

DUS characterization of lentil (*Lens culinaris* L.)

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ABSTRACT

The lentil core set of 105 genotypes was evaluated and characterized for 13 DUS descriptors. 11 out of 13 DUS characteristics showed variation indicating an exploitable amount of genetic variability. The DUS characteristics, leaf pubescence and plant height showed no variability and the remaining 11 DUS characteristics showed variability phenotypically and are considered for the identification, characterization and protection of genotypes. Pod anthocyanin colouration was seen in two genotypes *i.e.*, EC223397 and EC78414 and was absent in all other genotypes. Brown-coloured seed testa was exhibited by all the genotypes except EC223397 which showed black-coloured seed testa. All the genotypes showed orange-coloured cotyledon, except EC267634 which showed olive green-coloured cotyledon. Seed size (weight of 100 seeds) showed tetramorphic variation, foliage intensity of green colour, leaflet size (length) showed trimorphic variation while stem anthocyanin, time of flowering, plant growth habit, flower colour of standard petal and seed testa mottling showed dimorphic variation.

Keywords: Core set, DUS characterization, Genetic variability, Lentil and Protection

Lentil (*Lens culinaris* L.) is the first domesticated annual cool season grain legume having a genome size of 4 Gb (Polanco *et al.*, 2018). It is a diploid ($2n = 2X = 14$) self-pollinated crop belonging to the family Fabaceae (Leguminosae) and sub-family Papilionaceae (Arumuganathan and Earle, 1991). It is very well known as the “poor man’s meat” because of its huge amount of protein (Rani and Grewal, 2014). Systematic characterization of germplasm is required for its efficient use in future crop development programs. Morphological characterization of germplasm collection offers information about the variation available for various plant traits which can be exploited through various breeding processes. The PPV and FR Act is the legal framework of India adopted to protect plant varieties. Therefore, employing Distinctness, Uniformity and Stability (DUS) for characterization is crucial in the context of intellectual property regime to safeguard lines, ensure quality seed production and facilitate protection (Janghel *et al.*, 2020). DUS characterization of genotypes is vital for ensuring purity, identity, and protection of varieties. These descriptors are simple, cost-effective and do not necessitate specialized laboratory techniques.

In recognition of the importance and value of varietal classification, the current study was carried out with a lentil mini core set of 105 genotypes to characterize 13 DUS descriptors *i.e.*, foliage (intensity of green colour), stem (anthocyanin colouration), time of flowering, leaf pubescence, leaflet size (length), plant growth habit, flower colour of standard petal, plant height, pod anthocyanin colouration, seed size (weight of 100 seeds), seed testa colour, seed testa mottling and cotyledon colour.

MATERIAL AND METHODS

The experimental material consisted of 105 genotypes of lentil mini core set. The experiment was conducted in alpha Lattice design during *rabi* 2023-2024 at Agricultural College Farm, Bapatla, Andhra Pradesh. All the genotypes were planted in two rows of three meters in length adopting 30 x 10 cm spacing among the genotypes and plants, respectively for the morphological characterization. The observations were made at specific crop growth stages when the characteristics under investigation were fully expressed. Foliage (intensity of green colour) was observed at the flower bud stage. The DUS characteristics, stem anthocyanin colouration, time of

flowering, leaf pubescence, leaflet size (length), plant growth habit and flower colour of standard petal were observed at the days to 50% flowering stage. Plant height was recorded at the harvest maturity stage and pod anthocyanin colouration was observed in the fully developed green pod. The DUS characteristics seed size (weight of 100 seeds), seed testa colour, seed testa mottling and cotyledon colour were observed in fully matured seeds.

RESULTS AND DISCUSSION

Characterization of lentil mini core set of 105 genotypes was carried out using DUS descriptors. Out of 13 DUS characters, 11 characters showed exploitable amount of genetic variability indicating their use in characterization studies for identification and protection.

The character, foliage intensity of green colour showed sufficient variability among the genotypes. Foliage intensity was light green in 22 genotypes, medium green in 80 genotypes and dark green in 3 genotypes. Leaves with dark-coloured foliage intensity have a higher chlorophyll concentration which affects photosynthesis and gives a selection advantage that contributes significantly to the plant's biomass as they persist for a long time and remain green even in the advanced phases of crop growth, whereas light green plants mature and turn brown (Choudhary *et al.*, 2017).

