

## DEVELOPMENT OF CHICKPEA BLIGHT RESISTANT VARIETY (*DASHT*) USING COMBINATION OF BULK POPULATION AND PEDIGREE BREEDING METHOD

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### Abstract

“*Dasht*” is a blight resistant variety of Desi chickpea with intermediate growth habit. It was released in the year 2002 for Potowar and rice based areas. *Dasht* originated from a cross between C 44 and ICC 7770. C 44 is a local genotype well adapted to chickpea growing area of Punjab, whereas ICC 7770 is an *Ascochyta* blight resistant line obtained from ICRISAT, India. F<sub>1</sub> to F<sub>6</sub>, progenies of this cross were advanced by modified bulk method, negative selection/eradication of undesirable plants. Single plant selection was made from F<sub>6</sub> generation and subsequently, their progenies were evaluated, using an approach based on bulk population and pedigree breeding methods. Resistance against *Ascochyta* blight (blight score 3 – 5) and high yield potential (2400 kg ha<sup>-1</sup>) are the major attributes of *Dasht* that make it a superior variety for its target regions. *Dasht* is more cold tolerant and more efficient in iron uptake than C 44 and CM 98. The hundred seed weight of this variety is 24 gm and seeds are round in shape having 23% protein, 50% carbohydrates, 5.5% fats and 9.5% crude fiber. Plant type of *Dasht* is semi erect with plant height 50 – 80cm and 1.5-2.7 inter-nodal lengths. Pods are ovate with little depression on dorsal side and contain 1 – 2 seeds. Number of leaflets vary from 13–19, flowers are pink with 15–18 mm long pedicel and 2–6 mm long bracts and 7–9 mm long keel. The cooking time of *Dasht* is 8 minutes less than those of C 44 and Paidar 91. *Dasht* performs better in Potowar when planted in 2<sup>nd</sup> fortnight of October, keeping 20–25 kg acre<sup>-1</sup> seed rate and 30 cm row spacing.

### Introduction

Chickpea is predominantly grown under rain-fed conditions in Pakistan. The total area under chickpea in Pakistan is about one million hectares and 75% of that is located in Punjab. Potowar used to be one of the major chickpea growing areas before partition. The area of chickpea in Potowar before 1950 was 54,000 ha which accounted for 5.3% of its total area and contributed 7% to the total production (Anon., 1948). The epidemics of blight in the following years resulted in gradual decrease of area in this region. Presently, Potowar region contributes only 0.105% and 0.8% respectively to the total area and production. Since blight is a serious disease of chickpea, it causes severe yield losses in epidemic years. The low seed yield is also attributed to non-availability of pure seed of improved varieties alongwith low inputs and poor management practices (Sarwar *et al.*, 1993). The yield per unit area may be increased substantially through the evolution of high yielding genotypes, bearing a good combination of all yield components (Sarwar & Ahmad, 2003). According to Malik & Bashir (1984) and Nene (1980) *Ascochyta* blight may cause 48% to 70% grain yield losses. Sattar *et al.*, (1953) reported that Pakistan suffered losses worth of Rs. 12 million annually in the decade of fifties due to *Ascochyta*

blight. The repeated occurrence of disease epidemics in Potowar resulted in gradual decline of its area and production. Ultimately, the contribution of Potowar to production came down from 7% to 0.7% and area decrease from 54,000 to 10,000 ha. The environmental conditions of Potowar area are conducive for chickpea production except for *Ascochyta* blight disease that limits its cultivation in this area. This potential can only be realized by solving the problem of blight through the development of a resistant variety.

