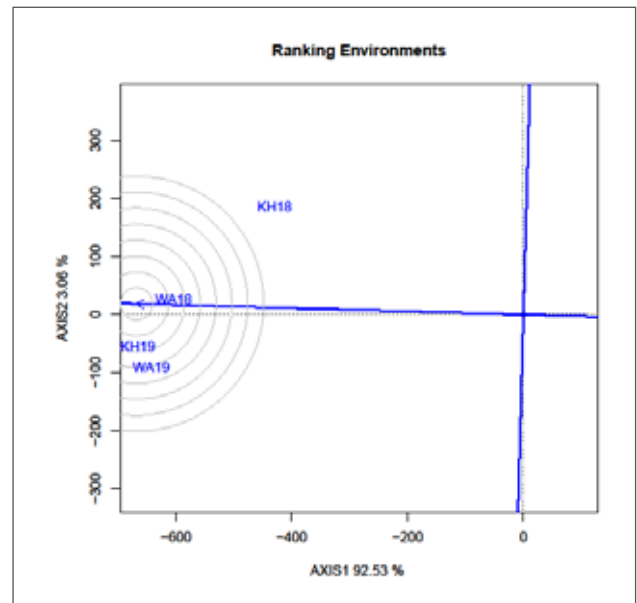


Figure 5. GGE-biplot based on environment-focused scaling for comparing the environments with the ideal environment.

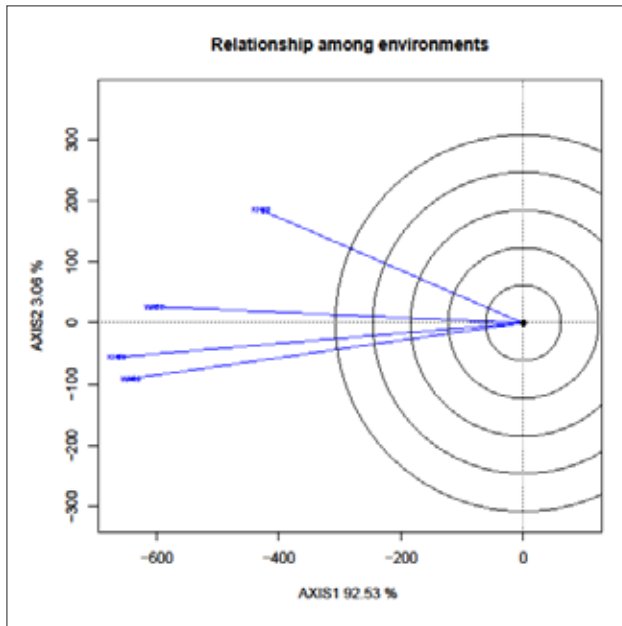


Figure 6. GGE-biplot based on environment-focused scaling for environments.

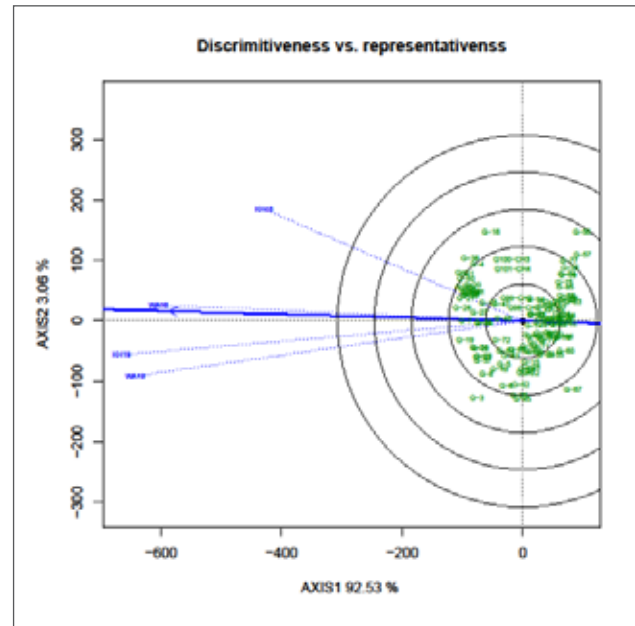


Figure 7. Vector view of the genotype main effect and GGE biplot showing the discriminating ability and representativeness of the test environments.

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Evaluation of Bread Wheat (*Triticum aestivum* L.) Genotypes for Yellow Rust Resistance in Relation to Meteorological Parameters

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ABSTRACT

Three bread wheat varieties (*Triticum aestivum* L.), i.e., HD 2967, WH 711 and PBW 343, were grown in randomised block design with four replications under field conditions over two years in winter season at wheat and Barley Research farm, at CCS Haryana Agricultural University Hisar to evaluate the stripe rust severity in relation to meteorological factors like minimum and maximum temperature (°C) and relative humidity (%), rainfall (mm), soil temperature (°C), canopy temperature (°C), cloud cover (Okta), wind speed (km/h), sunshine hours (h/day) and morning and evening vapour pressure (mm Hg). Observations were recorded on five randomly selected plants in each replication on stripe rust severity as percent leaf area covered using Modified Cobb's scale. Results revealed that low temperature (10-12°C) and high relative humidity (90%) along with intermittent rainfall were found conducive for the onset of stripe rust. Low temperature and moderate humidity favored the disease initiation and development. At the initiation of disease, the severity was more in PBW 343 (0.68%) followed by WH711 (0.59%) during first SMW whereas HD 2967, exhibited disease initiation during 2nd SMW with 2.19% severity. Disease progression was also different in different wheat varieties. Disease gradually advanced with the crop age, and increased sharply from 15.00 to 38.00% in HD 2967, 27.50 to 46.00% in WH711 and 35.00 to 43.6% in PBW 343 during 6-8th Standard Meteorological Week, when the crop was at stem elongation to jointing stage, i.e., GS 36-47 (64-78 DAS). Yield loss due to disease severity was estimated to be 356 kg/ha, 690 kg/ha and 400 kg/ha in wheat varieties HD2967, WH711 PBW343, respectively. Step-wise multiple regression showed high R² value of 0.885, 0.919 and 0.952 for the predictive model of stripe rust in HD 2967, WH 711 and PBW 343, respectively

Keywords: Stripe rust, bread wheat, *Triticum aestivum* L., temperature, rainfall, relative humidity

Introduction

Bread wheat is one of the most important cereal crops meeting daily calorie need of about 35% population worldwide. Although bread wheat has a wide range of climatic adaptability, but it is usually affected by many fungal diseases, the most devastating of which is the rust. Stripe (yellow) rust is widespread disease across the major bread wheat growing regions with diverse cropping systems, growing seasons and germplasm characteristics (Wellings, 2011). The worldwide loss in bread wheat production due to stripe rust has been estimated to be at least 5.5 million tonnes per year. (Beddow et al. 2015). Globally, the

epidemics of stripe rust was reported in United States (Line, 2002), Australia (Wellings et al. 2000), Middle East (Akar et al. 2007), New Zealand and China, (Chen et al. 2009) while in South Asia (India, Pakistan and Nepal), the yield loss was reported moderate to high over the years (Singh et al. 2004). The yield loss was usually due to the result of poor root growth, reduced dry matter, low test weight, reduced kernel number size and quality (Wellings, 2011).

Stripe rust caused by the obligate biotroph *Puccinia striiformis*, is a serious fungal disease of bread wheat, especially under the cool and moist conditions (Chen et al. 2014). The minimum temperature for the

occurrence of stripe rust infection is 0°C, optimum 11°C and maximum 23°C (Curtis et al. 2002), however, due to the changed climate conditions occasionally, yellow rust predominated over leaf rust (Jevtić et al. 2017).

To avoid epidemics and yield loss, breeding for disease resistance is the main strategy for the management of diseases (Chen et al. 2014). Bread wheat cultivars become susceptible to rusts due to their narrow genetic base and the rapid evolution of new pathogen races, making it necessary for the search of new resistance sources. An effective and economical approach to reduce yield loss caused by the diseases is the development of disease resistant varieties. The disease severity is determined by the congeniality of pre-disposing meteorological factors and/or abiotic stresses such as drought and salinity affecting plant, water relationships, besides genetic factors in wheat varieties. (Pandey et al. 2017). To develop an effective management strategy for the management of stripe rust, it is essential to evaluate key factors involved in initiation and progress of diseases. Understanding of meteorological parameters is prerequisite for providing baseline information to develop simple and reliable disease prediction system. Keeping this fact in view, it is imperative to develop a location specific reliable prediction system for stripe rust in bread wheat. The present paper deals with stripe rust severity in three wheat varieties in relation to prevailing meteorological conditions and to develop a model for prediction of disease severity in semi-arid conditions.

Materials and Methods

Three bread wheat (*Triticum aestivum* L.) varieties, i.e., HD 2967, WH711 and PBW 343, were grown in randomized block design with four replications under field conditions over two years in winter season at Wheat and Barley Research farm, Department of Genetics and Plant Breeding, CCS Haryana Agricultural University Hisar to evaluate the effect of different meteorological factors and stripe rust severity. These selected varieties were sown under late sown conditions in first week of December in plots of 8 m² size at a row spacing of 22.5 cm with 12 lines in each plot. The infector rows were placed after the interval of 10 lines as border rows of the experiment to ensure uniform infection. Inoculums were sprayed at tillering stage with urediniospores of *Puccinia striiformis* (concentration 10⁻⁶ spore/ml). Plants were screened under epiphytotic conditions and data in terms of per cent leaf area infected were recorded by using Modified Cobb's Scale (Peterson et al. 1948) on five randomly selected plants until crop

maturity (14th Standard Meteorological Week). Disease severity was recorded in terms of per cent leaf area infection, and pustule type was recorded as response.