Stem anthocyanin colouration is a highly used trait for characterizing the genotypes and it is an easily distinguishable trait phenotypically. Stem anthocyanin was absent in 52 genotypes but observed in 53 genotypes. The presence of anthocyanins in the stem may allow the plant to develop resistance to several environmental stresses.

Early flowering (<60 days) was seen in 94 genotypes, while medium flowering (60-80 days) was found in 11 genotypes. These 94 genotypes of early flowering can be exploited in the development of early maturity short-duration types.

Leaflet size was small in 23 genotypes, medium in 53 lines and large in 30 genotypes. This characteristic plays an important role in the yielding ability of the genotypes as the leaves are the source of food synthesis and the transpiration site of the plants which provide a selective advantage. This plays an important role in increasing the biomass of the plant.

The plant growth habit was erect in 58 genotypes and semi-erect in 47 genotypes. Erect plant forms are generally preferred since they receive plenty of sunlight and efficiently synthesize food for plant growth and development.

The standard petal colour of genotypes is a reliable morphological marker for characterizing the genotypes and helps to easily distinguish the traits. Out of the 105 genotypes, 17 lines had white-coloured standard petals and 88 lines had violet-coloured standard petals.

Pod anthocyanin colouration of genotypes is a useful characteristic for genotype characterization and is simple to identify phenotypically. Pod anthocyanin colouration was absent in 103 genotypes and present in two genotypes i.e., EC78414 and EC223397.

The trait, 100 seed weight, is considered an important yield contributing character and is exploited as a descriptor in DUS characterization. Out of 105 genotypes studied, 6 genotypes are small (less than 2g), 36 genotypes are medium (2-2.5g), 39 genotypes are large (2.6-3g) and 24 genotypes are very large (more than 3g).

The seed testa colour of genotypes is a highly used trait for characterizing the genotypes and easily distinguishable trait phenotypically. Black lentils consist of higher amount of polyphenols when compared to brown lentils. The genotype EC223397, was categorized as black type and other genotypes were grouped as brown type.

Cotyledon colour was observed in mature seeds and it is a highly used trait for characterizing the genotypes and helps to distinguish the trait phenotypically. Carotenoid concentration in green cotyledon is higher than in yellow cotyledon lentils. The genotype, EC267634, had olive green coloured cotyledon and the remaining genotypes had orange coloured cotyledon. Similar results of DUS characterization in lentil were reported by Mohammed *et al.* (2019), Kumar *et al.* (2020) and Harini *et al.* (2022).

Thus present study revealed notable genetic variation for various plant traits which can then be exploited through various breeding programmes. According to the PPV & FR Act, the information acquired will be helpful not only for registration but also in the wider context of varietal purity, identity,

Table 1 Classification of 105 genotypes of lentil (*Lens culinaris* L.) based on eleven variable DUS characteristics

S. No.	Genotypes	Foliage intensity of green colour	Stem anthocyanin colouration	Time of flowering	Leaflet size (length)	Plant growth habit	Flower colour of standard petal	Pod anthocyanin colouration	Seed size (weight of 100 seeds)	Seed testa colour	Seed testa mottling	Cotyledon colour
1	IC560181	1	1	5	7	1	1	1	5	4	3	3
2	L7920	2	1	3	5	3	4	1	7	4	3	3
3	DPL15	2	9	3	5	3	4	1	9	4	1	3
4	IG69568	1	9	3	3	1	4	1	5	4	3	3
5	PI4105	2	9	5	3	3	1	1	3	4	3	3
6	P3235	1	9	3	5	1	1	1	9	4	1	3
7	P8112	2	9	3	5	3	1	1	5	4	1	3
8	L4649	3	1	3	5	3	4	1	7	4	3	3
9	ILWL118	2	1	3	7	3	4	1	7	4	3	3
10	PI17	2	9	3	7	1	4	1	9	4	3	3
11	IC346092	2	1	3	5	3	4	1	7	4	3	3
12	PI3145	2	1	3	5	1	4	1	7	4	3	3
13	PI5111	2	1	3	3	3	4	1	7	4	3	3
14	PI4903	2	1	3	5	3	4	1	3	4	3	3
15	MC6	3	1	3	7	1	4	1	9	4	3	3
16	PI3142	2	9	3	5	1	4	1	7	4	3	3
17	PI3130	2	1	3	7	3	4	1	7	4	3	3
18	EC223212-A	2	1	3	5	1	4	1	9	4	3	3
19	EC223209-B	1	1	3	7	1	4	1	9	4	3	3
20	EC223205-B	2	1	3	5	3	4	1	9	4	3	3
21	EC223201	2	1	3	3	3	1	1	5	4	3	3
22	PI3131	2	1	3	7	1	4	1	7	4	3	3
23	EC78414	2	9	3	7	1	4	9	9	4	3	3
24	EC223197-A	1	9	3	5	3	4	1	5	4	3	3
25	EC223191	2	1	3	7	1	4	1	5	4	3	3
26	EC223150	2	9	3	5	1	4	1	5	4	3	3
27	EC223221	2	1	3	3	1	4	1	9	4	3	3
28	EC223215	2	1	3	5	1	4	1	5	4	3	3
29	EC78545	2	9	3	5	3	4	1	5	4	3	3
30	EC78543	1	1	3	7	1	1	1	5	4	3	3
31	EC78541-A	2	9	3	7	3	4	1	5	4	3	3
32	EC78539	2	1	5	7	1	4	1	5	4	3	3
33	EC78532	1	9	3	3	3	1	1	5	4	3	3
34	EC78529	1	9	3	7	1	4	1	7	4	3	3
35	EC78528	2	9	3	3	1	4	1	7	4	3	3