Incorporation of resistance against the disease in the host plant has been proposed as the most economical, efficient mean of disease control and could be utilized in the hybridization programme for the development of chickpea resistant cultivars for commercial cultivation (Iqbal, 2002, Hussain, 2002). Therefore, continuous efforts are made across the world to develop blight resistant varieties. Consequently, many blight resistant varieties have been released in various countries such as in USA, (Muehlbauer, 2004); India (Zope *et al.*, 2002; Singh *et al.*, 2002; Sandhu *et al.*, 2002). Blight resistance in chickpea is not a simple trait. It is controlled by many genes acting in additive manner (Malik, 1990). A combination of bulk and pedigree method was used to breed the blight resistant variety "*Dasht*". Selection of disease free single plants was delayed to F<sub>6</sub> generation (Pawar *et al.*, 1985 & Luedders *et al.*, 1973) and their progenies were evaluated as proposed by Allard (1966) till the adaptability studies.

The relationship between yield traits remains stable in late generation of legumes, which are normally different from those of early generation (Pushpendra & Ram, 1988). Therefore, delayed selection of single plants was opted for stability in the performance of progenies thus increasing the probability of better genotypes at the stage of homozygosity. The loss of additive genetic variability that is associated with bulk population breeding method during the course of generation advancement (Muehlbauer *et al.*, 1981) was reduced by the use of combination of bulk population and pedigree breeding methods.

### **Materials and Methods**

*Dasht* was developed from a cross between two genotypes ICC 7770, (tolerant to *Ascochyta* blight) and C 44 (a locally adapted high yielding variety). The hybridization was performed during 1983–84 and modified bulk selection method was used to develop this variety. The hybrid population was evaluated for target traits and advanced to F<sub>6</sub> generation before making single plant selections. Two generations were obtained in a year by exploiting summer environment of Himalayan Agricultural Research Station (HARS), Kaghan. Single plant selections were made from F<sub>6</sub> generations and their progenies were evaluated for yield potential and disease reaction under artificial infection conditions. High yielding progenies with blight resistance were selected in F<sub>8</sub> generation for evaluation in replicated trials. The selected lines were studied in replicated yield trials for grain yield evaluation and disease reaction. These trials were conducted at National Agricultural Research Center (NARC), Islamabad. Finally, the selected lines were evaluated in multi-location and national uniform yield trials. *Dasht* was also evaluated on farm yield trials before its recommendation for general cultivation. Various steps involved in the development of *Dasht* are given in Table 1.

Table 1. Various steps involved in the Development of <i>Dasht</i> .		
Study year	No. of filial generation	Remarks
1983-84	Parental genotypes were inter-crossed at NARC.	F <sub>1</sub> hybrid seeds were harvested for plantation at HARS, Kaghan
May 1984	F <sub>1</sub> population was raised at HARS, Kaghan	F <sub>2</sub> seeds from this population are bulk harvested
1984-85	F <sub>2</sub> population was raised at NARC	F <sub>3</sub> seeds from F <sub>2</sub> plant population were bulk harvested.
May 1985	F <sub>3</sub> plant population was grown at HARS, Kaghan.	Blight free single plants were selected from this population and their seed was bulked.
May 1985-86	F <sub>4</sub> bulk population was grown at NARC, Islamabad and was subjected to disease pressure.	The blight free single plants were selected and their seeds were bulked.
May 1986	F <sub>5</sub> bulk population was grown at HARS, Kaghan.	Best progenies were selected for yield screening.
1986-87	Bulk F <sub>6</sub> population was space planted at NARC, Islamabad for initial observation.	Blight resistant single plants were selected for yield evaluation.
1987-88	Yield screening nursery plant progeny rows in F <sub>7</sub> generation were planted at NARC, Islamabad.	The proposed line was high yielding and blight resistant, hence was selected for preliminary yield trial.
1988-89	F <sub>8</sub> generation was included in preliminary yield trial.	It was forwarded to advance yield trial after confirmation of yield superiority.

**Hybridization:** Crossing between parental lines was undertaken early in the morning at the experimental field of pulses programme, National Agricultural Research Center, Islamabad. For this purpose the parental lines (male and female) were sown alternatively, in Octobers (1983–1984), keeping line to line distance at 60 cm and plant to plant distance at 15 cm. At the time of flowering the pollination of female flowers was done immediately after emasculation. The emasculation and pollination techniques as described by Singh 1987 were followed. The F<sub>1</sub> seeds harvested from female parent were advanced to F<sub>2</sub> population by space planting.