The effect of different meteorological factors viz., minimum and maximum temperature (°C), minimum and maximum relative humidity (%), rainfall (mm), soil temperature (°C), canopy temperature (°C), cloud cover (Okta), wind speed (km/h), sunshine hours (h/day) and morning and evening vapour pressure (mm Hg) were studied on the development of stripe rust in bread wheat. The Meteorological data of one week prior to the date of observation were collected from the meteorology department of CCS Haryana Agricultural University Hisar. The canopy temperature was recorded with infrared thermometer (Ramson make) and soil temperature with soil thermometer (Japson make) at the depth of 5, 10 and 20 cm. During both the seasons, from the last week of December onward, the selected varieties in the experimental plots were monitored regularly for the initial foci of *Puccinia striiformis*.

Statistical analysis

To determine the cumulative effect of meteorological parameters and prediction of stripe rust severity, multiple stepwise regression was computed to generate models through SPSS 16.0 (Cornell and Berger 1987), having coefficient of determination (R²) and F_{cal} value e< 0.0001. The correlation coefficients (r) were determined to find the effect of single as well as combined meteorological parameters on the severity of stripe rust in bread wheat as proposed by Madden, 1986). Further, the graphs of recorded and predicted values were determined by the stepwise regression model to evaluate the accuracy of equations in predicting the disease.

Results and Discussion

Effect of meteorological factors on stripe (yellow) rust of bread wheat

The disease severity *vis-a-vis* meteorological parameters in all the selected bread wheat varieties exhibited that under late sown conditions appeared in 1st Standard Meteorological Week, when the crop was at tillering stage GS 21, i.e., 28 days after sowing (Table 1). The minimum and maximum temperature was 7°C and 18°C, minimum and maximum relative humidity 49 and 92%, sunshine 2.9 h/day, morning and evening cloud cover 3.8 and 6.5 okta, mean wind velocity 1.5 km/h, morning and evening vapour pressure 7.2 and 9.0 mmHg, soil temperature 11.4°C and canopy temperature 13.4°C. Singh et al. (2002) reported that minimum, optimum and maximum temperature for urediniospores penetration, growth and sporulation in host were 2, 12-15 and 23, 3, 12-15 and 20 and 5,

12-15 and 20°C, respectively. deVallavielle-Pope et al. (2002) documented that urediniospores infection by *Puccinia striiformis* was higher under the conditions of long light hours and prolonged clear sky. It was reported that yellow rust would predominate over leaf rust if requirement for high winter temperature was met (Hovmøller et al. 2016).

At the initiation of disease, the severity was more in PBW 343 (0.68%) followed by WH711 (0.59%). In HD 2967, the disease appeared in 2nd SMW with 2.19% severity, when crop was at late tillering stage GS 36 (36 DAS). Thereafter, disease gradually advanced with the crop age, but increased sharply from 15.00 to 38.00% in HD 2967, 27.50 to 46.00% in WH711 and 35.00 to 43.6% in PBW 343 during 6-8th Standard Meteorological Week, when the crop was at stem elongation to jointing stage, i.e., GS 36-47 (64-78 DAS). This coincided with minimum and maximum temperature of 8.2-10.6 and 19.5-23°C, minimum and maximum relative humidity of 51-62 and 81-97%, mean wind velocity of 3.0-1.2 km/h, morning and evening vapour pressure of 11.0-13.5 and 12.5-16.0 mmHg, sunshine of 3.6-6.8 h/day, morning and evening cloud cover of 6.8-4.0 and 3.0-1.5 okta, soil and canopy temperature of 13.8-15.9 and 17.0-18.6°C.

At the milk development stage, i.e., GS 75 (120 DAS) in 14th Standard Meteorological Week, maximum disease severity was recorded in WH711 (74.0%) followed by PBW 343 (73.2%) and HD 2967 (61.5%), during which, the minimum and maximum temperature was 11.1 and 28.0°C, minimum and maximum relative humidity 46.5 and 81.0%, mean wind velocity 5.3 km/h, morning and evening vapour pressure 8.5 and 14.0 mmHg, sunshine 9.4 h/day, rainfall 18.3 mm, morning cloud cover 1.0 okta and soil and canopy temperature 20.6 and 24.8°C, respectively. Chen et al. (2014) also reported that cool and moist conditions with low temperature (7-12°C) were favourable for stripe rust epidemic. The minimum temperature of 11.7°C coupled with 8.2 h of bright sunshine per day was found favourable for faster development of stripe rust of bread wheat (Singh and Tewari, 2001).

Correlation of meteorological factors with the severity of stripe rust

Correlation coefficient between various meteorological factors and stripe rust were computed, which revealed that in HD 2967, the minimum and maximum temperature, morning and evening vapour pressure, canopy temperature, soil temperature and age had significantly positive correlation with the disease severity (0.836, 0.848, 0.685, 0.836, 0.812, 0.936 and 0.978), respectively (Table 2). In WH711 and PBW 343, the 'r' value was 0.843, 0.844, 0.700, 0.856, 0.830,

0.926 and 0.992, and 0.859, 0.846, 0.705, 0.844, 0.850, 0.922 and 0.990, respectively. The maximum relative humidity had significantly negative correlation with the disease severity in the tested varieties, wherein the correlation coefficients were -0.636, -0.651 and -0.650 respectively. Likewise, correlation coefficients for the three varieties between disease severity and rainfall were 0.550, 0.546 and 0.513, respectively indicating positive association. Ahmed et al. (2010) recorded strong correlation of minimum and maximum temperature and sunshine radiation with stripe rust. Christensen et al. (1993) found that temperature in January and February was significantly correlated with the severity of stripe rust in bread wheat. Chen., (2005) recorded that moisture enhanced spores' germination and infection, but the availability of free water inhibited infection by reducing the viability and survival of the pathogen. Yield reduction was observed in tested bread wheat varieties due to stripe rust (Table.4). In general, a variable yield loss was observed in tested varieties due to different levels of stripe rust severity at anthesis (Fig. 2).

Development of predictive model

Results of this study have been used to develop a predictive model for predicting the possible incidents and severity of the yellow rust in different varieties of wheat cultivated in north west plain zone of India. The model is based on analysis of variance, correlation and regression coefficients among, meteorological parameters and disease severity. In WH711, the model was highly significant, having $F_{cal}(3, 10) = 38$ at $p \leq 0.01$ in predicting severity of stripe rust with R^2 value of 0.919. This indicated that cumulative effect of minimum temperature, maximum temperature including rainfall explained 91.90% variation. The results revealed an increase or decrease in disease severity by 0.90% per week by per unit increase or decrease in rainfall, if all the other parameters remained congenial (95% CI from 0.03 to 1.80%), whereas, 2.77 and 3.82% increase or decrease in disease severity was predicted per week by per unit increase or decrease in maximum and minimum temperature, if all the other parameters remained favourable (95% CI from 1.20 to 4.33 and 1.83 to 5.82%), respectively. The contribution of minimum and maximum temperature and rainfall in the prediction model was 51, 37 and 12%, respectively. Plotting of observed versus predicted value of disease severity (Fig. 1a) showed good association ($R^2 = 91.90\%$).

In HD 2967, the developed model for predicting severity of stripe rust was highly significant having $F_{cal}(2, 11) = 42$ at $p \leq 0.01$, explaining 88.50% variation in the prediction model by minimum and maximum temperature. The contribution of minimum and

maximum temperature was 43 and 57%, respectively. The observed versus predicted disease severity value (Fig.1b) showed good association ($R^2= 88.50\%$). It further revealed that, in PBW 343, the prediction model was highly significant, having $F_{cal}(3,10)= 73$ at $p \leq 0.01$, with R^2 value of 0.952, which indicated that 95.20% variation was due to canopy temperature, morning vapour pressure and rainfall having contribution of 34, 52 and 24%, respectively. Plotting of observed versus predicted value of severity (Fig.1c) exhibited good association (95.20%). The prediction models developed through stepwise multiple regression analysis depicted that under late sown conditions the maximum and minimum temperature, vapour pressure and canopy temperature and rainfall variables were found to account for approximately 85-97% variation. In all the three developed prediction models, temperature was the key variable, which influenced the stripe rust severity and epidemics. Coakley et al. (1988) also developed model for the prediction of stripe rust based on temperature, total precipitation and frequency of precipitation for susceptible cultivar (Omar). The best model describing disease severity of *stripe* rust consisted of maximum temperature and rainfall with R^2 value of 86.40% (TeBeest et al. 2009). Jarroudi et al. (2017) reported that the prediction models are useful in predicting the yellow rust severity, so that the appropriate management strategies can be adopted.

Conclusions

Yellow rust resistance has assumed significance in recent years in North-West plain zone of India due to climate change and consequent combination of pre-disposing factors like low and high temperatures, relative humidity, vapour pressure deficit, sunshine etc. The importance of wheat breeding for yellow rust resistance has been realized in recent years to avoid yield losses to ensure higher wheat production for food security. In present study, three wheat varieties grown extensively in the region have been investigated for severity of the yellow rust *vis-a-vis* meteorological parameters. Attempts have been made to develop a prediction model based on stepwise multiple regression using data on meteorological parameters and yellow rust severity. All the three varieties established significance of different meteorological parameters being rainfall, temperature and vapour pressure deficit. Future attempts are needed to develop comprehensive prediction model by including more variables for meteorological parameters like radiation intensity, photoperiod, number of spores in ambient air monitored through spore trap, *Yr* genes, pustules number and

size onset of disease infection and consequent disease progression. Such a model will be useful in developing a management strategy integrating genotypic variability for yellow rust resistant genes, yield attributes and grain yield *per se* as well as agronomic factors such as staggered sowing so that the disease severity could be kept controlled or bare minimum for sustainable production of wheat.

Table 1. Effect of different meteorological factors on the severity of stripe rust of bread wheat (pooled data).