36	EC267634	2	9	5	7	1	4	1	5	4	3	2
37	EC78395	1	9	5	3	3	4	1	3	4	3	3
38	EC78408	2	9	3	5	1	4	1	5	4	3	3
39	EC78453	2	9	3	7	3	4	1	9	4	3	3
40	EC78441-B	2	9	3	5	3	1	1	7	4	3	3
41	EC78438	2	9	3	7	1	4	1	5	4	3	3
42	EC78437	1	1	3	7	1	4	1	7	4	3	3
43	EC78434	2	9	3	5	1	4	1	5	4	3	3
44	EC78933	1	1	5	7	1	4	1	5	4	3	3
45	EC78554	2	9	3	5	3	4	1	7	4	3	3
46	EC329166	2	9	3	7	1	4	1	3	4	3	3
47	EC267709	2	1	3	3	1	4	1	7	4	3	3
48	EC267696	1	9	3	3	1	4	1	5	4	3	3
49	EC223230	2	1	5	3	1	4	1	5	4	3	3
50	EC223229-B	2	9	5	5	1	4	1	3	4	3	3
51	EC223229-A	2	9	3	7	1	1	1	5	4	3	3
52	EC223226	2	1	3	3	1	4	1	7	4	3	3
53	EC223222	2	1	3	5	3	4	1	9	4	3	3
54	EC78459	1	9	3	3	1	4	1	5	4	3	3
55	EC267641	2	9	5	5	3	4	1	9	4	3	3
56	EC267638	2	9	3	3	1	4	1	9	4	3	3
58	EC267628-A	1	1	3	5	3	4	1	5	4	3	3
59	EC267625-C	2	9	3	7	1	4	1	5	4	3	3
60	EC267609	1	9	3	7	1	4	1	5	4	3	3
61	EC267604	1	9	5	5	1	4	1	9	4	3	3
62	EC267605	2	9	3	3	1	4	1	5	4	3	3
63	EC267595-C	2	9	3	5	1	4	1	7	4	3	3
64	EC78498	2	1	3	7	1	4	1	9	4	3	3
65	EC267676	2	9	3	7	1	4	1	5	4	3	3
66	EC267544-A	2	9	3	3	1	4	1	7	4	3	3
67	EC78511	2	9	3	3	3	4	1	9	4	3	3
68	EC78513	1	9	3	3	1	4	1	7	4	3	3
69	EC78509	2	1	3	5	1	4	1	5	4	3	3
70	EC78506	2	1	3	5	1	4	1	7	4	3	3
71	EC78499	2	1	3	3	3	4	1	5	4	3	3
72	EC255491	1	1	3	3	1	4	1	7	4	3	3
73	EC2675471	2	1	3	5	3	4	1	7	4	3	3
74	EC267569-B	2	9	3	3	1	4	1	7	4	3	3
75	EC267569-A	2	1	3	5	1	4	1	7	4	3	3
76	EC267563	2	9	3	7	1	4	1	7	4	3	3