**Generation advancement (F<sub>1</sub>-F<sub>6</sub>):** A single row of 4 m length was planted with F<sub>1</sub> hybrid seed. The seeds were planted by dibbling maintaining 20 cm plant to plant distance at Himalayan Agricultural Research Station, Kaghan, under irrigated conditions. However, irrigation was not applied due to good precipitation received during summer 1984. The F<sub>2</sub> seeds were harvested in the end of October and planted at NARC experimental field. Planting was done in 60 cm wide rows with 15 cm plant to plant distance. This bulk planting was continued upto F<sub>6</sub> generation with negative selection of undesirable diseased plants under natural infection conditions. The seed from selected plants were bulk harvested. Single plant selection was made in F<sub>6</sub> generation.

**Planting of yield trials:** *Dasht* was consecutively evaluated for three year in on-station yield evaluation trials. Planting of on-station trials was done in the first week of November. The testing was done in Preliminary, Major and Multilocational Yield Trials before testing in National Uniform Yield Trials. All the yield trials were laid out in RCBD with 4 replications. The row to row and plant to plant distance was maintained at 30 cm and 10 cm, respectively. Planting was done with single row drill in 4 row plots of 4 m length. Plant population was maintained by thinning at seedling stage. One to two standard checks were included in every experiment for comparison. Disease reaction was separately recorded under artificial inoculation conditions.

**Screening against *Ascochyta* blight:** The different genotypes of chickpea and two standard check varieties (CM 72 and C 44) and a susceptible check were planted at NARC Islamabad in early October in Augmented design with one replication. During winter monsoon, i.e., in the month of February and March, the crop was sprayed with the inoculum of *Ascochyta rabiei* and humidity in the experiment was maintained with water spray. As NARC Islamabad is a hot spot of chickpea blight, severe blight incidence is obtained naturally as well. *Ascochyta* blight scoring of the test entries was done when susceptible check was completely killed. Blight scoring was done on 1–9 scale.

**Testing of *Dasht* in national uniform yield trials (NUYT):** *Dasht* was consecutively tested in NUYT trials for three years. This trial was conducted at seven locations in 1991-92 (Table 5); at 7 locations in 1992-93 and at 8 locations in 1993-94. At all the locations, the experiment was planted in RCBD with 4 replications. Six rows plots, where R x R distance was 30 cm and plant to plant distance was 10 cm, were planted with each entry. The sowing time and crop husbandry practices were different at all the locations. The replicated data of individual locations were averaged and converted to Kg ha<sup>-1</sup> for comparison.

Table 2. Grain yield of *Dasht* and checks varieties in preliminary, major and adaptation yield trials from 1988 – 89 to 1993 – 94.

Study year	Trial name	Grain yield (Kg ha <sup>-1</sup> ) of checks with <i>Dasht</i>				Remarks
		<i>Dasht</i>	CM 72	C 44	Punjab 91	
1988-89	Preliminary yield trial A-89	2036	1845	2240	-	In disease free years the performance of check variety, C 44 was better than that of <i>Dasht</i> .
1989-90	Major yield trial B-90	1074	-	-	-	The check varieties, CM 72 and C 44 were killed by blight at NARC, Islamabad
1990-91	Major yield trial C-1	2063	-	-	-	Both the check varieties were killed by blight at NARC.
1993-94	Adaptation trial	2433	-	-	2480	<i>Dasht</i> and Punjab 91 had non significant difference for grain yield under blight free condition

**Table 3. Summary of national uniform yield trials (Yield = Kg ha<sup>-1</sup>).**

Year	CM 72	C 44	Paidar 91	Punjab 91	Av. Yield of <i>Dasht</i>
1991-92	1643	1660			1582
1992-93	-	-	1715	1714	1973
1993-94	-	-	1393	1416	1332
Mean	1643	1660	1554	1565	1629