SMW	Disease Severity in Bread Wheat Varieties		Age (days)	C _{Temp} (°C)	T _{max} (°C)	T _{min} (°C)	RH _{Max} (%)	RH _{Min} (%)	M _{WV} (km/h)	Vp _{Mor} (mmHg)	Vp _{Evnt} (mmHg)	SS (h)	Rainfall (mm)	CC _{Morn} (okta)	CC _{Evnt} (okta)	S _{Temp} (°C)
	HD2967	WH711														
1 st	0.00	0.59	0.68	13.4	18.0	7.0	92.0	49.0	1.5	7.2	9.0	2.9	0.0	3.8	6.5	11.4
2 nd	2.19	3.17	3.17	14.0	14.7	5.8	94.0	68.5	2.0	8.3	9.3	0.8	0.6	6.5	5.0	11.8
3 rd	3.83	5.50	7.50	15.0	15.7	4.3	95.0	71.0	4.8	7.3	10.1	7.2	0.0	0.5	0.5	11.6
4 th	6.50	11.67	10.83	14.9	16.0	7.9	91.5	75.0	1.6	9.3	11.0	0.0	5.0	3.0	5.0	13.3
5 th	10.00	17.50	24.17	17.5	20.6	4.6	91.5	50.5	2.0	9.4	12.1	4.7	0.0	2.0	1.5	13.2
6 th	15.00	27.50	35.00	18.6	19.5	10.6	81.0	62.0	3.0	11.0	12.5	3.6	0.0	6.8	3.0	13.8
7 th	25.00	38.33	40.00	20.3	22.6	6.1	93.0	48.5	2.1	10.7	12.8	6.2	0.0	2.5	2.5	14.9
8 th	38.00	46.00	43.60	17.0	23.0	8.2	97.0	51.0	1.2	13.5	16.0	6.8	0.0	4.0	1.5	15.9
9 th	40.00	50.00	50.00	18.0	17.5	11.7	88.5	77.0	1.9	13.3	14.8	0.5	0.0	5.0	5.0	14.8
10 th	43.33	55.00	55.00	18.1	21.5	9.3	89.5	54.0	3.0	12.4	12.3	5.9	14.6	2.0	2.5	15.1
11 th	48.33	60.00	60.00	17.2	22.7	9.4	81.0	52.0	2.2	11.8	13.7	4.7	9.8	4.0	4.0	16.8
12 th	53.33	63.33	65.00	22.2	27.5	11.7	87.0	46.5	2.1	15.5	15.5	6.8	0.0	2.0	0.5	19.9
13 th	56.67	66.67	70.00	23.4	24.0	14.7	83.0	72.0	5.8	14.5	14.1	2.2	3.7	5.5	5.0	20.9
14 th	61.50	74.00	73.20	24.8	28.0	11.1	81.0	46.5	5.3	8.5	14.0	9.4	18.3	1.0	0.0	20.6

Note: SMW = Standard meteorological week, C_{Temp} = Canopy temperature, T_{Max} = Maximum temperature, T_{Min} = Minimum temperature, RH_{Max} = Maximum relative humidity, RH_{Min} = Minimum relative humidity, M_{WV} = Mean wind velocity, Vp_{Mor} = Morning vapour pressure, Vp_{Evnt} = Evening vapour pressure, SS = Sunshine, CC_{Morn} = Morning cloud cover, CC_{Evnt} = Evening cloud cover, S_{Temp} = Soil temperature

Table 2. Correlation of different abiotic factors with the severity of stripe rust of bread wheat (pooled data).

Epidemiological Factor	Disease Severity (%) in Bread Wheat Varieties		
	HD2967	WH711	PBW343
T_{max} (°C)	0.836	0.843	0.859
T_{min} (°C)	0.848	0.844	0.846
RH_{Max} (%)	-0.636	-0.651	-0.650
RH_{Min} (%)	-0.356	-0.385	-0.394
M_{WV} (km/h)	0.388	0.362	0.392
Vp_{Mor} (mmHg)	0.685	0.700	0.705
Vp_{Evn} (mmHg)	0.836	0.856	0.844
SS(h)	0.406	0.416	0.420
Rainfall (mm)	0.550	0.546	0.513
C_{Temp} (°C)	0.812	0.830	0.850
CC_{Morn} (okta)	-0.099	-0.084	-0.071
CC_{Evn} (okta)	-0.294	-0.310	-0.331
S_{Temp} (°C)	0.936	0.926	0.922
Age (days)	0.978	0.992	0.990

Note: Values in bold are highly significant at (p= 0.05). C_{Temp} = Canopy temperature, T_{Max} = Maximum temperature, T_{Min} = Minimum temperature, RH_{Max} = Maximum relative humidity, RH_{Min} = Minimum relative humidity, M_{WV} = Mean wind velocity, Vp_{Mor} = Morning vapour pressure, Vp_{Evn} = Evening vapour pressure, SS= Sunshine, CC_{Morn} = Morning cloud cover, CC_{Evn} = Evening cloud cover, S_{Temp} = Soil temperature

Table 3. Predictive Model for stripe rust of bread wheat.

Bread Wheat Variety	Model	R ²	F-Value	95% CI Lower-Upper
HD2967	$Y = -61.127 + 3.75X_2 + 2.75X_1$	0.885	42.4	$X_2 = 1.81-5.68$ $X_1 = 1.24-4.25$
WH711	$Y = -61.52 + 3.81X_2 + 2.67X_1 + 0.90X_5$	0.919	37.6	$X_2 = 1.83-5.82$ $X_1 = 1.20-4.33$ $X_5 = 0.03-1.80$
PBW343	$Y = -82.41 + 3.33X_4 + 4.99X_3 + 1.36X_5$	0.952	72.6	$X_4 = 2.00-4.66$ $X_3 = 3.33-6.64$ $X_5 = 0.65-2.08$

Note: Y= Disease severity of HD2967, WH711 and PBW343, $T_{Max} = X_1$, $T_{Min} = X_2$, $Vp_{Morn} = X_3$, $C_{Temp} = X_4$ and Rainfall= X_5 .

Table 4. Yield reduction in different bread wheat varieties due to stripe rust (pooled data).

Variety	Yield in Control (kg/ha)	Yield in Diseased Field (kg/ha)	Yield Loss (kg/ha)	Yield Loss (%)	Disease Severity (At Anthesis Stage)
PBW343	4260	3820	440	10.32	40
WH711	4630	3940	690	14.90	40
HD2967	4336	3980	356	8.21	30

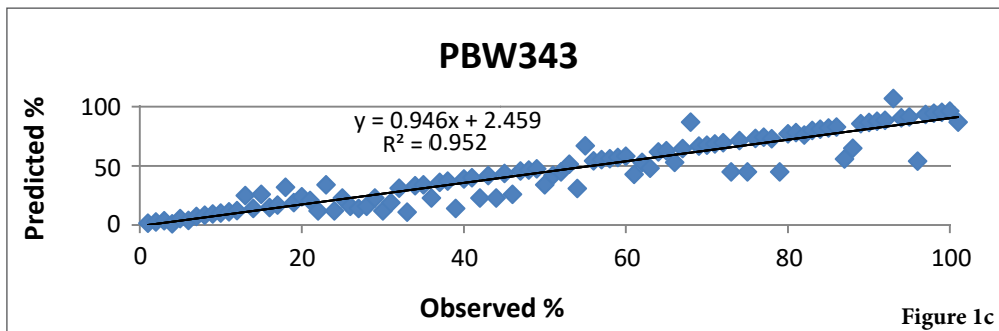
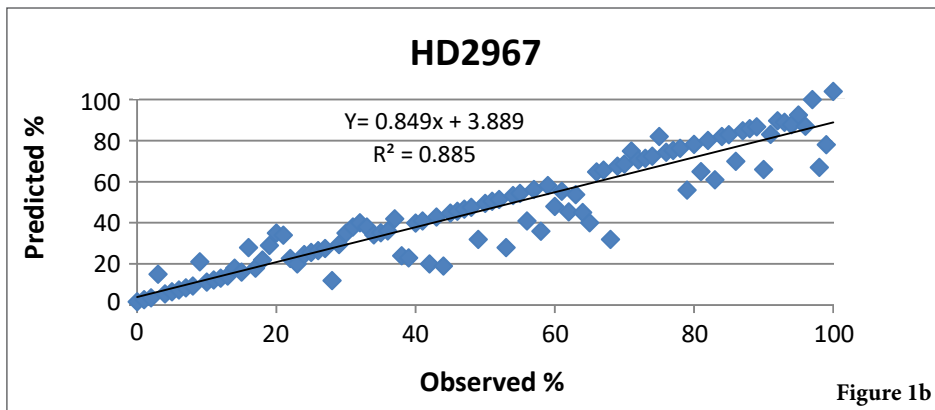
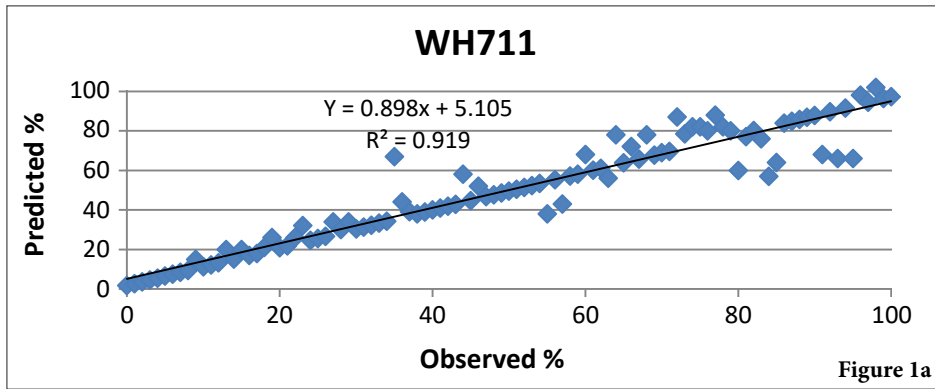


Figure 1a-c. Comparisons between observed and predicted values of the severity of bread wheat stripe rust in different bread wheat varieties.