77	EC267557-D	2	9	3	5	3	4	1	9	4	3	3
78	EC267554	2	9	3	5	1	4	1	5	4	3	3
79	EC267545-D	2	1	3	5	3	4	1	5	4	3	3
80	EC225501	2	1	3	5	3	4	1	7	4	3	3
81	EC223242	2	1	3	5	3	4	1	7	4	3	3
82	EC223188	1	1	3	5	3	4	1	9	4	3	3
83	EC78551-A	2	1	3	5	3	4	1	9	4	3	3
84	EC267540	1	1	3	5	1	4	1	9	4	3	3
85	EC223397	2	9	3	5	3	4	9	7	5	1	3
86	EC267539	2	9	3	5	1	4	1	9	4	3	3
87	EC267536	2	9	3	5	3	4	1	7	4	3	3
88	EC267533	2	9	3	7	1	4	1	7	4	3	3
89	EC267529	2	9	3	5	3	4	1	7	4	3	3
90	EC78488	2	1	3	7	1	4	1	7	4	3	3
91	EC78474	1	9	3	7	3	4	1	7	4	3	3
92	EC78472	2	1	3	7	3	4	1	9	4	3	3
93	EC78542-A	2	9	3	3	3	4	1	5	4	3	3
94	EC78477-A	2	1	3	5	3	4	1	5	4	3	3
95	EC78476	2	1	3	5	1	4	1	9	4	3	3
96	EC78425	2	1	3	3	3	4	1	5	4	3	3
97	EC78423	1	1	3	5	3	4	1	5	4	3	3
98	EC78421	2	9	3	5	3	1	1	3	4	3	3
99	EC241476	2	9	3	5	3	1	1	7	4	3	3
100	EC78524-A	2	1	3	5	3	1	1	7	4	3	3
101	EC78520	2	1	3	5	3	1	1	7	4	3	3
102	EC78519	2	1	3	5	3	1	1	7	4	3	3
103	EC78515	2	1	3	5	3	4	1	7	4	3	3
104	EC78487	2	1	3	5	1	1	1	9	4	3	3
105	EC78495	2	1	3	5	1	1	1	5	4	3	3

Note:

Light-1
Medium-2
Dark-3Absent-1
Present-9Early(<60days)3
Medium(60-80days)-5
Late (>80 days)-7Small-3
Medium-5
Large-7Erect (< 30°)1
Semi-erect(30°-60°)-3
Horizontal (>60°)-5White-1
Pink-2
Blue-3
Violet-4
Others(specify)-5Absent-1
Present-9Small (< 2.0g)3
Medium (2.0-2.5g)-5
Large (2.6-3.0g)-7
Very large (>3.0 g)-9Green-1
Grey-2
Pink-3
Brown-4
Black-5Absent-1
Present-3
Yellow-1
Olive green-2
Orange-3

Table 2. Genotypes and their specific DUS characteristics in lentil (*Lens culinaris* L.)

S.No.	Genotypes	No. of Specific DUS characteristic	Specific DUS Characteristics (s)
1	IC 560181	3	Light foliage intensity; large leaflet size and white colour standard petal
2	L7920	3	Medium foliage intensity; semi-erect plant growth habit and violet colour standard petal
3	DPL15	3	Seed testa mottling absent; brown colour seed testa and orange colour cotyledon
4	IG69568	2	Large leaflet size and stem anthocyanin colouration absent
5	P14105	2	Small seed size and semi-erect plant growth habit
6	P3235	2	White colour standard petal and seed testa mottling absent
7	P8112	2	Pod anthocyanin colouration absent and seed testa mottling absent
8	L4649	2	Dark foliage intensity and stem anthocyanin colouration absent
9	ILWL118	2	Medium foliage intensity and large leaflet size
10	P117	2	Erect plant growth habit and orange colour cotyledon colour
11	IC346092	3	Medium foliage intensity; semi-erect plant growth habit and brown colour seed coat
12	P13145	2	Large seed size and medium leaflet size
13	P15111	2	Small leaflet size and brown colour seed testa
14	P14903	2	Pod anthocyanin absent and seed testa mottling present
15	MC6	2	Dark foliage intensity and very large seed size
16	P13142	2	Erect (plant growth habit) and absent (pod anthocyanin colouration)
17	P13130	2	Medium foliage intensity and seed testa mottling present
18	EC223212-A	2	Medium seed size and stem anthocyanin colouration present
19	EC223209-B	2	Very large seed size and brown colour seed testa
20	EC223205-B	2	Brown colour seed testa and orange colour cotyledon
21	EC223201	2	Stem anthocyanin colouration absent and small leaflet size
22	P13131	2	Large leaflet size and orange colour cotyledon
23	EC78414	2	Pod anthocyanin colouration present and very large seed size
24	EC223197-A	3	Light foliage intensity; stem anthocyanin colouration present and brown colour seed testa
25	EC223191	3	Medium seed size; erect plant growth habit and violet colour standard petal
26	EC223150	2	Medium leaflet size and medium seed size
27	EC223221	3	Small leaflet size; erect plant growth habit and violet colour standard petal
28	EC223215	3	Stem anthocyanin colouration absent; medium seed size and violet colour standard petal
29	EC78545	3	Semi erect plant growth habit; brown colour seed testa and seed testa mottling present
30	EC78543	3	Light foliage intensity; large leaflet size and white colour standard petal
31	EC78541-A	2	Semi-erect plant growth habit and seed testa mottling present
32	EC78539	3	Large leaflet size; violet colour standard petal and orange colour cotyledon
33	EC78532	3	Medium foliage intensity; violet colour standard petal and seed testa mottling present
34	EC78529	3	Light foliage intensity; large leaflet size and brown seed coat colour
35	EC78528	3	Medium foliage intensity; small leaflet size and large seed size
36	EC267634	1	Olive green colour cotyledon
37	EC78395	3	Light foliage intensity; violet colour standard petal and orange colour cotyledon
38	EC78408	3	Erect plant growth habit; medium seed size and stem anthocyanin colouration present
39	EC78453	3	Stem anthocyanin colouration present; violet colour standard petal and brown colour seed testa
40	EC78441-B	2	Semi-erect plant growth habit and white colour standard petal