**Table 4. Grain yield (Kg ha<sup>-1</sup>) of *Dasht* as compared to other candidate varieties tested at different locations in national uniform yield trial 1992-93.**

Locations	Paidar 91	Punjab 91	<i>Dasht</i>
NARC, Islamabad	2280	1354	2148
AARI, Faisalabad	2470	2680	2475
BARI, Chakwal	2180	1914	2132
NIAB, Faisalabad	1608	1798	1950
ARS, Sarainaurang	1114	802	1778
NIFA, Peshawar	787	1845	1740
Seri, Potowar	1566	1611	1590
Mean	1715	1714	1973

**Table 5. Grain yield (Kg ha<sup>-1</sup>) of *Dasht* as compared to other candidate varieties tested at different locations in national uniform yield trial 1991- 92.**

Locations	C 44	CM 72	<i>Dasht</i>
NARC, Islamabad	2220	2759	2718
AZRI, Bahawalpur	780	1300	754
NIAB, Faisalabad	2598	1904	2161
UAF, Faisalabad	681	1024	936
NIFA, Peshawar	1226	1880	2246
BARI, Chakwal	1149	1398	936
AARI, Faisalabad	3339	2250	2500
ARS, Kalurkot	2474	1588	1536
RRI, Dokri, Sindh	468	590	541
Mean	1660	1643	1582

**Table 6. Grain yield (Kg ha<sup>-1</sup>) of *Dasht* as compared to other candidate varieties tested at different locations in national uniform yield trial 1993-94.**

Locations	Paidar 91	Punjab 91	<i>Dasht</i>
NARC, Islamabad	1524	1872	2058
AARI, Faisalabad	1600	744	1133
RRI, Dokri, Sindh	1274	1064	1065
UAF, Faisalabad	1136	1407	1032
BARI, Chakwal	937	1066	678
AZRI, DIK	1272	1352	1435
NIFA, Peshawar	1668	1800	1633
NIAB, Faisalabad	1734	2022	1626
Mean	1393	1416	1332

NARC- National Agricultural Research Center, Islamabad; AARI- Ayub Agricultural Research Institute, Faisalabad; RRI- Rice Research Institute, Dokri, Sindh; UAF- University of Agriculture Faisalabad; AZRI- Arid-zone Research Institute, Dera Ismail Khan; AZRI- Arid-zone Research Institute, Bahawalpur; BARI- Barani Agricultural Research Institute, Chakwal; NIAB- Nuclear Institute for Agriculture & Biology; ARS- Agricultural Research Station Sarainaurang; ARS- Agricultural Research Station, Kalurkot; NIFA- Nuclear Institute for Food & Agriculture; Seri- A farmer field in Potowar region.

**Physio-chemical properties:** Protein contents of flour samples were determined by micro Kjeldhal procedure (AACC 1969). After determination of Nitrogen contents total protein was calculated by converting Nitrogen values into protein values by multiplying these with 6.25 (Anon., 1969) by approved method of the AACC, method No 44-15A, American Assoc. Cereal Chem. St Paul, MN. For cooking time 30 gm of air dried unsoaked seeds were boiled to softness under reflux in tap water. Temperature of the boiling tap water was measured at 99.5°C. Twenty minutes after boiling, five seeds were removed and processed for softness and consistency. This was repeated after two minutes interval until seeds became soft and time required was noted. Crude fiber and ash were determined by AACC procedures (Anon., 1975). Total starch was analysed by using a procedure based on total enzyme digestion to glucose (Carpita & Kanabus 1987).