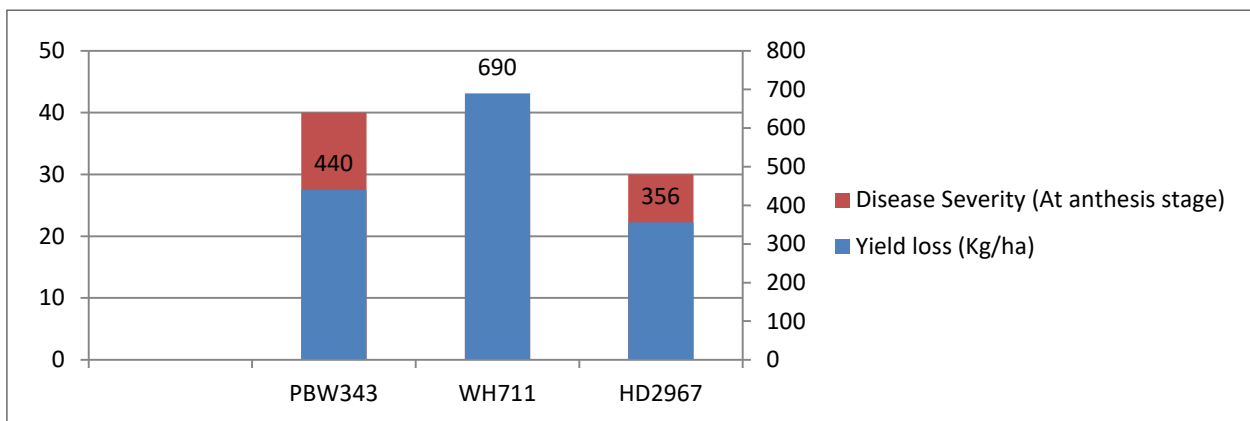


Figure 2. Estimated yield losses in response to different levels of stripe rust at anthesis.

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Combining Ability Analysis for Yield and Fibre Quality Traits in American Cotton (*Gossypium hirsutum* L.) Genotypes

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ABSTRACT

The present study was conducted with the aim of determining combining ability and variances for different traits and to identify the best combiners with respect to seed cotton yield and fiber quality. Forty eight F_1 hybrids along with parents were grown in randomized block design with three replications and the data were recorded for 14 characters namely, days to 1st flower, plant height, number of monopodial branch per plant, number of sympodial branch per plant, number of bolls/plant, boll weight, seed index, ginning out turn lint index, seed cotton yield per plant, uniformity index, fibre strength, upper half mean length and micronaire value. The variance gca/sca ratio depicted predominance of non-additive gene action for days to first flowering, number of bolls per plant, boll weight, seed index, seed cotton yield per plant, fibres strength, uniformity index, upper half mean length and micronaire value. The characters lint index and number of monopods per plant revealed non-additive and additive gene action while plant height, number of sympods per plant and ginning out turn were predominantly controlled by additive gene action. The lines H1471, H 1472, H 1522 and H 1098i and the tester C 201 were found best combiners to be considered for future breeding programme. The cross combination H 1480 × C 201 recorded highest significant SCA effects for seed cotton yield per plant followed by H1098i × C 211 and H 1522 × C210.

Keywords: Combining ability, American cotton genotypes, Line x tester, *Gossypium hirsutum*

Introduction

Cotton is the leading natural fiber and grown in more than 50 countries in the world. It is one of the commodities with global importance and high commercial value providing income to millions of farmers worldwide. India is at rank number one in terms of area under cotton cultivation and raw cotton production in the world. Per hectare productivity in India still much lower compared to many leading cotton growing countries. (ICAR-AICRP (Cotton), Annual Report 2020-21). Development of new varieties/hybrids of different cultivated cotton species with high lint yield and lint quality is the primary objective of all cotton breeders. For registration of a new variety, it is required to test for DUS traits. In the cotton

crop, a plethora of studies has been conducted for the classification of genotypes on the basis of DUS traits (Sagar et al. 2019; Kumar et al. 2021a). The yield contributing traits and fibre quality parameters are the important character in cotton improvement programme. Heterosis breeding is an important breeding technique to facilitate yield enhancement and help to enrich many other desirable quantitative and qualitative traits in crops. Generally, the development of cotton hybrids/varieties for higher lint yield with desirable fibre quality parameters is the most important objective of the cotton improvement programs. Cotton fiber quality is expressible by a multitude of measurements (Hake et al. 1996). Fiber length, fiber fineness, fiber strength, short fiber index and the spinning consistency index

are the most important fiber quality traits (Ekinçi and Basbag 2018). However, further increase in raw cotton productivity is a challenging mission for breeders due to extensive use of local available germplasm (Tyagi et al. 2014; Zhang et al. 2020 and Kumar et al. 2021b) and the high impact of environmental fluctuations on the yield and yield contributing traits.

To achieve higher lint yield with good fibre quality in cotton, it is essential to select appropriate parents for a successful breeding programme. Line x Tester analysis provides a systematic approach for finding of suitable parents and F_1 hybrids for various investigated traits. Combining ability describes the breeding value of parental lines in hybrid production. Sprague and Tatum (1942) reported that GCA effects were due to additive type of gene action however SCA effects were due to genes which are non-additive (dominant and epistatic) type of gene action.

The present investigation was conducted to estimate GCA effects of Parents as well as the SCA effects of the crosses for yield and fibre quality traits in American cotton and to determine appropriate parents and hybrids for the tested characters.

Materials and Methods

The materials used in present investigation were selected based on their genetic diversity. The parental materials were selected from different experiment sown during *Kharif* 2016 at Cotton section, Department of Genetics and Plant Breeding, research area (Table 1). The selected material comprising of sixteen parents (12 lines + 4 testers) were grown in crossing block during *Kharif* 2017 and crossing were made by Line x Tester mating design which results into development of forty eight F_1 hybrids.

The 48 F_1 hybrids and their parents (16) i.e. twelve lines and four testers along with the check hybrid HHH 223 were grown during *Kharif* 2018. Each entry was sown in two rows of 3.0 meter length adopting a spacing of 67.5 cm between rows and 60 cm between the plants in randomized block design with three replications. All the recommended packages of practices were followed from sowing to picking. Five competitive plants were randomly selected and observations were recorded from them from each entry i.e. parents, hybrids and check for days 1st flowering, plant height (cm), number of monopods per plant, number of sympods per plant, number of bolls per plant, boll weight (g), seed index (g), seed cotton yield per plant (g), ginning outturn (%) and lint index (g). Lint samples were sent to CIRCOT Lab Sirsa for analysis of four fibre quality traits, uniformity index (%), fibre strength (g/tex), upper half mean length (mm) and micronaire value ($\mu\text{g}/\text{inch}$).

The analysis of variance was carried out as per the standard statistical method to test the significance of differences among the tested materials. Evaluation of hybrids and parents involved in line x tester analysis was taken up separately by conducting a relevant RBD analysis first and later the variation among the crosses and parents was partitioned through combining ability analysis. The genetic variation among the F_1 hybrids was further partitioned into genetic components attributable to general combining ability (GCA) and specific combining ability (SCA) following the method as reported by Kempthorne (1957).

Results and Discussion

The analysis of variance for Line x Tester for different characters of American cotton genotypes as depicted in the table 2 indicated that significant variations were present among the genotypes for the characters i.e. days to 1st flower, plant height, number of monopods per plant, number of bolls per plant, boll weight, seed index, seed cotton yield per plant, ginning outturn, lint index, uniformity index, fibre strength, upper half mean length and micronaire value except for number of sympodial branches per plant revealing sufficient amount of wide genetic diversity as indicated by the significance of the mean squares.

The forty eight cross combinations obtained by crossing twelve lines and four testers were subjected into line x tester (L x T) analysis. The analysis revealed significant differences among lines for all parameters under investigation except number of monopods per plant, number of sympods/plant and all fiber quality parameters (Table 2). The significant differences were also observed among the testers for all traits except for number of sympods/plant and for all fiber quality parameters. Line x tester interactions was significant for number of bolls per plant, seed cotton yield, seed index, ginning out turn, lint index and fibre strength.

The study revealed greater magnitude of SCA variance than GCA in all the characters except for plant height, number of monopods per plant, number of sympods per plant and ginning out turn depicting the role of non-additive gene action for the said character. Therefore, heterosis breeding may be rewarding for the improvement of the said characters in cotton. The estimate of GCA and SCA variances was presented in Table 3. The ratio of $\sigma^2\text{GCA}/\sigma^2\text{SCA}$ was less than unity for the characters *viz.* Days to 1st flower, number of bolls per plant, boll weight, seed index, seed cotton yield and lint index indicating preponderance of non additive gene action (dominance and epistasis), which is an important in exploitation of heterosis through hybrid breeding. Several authors Vekariya et al. (2017),

Shinde et al. (2018) Swetha et al. (2018), Chinchane et al. (2018) and Premalatha et al. (2020) have reported the predominance of SCA variance in cotton for morphological, yield and its component characters. Similar results were observed for fibre quality traits *viz.*, upper half mean length, fibre strength and micronaire value *i.e.* the ratio of $\sigma^2GCA / \sigma^2SCA$ was less than unity. Variance of GCA effects were higher than variance of SCA effects ($\sigma^2GCA / \sigma^2SCA > 1$) for the characters plant height, number of sympods per plant and ginning outturn which indicated that these traits are controlled by additive gene action. The number of monopods per plant and lint index observed the ratio of $\sigma^2GCA / \sigma^2SCA$ near to unity which indicated that additive gene is prevailing with non-additive gene actions for the expression of these traits. The results are in agreement with earlier reported the findings of Lukonge et al. 2008; Bolek et al. 2010 and Ekinçi and Basbag 2018.

The GCA effects of parents presented in table 4 revealed that among female parents the genotype H 1471, H 1472, H 1522 and H 1098i were best combiners for seed cotton yield. The genotypes observed good general combiner for days to first flower *i.e.* earliness (H 1471), plant height (H 1480), no. of sympods per plant (H 1518), no. of bolls per plant (H 1480), boll weight (H 1522) seed index (H 1519), ginning out turn (H 1518) and lint index (H 1518). For fibre characters *i.e.* uniformity index, UHML, fiber strength and micronaire value good general combiners are H 1519, H 1523, H 1471 and H 1522 respectively. Among testers C 201 was the best combiner for earliness and good general combiner for seed cotton yield/plant, boll weight, number of monopods/plant, seed index, ginning out turn and lint index. Thus, high gca effect in desirable indicates the presence of additive genes for that character in the parent therefore, selection is effective for improvement of the characters. Bayyapu Reddy et al. (2017), Bilwal et al. (2018) also reported different parents with good general combining ability for seed cotton yield and yield attributing characters.

The specific combining ability effects are usually used to make out the best cross combinations for hybrid production. The SCA effects of the crosses for various characters were presented in table 5. Among forty eight hybrids, the following nine hybrids, H 1472 \times C 202, H 1480 \times C 201, H 1488 \times C 210, H 1508 \times C 210, H 1518 \times C 201, H 1518 \times C 202, H 1522 \times C 210, H 1523 \times C 211 and H 1098i \times C 211 registered positive and significant specific combining ability for seed cotton yield/plant. Similar findings were reported by Bayyapu Reddy (2017) and Premalatha et al. (2020).

The crosses H 1471 \times C 201, H 1523 \times C 202, H 1471 \times C 211, H 1471 \times C 211, H 1523 \times C 202, H 1489 \times C 210, H 1098i \times C 202, H 1508 \times C 211, H 1472 \times C 201, H 1520 \times C 211, H 1508 \times C 211, H 1520 \times C 211 and H 1520 \times C 201 recorded the highest SCA effects for days to 1st flowering, plant height, number of monopods/plant, number of sympods/plant, number of bolls/plant, boll weight, seed index, ginning outturn, lint index, Uniformity index, upper half mean length, fiber strength and micronaire value, respectively. The SCA effects may not only be the appropriate choice for exploitation of heterosis, because the hybrids with low mean value may also depict high sca effect. Hence, the cross having significant sca effects should be evaluated under different locations over the years prior to suggest for commercial cultivation.

Conclusions

The characters, days to 1st flowering, number of bolls per plant, boll weight, seed index, seed cotton yield per plant, uniformity index, upper half mean length, fibre strength and micronaire value revealed predominance of non-additive gene action. Additive gene action was depicted by the characters, plant height and number of sympods per plant and ginning out turn. The characters, number of monopods per plant and lint index were governed by both additive and non-additive gene action. It may be concluded that H1471, H1472, H 1522, H1098i and C 201 were the best parents on the basis of their gca effects for yield and yield contributing traits as well as fiber quality traits. The crosses involving these parents would bring about superior hybrids. The cross combinations H 1480 \times C 201, H 1098i \times C 211 and H 1522 \times C 210 were the best based on their high sca effects for seed cotton yield per plant. Therefore, these cross combination may be utilized as hybrid cultivar after thorough evaluation over time and space.

Table 1. Details of experimental genotypes with yield performance.

No.	Females (Lines)	Seed Cotton Yield (kg/ha)		
		2016	2017	Mean
1	H1471	3534	1914	2724
2	H1472	3179	2083	2631
3	H1480	3272	2454	2863
4	H1488	3441	2222	2832
5	H1489	3595	2917	3256
6	H1508	3503	2346	2925
7	H1518	3025	2145	2585
8	H1519	2819	3302	3061
9	H1520	2767	2978	2873
10	H1522	3354	1960	2657
11	H1523	3519	3256	3388
12	H1098i	2932	2500	2716
Males (Testers)				
1	C-201	2787	-	2787
2	C-202	2316	-	2316
3	C-210	2187	-	2187
4	C-211	1952	-	1952

Table 2. Analysis of variance for Line x Tester for different characters in American cotton (*G. hirsutum* L.).

Source of Variation	Df	Days to 1 st Flower	Plant Height	Number of Monopods Per Plant	Number of Symopods Per Plant	Number of Bolls Per Plant	Boll Weight	Seed Index	Seed Cotton Yield	Ginning Out Turn	Lint Index	UI	Fibre Strength	Micronaire Value	UHML
Replication	2	6.05	500.69	2.30	446.02	72.44	0.32	0.05	131.58	15.44	0.88	0.002	0.30	0.14	0.03
Crosses	47	16.79**	203.43	0.39	10.07	42.91**	0.42	0.92**	1319.48**	20.09**	1.09**	0.62*	3.86**	0.19**	1.92
Line	11	19.54**	471.36**	0.41	19.93	88.76**	0.50*	1.85**	1175.59**	47.01*	1.39**	0.82	5.75	0.41	2.76
Tester	3	60.92**	607.94**	2.00**	11.41	26.62*	0.93**	1.04**	6376.16**	69.97*	5.66**	0.11	1.06	0.03	0.60
Line×tester	33	11.87	77.35	0.24	6.67	30.77**	0.34	0.60**	907.74**	6.58**	0.57*	0.60	3.48*	0.13	1.75
Error	94	4.06	87.99	0.31	6.83	6.78	0.16	0.09	167.55	2.75	0.15	0.39	1.65	0.13	1.39

* Significant 5 at p=0.05, ** Significant at=0.01, UI= Uniformity Index, UHML=Upper Half Mean Length

Table 3. Estimates of genetic components of variance for different characters in American cotton (*G. hirsutum* L.).

Variance	Days to 1 st Flower	Plant Height	Number of Monopods Per Plant	Number of Symopods Per Plant	Number of Bolls Per Plant	Boll Weight	Seed Index	Seed Cotton Yield	Ginning Out Turn	Lint Index	UI	Fibre Strength	Micronaire Value	UHML
COV(HS)	1.18	19.26	0.05	0.38	1.02	0.02	0.04	119.51	2.16	0.12	0.06	0.13	0.14	0.00
COV FS	4.97	34.98	0.15	0.70	10.03	0.09	0.24	485.74	5.60	0.38	0.37	1.32	1.77	0.10
σ^2 GCA	1.18	19.26	0.05	0.38	1.02	0.02	0.04	119.51	2.16	0.12	0.06	0.13	0.14	0.00
σ^2 SCA	2.60	-3.55	0.05	-0.05	8.00	0.06	0.17	246.73	1.28	0.14	0.25	1.07	1.48	0.11
σ^2 GCA/ σ^2 SCA	0.45	-5.43	1.04	-6.94	0.13	0.25	0.20	0.48	1.69	0.91	0.26	0.12	0.10	-0.02

* Significant 5 at p=0.05, ** Significant at=0.01, UI= Uniformity Index, UHML=Upper Half Mean Length

Table 4. Estimation of general combining ability effects of parents for different characters in American cotton (*G. hirsutum* L.).

No. Parents	Days to 1 st Flower	Plant Height	Number of Monopods/Plant	Number of Symopds/Plant	Number of Bolls/Plant	Boll Weight	Seed Index	Seed Cotton Yield/Plant	Ginning Out Turn	Lint Index	UI	UHML	Fibre Strength	Micronaire Value	
A. Females															
1	H1471	-2.76**	-2.57	-0.11	-1.35	-1.85*	-0.04	-0.05	12.42**	-0.19	-0.07	0.19	0.56	1.08**	-0.20*
2	H1472	-2.26	0.35	0.06	0.81	2.32**	0.14	0.34**	13.00**	-1.01**	0.08	-0.14	-0.19	-0.27	0.02
3	H1480	-0.01	9.76*	0.22	0.98	5.15**	0.06	0.56**	5.00	-3.17**	-0.28*	0.02	0.20	-0.20	-0.18
4	H1488	0.74	5.51	-0.19	0.56	2.15**	-0.06	0.18*	3.17	-2.56**	-0.43**	-0.23	-0.38	-0.27	0.06
5	H1489	-0.01	4.10	0.06	0.98	1.99**	0.09	-0.57**	0.17	-0.89	-0.62**	-0.31	-0.65	-0.98	-0.03
6	H1508	1.57**	-14.65**	-0.28	-0.85	-4.35**	-0.36**	-0.01	-13.58**	-0.77	-0.19	-0.07	-0.02	0.29	-0.14
7	H1518	0.49	3.82	0.06	2.31**	-0.18	0.26*	-0.25**	-8.42*	3.63**	0.62**	0.17	0.23	0.42	0.28**
8	H1519	-0.01	3.26	-0.03	0.65	-0.43	0.04	0.65**	-8.67*	-0.90	0.26*	0.47**	0.47	0.57	0.12
9	H1520	0.10	0.68	-0.19	-0.02	-2.18**	-0.01	0.11	-13.75**	1.14*	0.33*	0.37*	0.60	0.15	0.14
10	H1522	0.74	-4.15	0.31	-0.52	1.24	0.33**	-0.10	8.92**	0.73	0.09	-0.06	-0.52	-0.40	0.27**
11	H1523	0.90	-4.82	-0.11	-2.27*	-1.93**	-0.24*	-0.54**	-6.83*	2.37**	0.12	-0.39*	-0.71*	-1.22*	-0.01
12	H1098i	0.74	-1.32	0.22	-1.27	-1.93**	-0.24*	-0.33**	8.58**	1.62**	0.07	-0.01	0.40	0.83	-0.31*
SE (d)		0.59	3.30	0.17	0.88	0.74	0.12	0.08	3.43	0.51	0.11	0.18	0.34	0.38	0.09
CD 5%		1.07	6.46	0.33	1.73	1.46	0.24	0.15	6.72	0.99	0.22	0.36	0.68	0.75	0.18
CD 1%		1.41	7.44	0.44	2.28	1.92	0.31	0.20	8.85	1.31	0.29	0.47	0.89	1.00	0.24
B. Males															
1	C 201	-1.54**	-4.86	-0.22*	-0.83	-1.18**	0.21**	0.13**	12.08**	2.02**	0.57**	-0.01	0.02	-0.10	-0.16
2	C 202	-0.60	-5.63**	-0.06	0.12	-0.07	-0.15*	-0.05	-19.00**	-0.18	-0.09	0.07	0.12	0.15	0.04
3	C 210	1.24**	3.29	0.33**	0.40	0.49	-0.09	0.14**	3.00	-0.88*	-0.11	-0.07	-0.18	0.14	-0.00
4	C 211	0.90*	2.82	-0.06	0.31	0.76	0.03	-0.22**	3.92*	-0.96*	-0.36**	0.02	0.05	-0.19	-0.02
SE (d)		0.32	1.09	0.10	0.62	0.43	0.07	0.04	1.98	0.29	0.06	0.10	0.20	0.22	0.053
CD 5%		0.62	3.73	0.19	1.00	0.84	0.14	0.09	3.90	0.57	0.13	0.21	0.39	0.44	0.11
CD 1%		0.81	4.91	0.26	1.32	1.11	0.18	0.11	5.11	0.75	0.17	0.27	0.52	1.00	0.14

* Significant at p=0.05, ** Significant at p=0.01, UI= Uniformity Index, UHML = Upper Half Mean Length

Table 5. Estimation of specific combining ability effects of crosses for different characters in American cotton (*G. hirsutum* L.).