### Results and Discussion

**On station yield trials:** The major objective of development of *Dasht* was to give the farmers of Potowar a high yielding and blight resistant variety of Desi chickpea. Therefore, the blight resistant exotic line was hybridized with a well adopted local variety. The segregating population was advanced following a modified bulk method of plant breeding. Following this method only blight resistant and high yielding plants were selected and their seeds were bulked till F<sub>6</sub> generation. The superior single plants in F<sub>6</sub> generation were grown to plant progenies rows and evaluated on row basis which provided the base for selection of homozygous genotypes. The lines/genotype released as *Dasht* proved to be high yielding and blight resistant. This line was then evaluated in a series of replicated trials before putting it into a National testing system that is mandatory for the release of a variety in Pakistan. Beside yield evaluation the disease reaction and quality parameters of the candidate variety were also studied.

*Dasht* was studied for grain yield in preliminary and major yield trials from 1988-89 to 1990-91 and in adaptation yield trial during 1993-94. The data on grain yield recorded in these trials are presented in Table 2. These data showed that grain yield of *Dasht* ranged from 1047 to 2430 kg ha<sup>-1</sup> as compared to CM 72 and C 44 for which yield ranged from 0–1845 kg ha<sup>-1</sup>, respectively. Whereas, its yield was little less as compared to that of checks under blight free conditions. However, this difference was non-significant. Under high disease pressure at NARC the check varieties were completely killed, whereas *Dasht* gave 1074, 2063 kg/ha grain yield during 1989-90 and 2063 kg/ha during 1990-91, in yield evaluation trials (Table 2). Since Potowar is considered a hot spot for blight where severe disease occurs almost every year, therefore only blight resistant genotypes can do better in this region. The released varieties which were already available in the market, although possessed relatively high yield potential but they appeared to be susceptible to blight as revealed in the results reported here and also reported by Iqbal *et al.*, (2004). Consequently, they could not survive in Potowar due to high disease prevalence and incidence. Since the major emphasis was placed on incorporation of blight resistance in the target variety aimed for release in Potowar, *Dasht* appeared to be a promising line with respect to this major trait wanted for chickpea cultivation in blight prone areas. Consequently, *Dasht* was forwarded to national testing system and for the study of seed quality parameters.

**Table 7. Evaluation of *Dasht* for blight resistance on 1–9 scale.**

Year of study	Blight score of <i>Dasht</i>	Blight score of CM 72	Blight score of C 44
1989-90	4	9	9
1991-92	5	9	9
1992-93	3	5	5
1993-94	3	7	7
1994-95	4	8	8

Blight scale 1-4= Highly resistant; 5-6= Tolerant; 7-9= Susceptible

**National uniform yield trials:** *Dasht* was tested in national testing system through National Uniform Yield Trial (NUYT) consecutively for three years across the country. The location wise comparison of *Dasht* with check varieties for grain yield is given in Table 4, 5 and 6. Table 5 shows that average grain yield of *Dasht* across the locations in first and second year of testing (1991-92 and 1993-1994) was slightly less than those of checks whereas in the second year (1992-93), its grain yield was higher than those of both checks Punjab 91 and Paidar 91. The mean of three years showed that *Dasht* had better or almost similar yield to those of checks (Table 3). During 1991-92 two checks, C44 and CM 72 were used whereas in the following years Paidar 91 and Punjab 91 released as blight tolerant varieties were used as check for comparison. The three years evaluation of *Dasht* over multiple locations confirmed the results of on-station studies where it was concluded that *Dasht* having similar grain yield to already released varieties possesses high tolerance against *Ascochyta* blight. It was also observed that with overall good performance it was well adapted to Potowar conditions and in Peshawar valley (Table 4, 5 and 6).