No.	Hybrids	Days to First Flower	Plant Height	Number of Monopods Per Plant	Number of Sym pods Per Plant	Number of Bolls Per Plant	Boll Weight	Seed Index	Seed Cotton Yield	Ginning Out Turn	Lint Index	UI	UHML	Fibre Strength	Micronaire Value
1	H1471×C201	-4.79**	1.82	-0.03	-0.92	-1.90	-0.13	0.41**	8.33	-0.45	0.22	-0.65	-1.10	-1.35	0.03
2	H1471×C202	1.93	-9.71	-0.53	-0.20	-1.35	-0.23	-0.37*	4.25	2.89**	0.31	0.26	0.73	0.60	-0.25
3	H1471×C210	1.43	-0.63	-0.08	-1.81	0.76	0.24	-0.10	-21.75**	-0.84	-0.24	0.39	0.63	0.54	-0.01
4	H1471×C211	1.43	8.51	0.47	2.94	2.49	0.12	0.06	8.67	-1.60	-0.29	-0.01	-0.26	0.21	0.23
5	H1472×C201	4.04**	-4.76	-0.19	-1.76	-5.74**	0.36	0.79**	9.92	1.94	1.07**	-0.32	-0.18	-0.14	0.08
6	H1472×C202	-3.57**	6.38	0.31	1.63	2.82	0.18	0.11	16.00*	-0.43	-0.04	-0.40	-0.81	-0.85	0.10
7	H1472×C210	1.60	-3.54	-0.42	0.02	0.93	-0.94**	-0.00	1.00	0.54	-0.02	0.07	0.16	-0.31	0.07
8	H1472×C211	-2.07	1.93	0.31	0.10	1.99	0.40	-0.84**	-26.92**	-2.05*	-1.00**	0.65	0.83	1.29	-0.25
9	H1480×C201	-1.21	2.82	-0.03	0.08	0.43	-0.23	-0.13	26.25**	0.59	-0.01	0.18	-0.07	0.66	0.11
10	H1480×C202	0.51	-1.38	0.14	-0.20	-0.35	0.27	0.12	-5.33	0.13	0.12	-0.23	0.67	0.02	0.02
11	H1480×C210	-0.32	-0.96	0.08	-0.15	0.43	0.011	-0.01	-6.33	-0.30	-0.07	0.23	-0.60	0.19	-0.21
12	H1480×C211	1.01	-0.49	-0.19	0.27	-0.51	-0.05	0.02	-14.58*	-0.42	-0.05	-0.18	0.14	-0.87	0.08
13	H1488×C201	1.71	2.74	0.06	0.16	-0.90	0.09	-0.45**	-16.58*	-1.95	-0.76*	-0.24	-0.02	-1.01	0.31
14	H1488×C202	-2.24*	-5.13	-0.11	-0.45	-1.35	-0.28	-0.10	-9.17	0.12	0.00	0.69	1.22	1.89*	-0.31
15	H1488×C210	0.60	-1.04	0.17	-0.73	-0.57	-0.07	0.31*	21.50**	0.96	0.42	0.15	-0.12	0.23	-0.01
16	H1488×C211	-0.07	3.43	-0.11	1.02	2.82	0.27	0.23	4.25	0.87	0.34	-0.60	-1.08	-1.11	0.01
17	H1489×C201	-0.21	-4.18	0.47	0.08	-1.07	0.07	0.13	2.08	-0.76	-0.13	0.51	0.58	1.01	0.10
18	H1489×C202	0.85	5.63	-0.03	-0.20	-1.51	-0.37	-0.18	10.17	1.04	0.06	0.44	0.85	1.07	-0.09
19	H1489×C210	-1.99	4.04	-0.42	1.52	3.93**	0.51*	0.03	-20.17**	0.35	0.14	-0.43	-0.58	-0.76	0.09
20	H1489×C211	1.35	-5.49	-0.03	-1.40	1.35	-0.22	0.02	7.92	-0.64	-0.07	-0.52	-0.85	-1.32	-0.10
21	H1508×C201	1.54	3.90	-0.19	0.58	0.26	0.26	-0.20	-11.83	-0.94	-0.37	-0.39	-0.49	-0.63	0.00
22	H1508×C202	0.26	-6.63	-0.03	-3.04	-5.18**	-0.12	0.05	-5.42	-1.07	-0.14	-0.34	-1.13	-1.22	-0.03
23	H1508×C210	-1.57	0.79	0.25	1.69	3.60*	-0.11	0.33*	17.58*	-1.10	-0.02	-0.08	0.08	-0.19	0.14
24	H1508×C211	-0.24	1.93	-0.03	-0.77	1.32	-0.03	-0.18	-0.33	3.11**	0.53*	0.81	1.54*	2.04**	-0.11
25	H1518×C201	-0.04	3.07	-0.19	1.41	2.76	0.04	0.35*	16.33*	-1.67	-0.07	0.57	0.47	1.14	-0.11
26	H1518×C202	-0.32	0.88	-0.36	-1.20	-1.68	0.40	0.03	19.75*	0.79	0.16	0.13	0.60	0.51	-0.00

Continuing Table 5

No.	Hybrids	Days to First Flower	Plant Height	Number of Monopods Per Plant	Number of Synpods Per Plant	Number of Bolls Per Plant	Boll Weight	Seed Index	Seed Cotton Yield	Ginning Out Turn	Lint Index	UI	UHML	Fibre Strength	Micronaire Value
27	H1518×C210	0.51	0.29	0.25	-0.48	1.76	-0.52*	-0.42*	-7.58	0.17	-0.29	-0.28	-0.25	-0.26	-0.06
28	H1518×C211	-0.15	-4.24	0.31	0.27	-2.85*	0.08	0.10	-28.50**	0.71	0.20	-0.42	-0.89	-1.40	0.17
29	H1519×C201	-0.21	-4.01	0.22	1.41	1.01	-0.21	-0.75**	-5.42	-0.47	-0.67**	0.06	0.45	0.16	0.07
30	H1519×C202	-2.15*	2.46	-0.28	-0.20	1.57	0.22	0.20	9.00	-0.47	0.05	0.32	0.31	0.29	0.10
31	H1519×C210	1.35	6.54	0.00	-0.48	-1.99	0.29	0.58**	-4.83	-0.30	0.37	-0.28	-0.72	-0.48	-0.06
32	H1519×C211	1.01	-4.99	0.06	-0.73	-0.60	-0.30	-0.03	0.75	1.25	0.25	-0.10	-0.04	0.03	-0.11
33	H1520×C201	-3.13**	-0.10	-0.28	0.41	-0.57	0.24	0.28	-5.33	-0.12	0.22	0.21	0.50	-0.04	-0.39*
34	H1520×C202	1.26	7.04	0.22	2.47	1.32	0.07	0.24	-8.25	-1.12	-0.06	-0.64	-0.76	-1.74*	0.23
35	H1520×C210	0.43	-4.88	0.17	-2.15	-2.90*	0.21	-0.46**	11.42	0.86	-0.14	-0.34	-0.79	-0.46	0.11
36	H1520×C211	1.43	-2.07	-0.11	-0.73	2.15*	-0.52**	-0.07	2.17	0.37	-0.02	0.78*	1.05	2.24**	0.04
37	H1522×C201	1.04	2.74	-0.11	-0.42	1.68	-0.03	0.17	10.00	1.49	0.49*	-0.17	-0.50	0.10	0.06
38	H1522×C202	1.10	-5.13	0.39	-0.70	-1.76	-0.13	-0.85**	-32.25**	0.23	-0.62**	-0.05	-0.31	-0.46	-0.04
39	H1522×C210	0.60	-2.04	-0.00	0.35	-2.99	0.08	0.40**	23.08**	0.13	0.34	0.12	0.49	0.29	-0.04
40	H1522×C211	-2.74*	4.43	-0.28	0.77	3.07**	0.08	0.28	-0.83	-1.85	-0.21	0.10	0.32	0.07	0.016
41	H1523×C201	0.21	6.60	-0.03	-1.34	1.51	-0.13	0.17	-17.25*	0.66	0.29	0.17	0.01	-0.53	-0.04
42	H1523×C202	1.60	8.21	0.14	1.72	5.07**	-0.30	0.01	1.83	0.99	0.19	-0.15	-0.02	-0.01	0.48*
43	H1523×C210	-1.90	0.63	0.08	2.10	1.85	0.11	-0.54**	-5.83	-1.43	-0.70**	0.18	0.22	0.43	-0.06
44	H1523×C211	0.10	-2.24	-0.19	-2.43	-8.43**	0.32	0.38*	21.25**	-0.22	0.22	-0.20	-0.22	0.11	-0.39*
45	H1098i×C201	1.04	2.57	0.31	0.33	2.51	-0.33	-0.77**	-17.00*	1.67	-0.29	0.07	0.35	0.62	-0.26
46	H1098i×C202	0.76	2.63	0.14	-0.38	2.40	0.30	0.81**	-0.58	-3.10**	-0.03	-0.03	-0.08	-0.09	-0.20
47	H1098i×C210	-0.74	0.79	-0.25	0.10	-4.82**	0.18	-0.05	-8.58	0.97	0.22	0.27	0.27	0.79	0.04
48	H1098i×C211	-1.07	0.74	-0.19	-0.81	-0.10	-0.15	-0.01	26.17**	0.46	0.10	-0.31	-0.55	-1.29	0.39*
SE _d		1.10	6.60	0.34	1.77	1.48	0.24	0.15	6.86	1.01	0.23	0.36	0.68	0.76	0.18
CD 5%		2.14	12.93	0.67	3.47	2.91	0.47	0.30	13.44	1.98	0.44	0.71	1.35	1.51	0.37
CD 1%		2.82	17.01	0.89	5.24	3.83	0.62	0.39	17.70	2.61	0.58	0.95	1.79	2.00	0.48

* Significant at p=0.05, ** Significant at p=0.01, UI= Uniformity Index, UHML = Upper Half Mean Length

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DUS Characterization of the Most Promising High Root Yielding Genotype HWS 8-18 of Ashwagandha (*Withania somnifera*)

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ABSTRACT

Present study on DUS characterization is about the most promising high root yielding genotype HWS 8-18 of Ashwagandha (*Withania somnifera*) was done during the late *Kharif*, 2018 and 2019 and established its distinctness among different genotypes. The genotype HWS 8-18 was tested for several morphological descriptors. The ashwagandha genotype HWS 8-18 has been found superior in dry root yield as compared to ASW-1 (best check) 10.78% at National level and 40.99% higher in the Haryana State. The mean seed yield of HWS 8-18 was 610.87 kg/ha compared to 551.43 kg/ha of ASW-1 (best check) recorded at National level while at Hisar the mean seed yield was found to be 767 kg/ha as against ASW-1 (544 kg/ha). The genotype HWS 8-18 has given numerically high seed yield (579.41 kg/ha) and at par with best check, AWS-1 (541.05 kg/ha). The morphological characters, lanceolate, smooth, medium dark green leaves, orange yellow berry colour and semi-erect are the marker characters of HWS 8-18. In addition to this, the characterization information will be helpful to maintain the genetic as well as proper identification. Moreover, high root yield and seed yield have greater importance and would be utilized in hybridization for further genetic improvement.

Keywords: Ashwagandha (*Withania somnifera*), characterization, diversity, DUS testing, descriptors

Introduction

Globally, Ashwagandha (*Withania somnifera* L.) Dunal (2n=48) is most demanding medicinal herbs and known for the medicinal utility of its root alkaloids as immunity booster against COVID. Now-a-days, due to spread of COVID pandemic all over the world, importance of this herb has been increased several folds. Ashwagandha is most valuable herbal plant for Indian culture because it is an important part of Ayurveda and Unani systems of medicine since ancient times (Koli et al. 2021). It is most interesting to note down that the cultivated plants have sizable differences in their therapeutic action. Ashwagandha belongs to the family Solanaceae and genus *Withania*. In Ashwagandha, only two species viz., *Withania somnifera* (L.) Dunal and *Withania coagulans* Dunal are found in India. In modern era, to protect a new variety under the Plant Varieties and Farmers Rights (PVP and FR Act in, 2001) DUS characterization of new variety is very important.

Under this act new variety is compared with other known variety on the basis of a set of appropriate traits and this information is required at the time of release of new variety (Yadav et al. 2013). Therefore, keeping the importance of above facts, the present study was undertaken to characterize the ashwagandha HWS 8-18 on the basis of quantitative and qualitative morphological characters and to identify distinctness among the newly developed genotypes.

Materials and Methods

In this experimental study, Ashwagandha (*Withania somnifera*) genotype HWS 8-18 was selected from germplasm of Medicinal and Aromatic plant section of Chaudhary Charan Singh Haryana Agricultural University, Hisar. The genotype was evaluated for morphological characters during late *Kharif* 2018 and 2019. For this study, five randomly selected plants of Ashwagandha were taken. Data was recorded on the

all the morphological characters. All the characters were observed at specified stage of crop growth, when the character under study had full expression. The characters, namely, growth habit, growth pattern, plant pigmentation, leaf texture, leaf color, leaf polishing, flower color, at time of flowering. At harvesting and threshing stage, observations on seed color, seed shape were recorded. For quantitative characters, days to 50% flowering and days to maturity were recorded as number of days from sowing to 50% plants initiated flowering or showed ripe berry at the relevant stage of genotype, respectively. The data on remaining characters i.e. plant height (cm), number of branches per plant, number of berries per plant, berries yield per plant (g), number of seeds per berries, seed yield per plant (g) and biological yield per plant (g) were recorded at the time of harvesting and threshing. The mean values of three replications were calculated and classified into different groups according to germplasm catalog developed by International Board for Plant Genetic Resources, Rome, Italy.

For molecular divergence study, genomic DNA of Ashwagandha was extracted from young leaves using CTAB (Cetyltrimethyl Ammonium Bromide) method with modifications. The quality of DNA was checked by using 0.8% agarose gel in electrophoresis (Parita et al. 2018). A set of three SSR markers was used to study the genetic divergence in ashwagandha genotypes at the molecular level. SSR is a co-dominant marker and it is distributed through the genome, variable, easily scorable, highly reproducible, multiallelic, abundant and in nature that's why it is a good choice for molecular characterization. It has been used in many researches those are based on diversity. Less numbers of SSR markers can be provide better genetic diversity spectrum because of its multi allelic and high polymorphic nature (Shah et al. 2013). All three primers used in this study possessed good transferability in *Withania somnifera* and hence, they were used in present study for diversity analysis. The three primers used in this study are (i) CAMS 340, (ii) CAMS 351 and (iii) CAMS 376 (Table 1).

Results and Discussion

Agro-morphological characters

In the present study, ashwagandha genotype HWS 8-18 was evaluated for its morphological traits. Plant height was one of the most variable characters in ashwagandha. On the basis of this character, genotype, HWS 8-18 was grouped into the intermediate category having about 63.99 cm tall. In ashwagandha, three types of growth habits are found i.e. erect, semi-erect and spreading type. Out of these, the genotype,

HWS 8-18 exhibited semi-erect growth habit. On the basis of number of branches per plant genotype was also grouped into the intermediate. The shape of leaves of genotype HWS 8-18 was lanceolate type, and color of foliage leaves was dark green (Table 2). The dark green color of leaves is responsible for harnessing maximum sum light for photosynthesis as compared to light green and pale green. The fruits of ashwagandha are known as berry; generally three types of berry color are reported in literature red, orange and light green. The genotype HWS 8-18 have orange yellow berry colour (Fig. 1). Its plant starts flowering in about 82-93 days. The variety HWS 8-18 takes about 163-171 days to mature and falls in Medium maturity group. Similar findings were also reported by Yadav et al. (2013) in Indian mustard.

Yield and its component characters

The ashwagandha genotype HWS 8-18 has been found superior in dry root yield compared to ASW-1 (best check) 10.78% at National level and 40.99% higher in the Haryana State (Table 3). The mean root yield of HWS 8-18 was 610.87 kg/ha compared to 551.43 kg/ha of ASW-1 (best check) recorded at National level. The genotype HWS 8-18 has given numerically high seed yield (579.41 kg/ha) and at par with best check, AWS-1 (541.05 kg/ha). Further, this DUS Study can be play an important role in maintain of genetic purity of a genotypes as well as in characterization of genotype. Above findings were also supported by Kumar et al. (2007), Joshi et al. (2015), Srivastava et al. (2018) and Shahaji et al. (2020).

DUS characterization of new variety is very important plant breeding program in the era of IPR (Intellectual Property Rights). It provides guarantee of distinct, uniform and stable which has been specified by the breeder. It is very important component of Plant Varieties and Farmers' Rights Act (2001) because it provides basic information of variety which helps in protection (Yadav et al. 2013).

Molecular characterization

With help of SSR markers, the promising genotype HWS 8-18 was also compared with other varieties/genotypes. It was found unique and different from all other genotypes/varieties. The detailed results on gene distribution among the genotypes/varieties are presented in Table 4. The genotype HWS 8-18 is differentiated on the basis of presence of CAMS 34 primer 50kb band which was absent in HWS-205, HWS-205 and HWS 12-12. In addition to this, it was differentiated from HWS 1203 and JA-20 due to absent of CAMS 34 primer 400kb band, as this was present in HWS 8-18. Parita et al. (2018) also worked on Ashwagandha by using SSR makers.

Conclusions

On the bases of above study on ashwagandha genotype, it can be concluded that HWS 8-18 has been found superior in dry root yield compared to ASW-1 (best check) 10.78% at National level and 40.99% higher in the Haryana State. The mean seed yield of HWS 8-18 was 610.87 kg/ha compared to 551.43 kg/ha of ASW-1 (best check) recorded at National level while at Hisar the mean seed yield was

found to be 767 kg/ha as against ASW-1 (544 kg/ha). The genotype HWS 8-18 has given numerically high seed yield (579.41 kg/ha) and at par with best check, AWS-1 (541.05 kg/ha). Lanceolate, smooth, medium dark green leaves, orange yellow berry colour and semi-erect are the marker characters for the HWS 8-18 genotype. On the bases of morphological traits and molecular characterization it can be concluded that HWS 8-18 is superior genotype.

Table 1. List of SSR primers used in the present study.