**Blight reaction:** The disease score of *Dasht* and two check varieties recorded from 1989-90 to 1994-95 is presented in Table 7. The comparison of *Dasht* with released varieties showed that the blight score of *Dasht* varied from 3–5 as compared to 5–9 recorded for CM 72 and C 44. This shows the resistant behavior of *Dasht* under high disease pressure. Resistance against *Ascochyta* blight makes *Dasht* a better option for blight prone areas. On the basis of data recorded in various trials it was concluded that under disease free conditions *Dasht*'s performance was equally good as those of check varieties whereas its performance was much better with regard to grain yield than those of checks under severe blight incidence (Table 4 & 7). *Ascochyta* blight of chickpea, beside yield reduction, damages the quality of grain as well. Therefore, the grain obtained from susceptible varieties grown under diseased conditions are of inferior quality, whereas, the resistant varieties produces better yield and grain of better quality. This phenomenon was recorded in the case of *Dasht* as well when grown in disease condition. As blight incidence is a common phenomenon in Potowar, *Dasht* was released for this area. The empirical observation on *Dasht* showed that it is a better variety with respect to cold tolerance as compared to other varieties. The chickpea varieties DDG4, BG 1053 were released in India on the basis of their superiority in blight resistance (Sarvjeet *et al.*, 2002; Sandhu *et al.*, 2002).

**Characteristics of *Dasht* variety:** *Dasht* has resistance against *Ascochyta* blight, its foliar parts are pubescent, and pedicle has strong anthocyanin pigmentation, fresh seed colour is brown, secondary branches are terminal. The level of cold tolerance in *Dasht* is significantly higher than that of C 44 and Paidar 91. This variety is more efficient in iron uptake and shows no iron deficiency symptom in iron deficient soils where C 44 and Punjab 91 suffer from iron chlorosis.



Table 8. Physico-chemical characters and chemical composition of seed of *Dasht*.

Genotype	Physico-chemical characters						Chemical composition				
	100 seed weight (gm)	Hydration Capacity (g/seed)	Cooking time in min (dry)	Cooking time in min (soaked in water)	Protein % (N x 6.25)	Fat %	Carbo-hydrate (%)	Crude fiber	Ash	Energy (Kcal/100gm)	
Punjab91	26	0.27	122	32	23.1	4.7	50.9	10.2	2.9	338	
Paidar-91	21.5	0.17	123	36	22.4	4.7	50.6	11.6	2.9	334	
C-44	26	0.22	132	45	22.7	5.4	49.4	10.4	3.0	337	
<i>Dasht</i>	24	0.21	125	35	23.2	5.6	48.6	10.9	2.7	338	

**Botanical description:** The plant of this variety is semi erect and dark green in color. Hairiness and presence of anthocyanin pigmentation on the stem is a prominent character. Number of primary branches varies from 3-6 whereas secondary vary from 3-16 depending upon growth conditions. Inter-nodal length ranges from 1.5 to 2.7cm. The inter-nodal length at the top and bottom of the stem is narrow and wide in the middle of the stem. The numbers of leaflets range from 13-19. The leaflets are toothed and ovate in shape. The flowers are pink/red and seed are brown in color.

**Flower:** The flowers are red with 15-18 mm long pedicel and 2-6 mm long bracts. Calyx is toothed with 5 teeth. Anthers are bright/pink. The wings are longer than the filaments. The keel is 7-9 mm long.

**Pod:** Pods are ovate with little depression on dorsal side. They are medium large to large in size containing 1-2 seeds/pod. Pedicle is pigmented.

**Seed:** *Dasht* has large seed size with yellow brown seed color. The seeds are round and 100 seed weight goes up to 24gm. However, seed size varies depending upon the position of pod and growth.

**Seed quality:** Seed quality is an important parameter that determines the acceptability of a commodity among the consumers (Bhatty, 1988). The comparison of quality parameters showed that protein (23.2%), carbohydrate (48.6%), fats (5.6%), crude fibber (10.9%) contents of *Dasht* were comparable with those of C 44. Its 100 seed weight (24g), hydration capacity (0.21) and cooking time were comparable with those of Punjab 91 (Table 8). However, cooking time of *Dasht* under dry (125 minutes) and soaking (35 min.) condition is 7 and 10 minutes less than cooking time of C 44, under similar conditions. Energy obtained from 100g of *Dasht* was either equal or slightly more than that of checks.

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