No	Primer Name	Primer Sequence	% GC Content	Tm (°C)
1	CAMS-340	F: TTTATGCCCATTCACAAAATAA R: GGACGAATTCACCGAGTGC	41.00	66.0
2	CAMS-351	F: CGCATGAAGCAAATGTACCA R: ACCTGCAGTTTGTGTTGGA	45.00	50.0
3	CAMS-376	F: GGTGCTGGCATAGATGAACA R: TATGTCTGGCTTGGTGCTGA	50.00	69.0

F: Forward primer; R: Reverse primer; Tm: Melting temperature

Table 2. Plant description of HWS 8-18 variety of Ashwagandha.

Morphological DUS Characters of HWS 8-18	Lenceolate, smooth, dark green leaves, yellow-orange berry colour and semi-erect plant type
Mean dry root yield (kg/ha)	610.87
Mean seed yield (kg/ha)	579.41
Mean plant height (cm)	63.99
Days to Flowering	87.56
Maturity days	167.45
Root length (cm)	20.59
Root diameter at collar region (mm)	11.89
Number of root branches/plant	2.54
Dry plant yield (kg/ha)	3265.54
Number of stem branches/plant	3.80
Withaferin-A (%)	0.42
Withanolide-A (%)	0.59
12-deoxy-withastramanolide (%)	0.27

Table 3. Dry root yield data of coordinated varietal trials.

Particulars	Year of Testing	No. of Trials	HWS 8-18	HWS 12-12	Check Variety AWS-1	Check Variety JA 20
Mean dry root yield (kg/ha)	2015-16	7	647.29	620.08	637.93	500.02
	2016-17	9	606.78	565.00	525.44	481.33
	2017-18	9	578.55	590.57	490.91	441.92
	Mean		610.87	591.88	551.43	474.42
Percent increase over best check	2015-16		1.47	-2.80		
	2016-17		15.48	7.53		
	2017-18		17.85	20.30		
	Mean		10.78%	7.34%		

Table 4. Profile of the gene distribution in entries and checks using SSR.

No	Genotype\Marker	CAMS 340			CAMS 351			CAMS 376				
		Allele Size	50kb	200kb	400kb	50kb	130 kb	420kb	130kb	170kb	200kb	350kb
1	HWS 8-18		+	+	+	-	-	+	-	-	-	-
2	HWS-205		-	+	+	-	-	+	-	+	-	-
3	HWS-222		-	+	+	-	-	+	-	+	-	-
4	HWS 1203		+	-	-	-	-	-	+	-	-	-
5	HWS 12-12		-	+	+	-	-	+	-	+	+	+
6	JA - 20		+	-	-	+	+	-	-	+	+	+

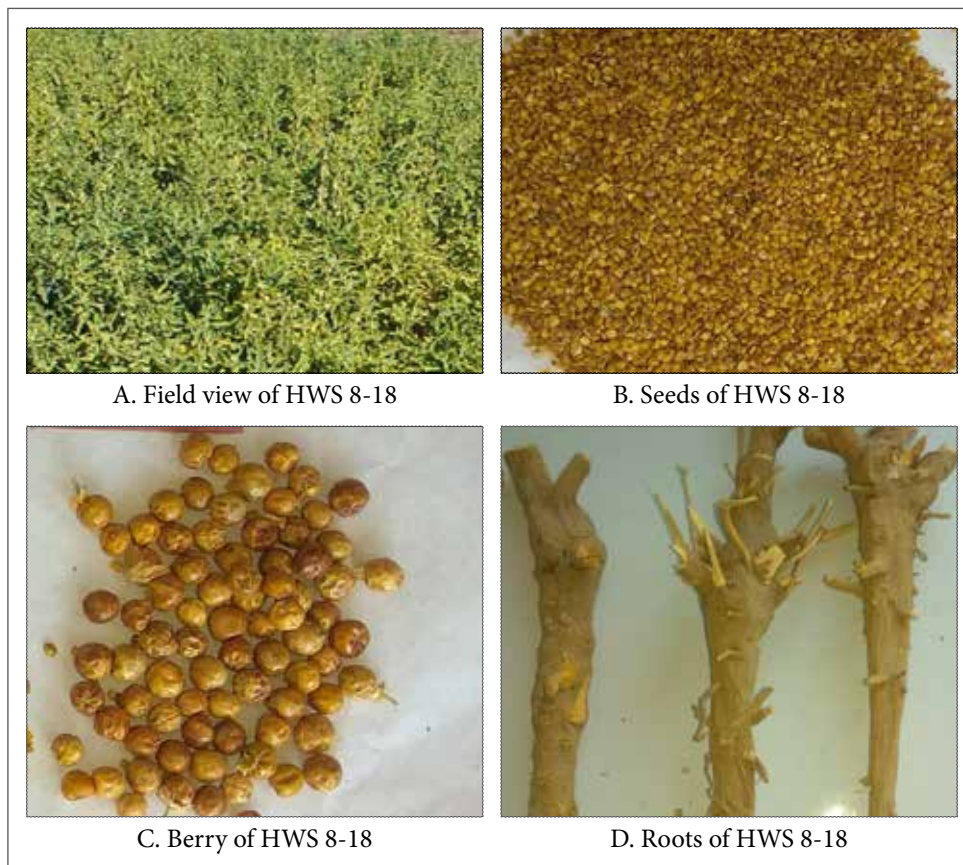


Figure 1. Ashwagandha HWS 8-18 (A) Field view (B) Seed (C) Berry and (D) Roots. (Original)

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Registration of "Albaşak" Bread Wheat (*Triticum aestivum* L.) Variety

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Albaşak is winter bread wheat (*Triticum aestivum* L.) variety developed by Trakya Agricultural Research Institute (TARI) and registered in 2021 (Figure 1). Albaşak is developed by crossing Tek/4/Lau/Agd/3/Odes95//Olv/B16 with TE6474-0T-0T-4T-3T-4T-0T through pedigree method. Crossing was made in 2004 and yield test were began in 2011-2012 growing year.

The spike of the Albaşak cultivar is moderately long, red, smooth, with awn and medium compact. The first red and awned variety registered in the Trakia region. The flag leaf is dark-green and with medium-low glaucosity. Grain is oval, hard and red colour. Albaşak is a medium-tall cultivar, similar to Köprü, Gelibolu, Yüksel and Saban. Plant height is between 93 and 103 cm depending on the growing conditions. It is medium early and as it has good adaptation ability, it has been grown throughout the Trakya-Marmara region and some other transitional-zone parts of Turkey. It gives high yield both on fertile and less fertile soils. It has resistance to winterkilling and is tolerant to medium drought conditions. Albaşak is tolerant to powdery mildew (*Erysiphe graminis* f. sp. *tritici*), and highly tolerant to stripe rust (*Puccinia striiformis* f. sp. *tritici*) and leaf rust (*Puccinia triticina*). It has susceptible to septoria leaf disease.

Its yield potential is high however, high yield can be obtained if environmental conditions are favorable and applied good agronomic practices. The highest grain yield obtained was 9874 kg ha⁻¹ in variety testing experiment. Mean yield of the variety testing experiment was 7924 kg ha⁻¹ in Trakya growing conditions. Suggested planting rate is between 450-500 seeds/m².

Its grain quality is good. The mean values of some bread-making qualities of the variety testing experiment (2019 and 2020) are; test weight 71.7-74.6 kg, thousand kernel weight 27.4-39.5 g, protein content 14.0-16.5%, absorption 55.6-61.6%, sedimentation (Zel) 45-72 ml, gluten index 97.3-99.5%, gluten value 27.3-32.1%, alveograph energy value (W) 237-429. The highest quality values in 2017-2018 growing seasons application of the variety testing experiment were; thousand kernel weight 48.7 g, test weight 80.2 kg, protein content 12.0%, gluten value 32.3%, gluten index 98.2% and sedimentation (Zel) 66 ml.

Pre-Basic and Basic seeds of the Albaşak cultivar have been produced by Trakya Agricultural Research Institute (TARI). Certified seed of the Albaşak are produced by both private companies and state farms.



Figure 1. Spike and grain of the cultivar Albaşak. (Original)

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Registration of "Vehbibey" Durum Wheat (*Triticum durum* Desf.) Variety

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Vehbibey is a winter durum wheat (*Triticum durum* Desf.) cultivar, released in 2019 by Central Research Institute for Field Crops (Figure 1). The cultivar is characterized by high grain yield, tall plant height, resistance to drought, cold, lodging, and is mildly resistant to yellow rust. Its spikes are white and grains are highly vitreous when sufficient fertilizers are applied.

The cultivar has high water and fertilizer use efficiency. It is adapted to Central Anatolia Regions' climate which receives low annual precipitation (300-350 mm). Vehbibey's pedigree is Kunduru414-44/66T11//ANK-98 and YA:08838-OA-OA-OA-13A-0A. The cross was made in 2006 and yield trials began in the 2013-14 growing season. Application for registration of Vehbibey durum wheat variety was submitted to the Seed Registration and Certification Center in 2016.

The average yield of the cultivar ranged from 3000-4000 to 4500-7000 kg/ha⁻¹ under rainfed and irrigated conditions, respectively. Vehbibey was the top-yielding cultivar in yield trials during the registration process.

The cultivar has very good quality characteristics such as thousand-kernel weight: 33.6-46.5 g, hectoliter weight: 70.4-80.4 kg/hl, protein content: 13.0-16.4%, kernel vitreousness: 84-100%, SDS sedimentation: 30-49, B color: 24.9-26.4, and semolina yield: 60.4-67%. Vehbibey has both better quality characteristics as well as higher yield than Kızıltan-91 which is one of the most widely grown durum wheat cultivars in semi-arid areas in Central Anatolia.

The cultivar is recommended for semi-arid and irrigated areas of the Central Anatolian Region and the Transitional Zones. The appropriate sowing rate is 200-220 kg/ha and suggested sowing dates are between October 15-November 15.



Figure 1. Spike, grain and field view of the cultivar Vehbibey. (Original)

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