41	EC78438	2	Large leaflet size and medium seed size
42	EC78437	3	Stem anthocyanin colouration absent; erect plant growth habit and brown colour seed testa
43	EC78434	2	Erect plant growth habit and pod anthocyanin colouration absent
44	EC78933	3	Light foliage intensity; violet colour standard petal and brown colour seed testa
45	EC78554	2	Stem anthocyanin colouration present and pod anthocyanin colouration absent
46	EC329166	2	Large leaflet size and small seed size
47	EC267709	3	Erect plant growth habit; medium seed size and pod anthocyanin colouration absent
48	EC267696	2	Stem anthocyanin colouration present and violet colour standard petal
49	EC223230	3	Pod anthocyanin colouration absent; violet colour standard petal and orange colour cotyledon
50	EC223229-B	2	Violet colour standard petal and small seed size
51	EC223229-A	2	Large leaflet size and white colour standard petal
52	EC223226	2	Small leaflet size and orange colour cotyledon
53	EC223222	3	Large seed size; pod anthocyanin colouration absent and orange colour cotyledon
54	EC78459	3	Light foliage intensity; violet colour standard petal and brown colour seed testa
55	EC267641	2	Stem anthocyanin colouration present and semi-erect plant growth habit
56	EC267638	3	Small leaflet size; violet colour standard petal and very large seed size
57	EC267636	3	Dark foliage intensity; white colour standard petal and orange colour cotyledon
58	EC267628-A	3	Medium foliage intensity; medium leaflet size and medium seed size
59	EC267625-C	2	Stem anthocyanin colouration present and orange colour cotyledon
60	EC267609	3	Erect plant growth habit; pod anthocyanin colouration absent and brown colour seed testa
61	EC267604	3	Violet colour standard petal; very large seed size and seed testa mottling present
62	EC267605	3	Medium foliage intensity; pod anthocyanin colouration absent and medium seed size
63	EC267595-C	3	Stem anthocyanin colouration present, brown colour seed testa and erect plant growth habit
64	EC78498	2	Large leaflet size and seed testa mottling present
65	EC267676	3	Erect plant growth habit; medium seed size and violet colour standard petal
66	EC267544-A	2	Stem anthocyanin colouration present and pod anthocyanin colouration absent
67	EC78511	3	Semi-erect plant growth habit; very large seed size and small leaflet size
68	EC78513	2	Large seed size and pod anthocyanin colouration absent
69	EC78509	3	Brown colour seed testa; stem anthocyanin colouration absent and medium leaflet size
70	EC78506	2	Pod anthocyanin colouration absent and seed testa mottling present
71	EC78499	2	Small leaflet size and seed testa mottling present
72	EC255491	3	Light foliage intensity; large seed size and stem anthocyanin colouration absent
73	EC2675471	3	Semi-erect plant growth habit; orange colour cotyledon and seed testa mottling present
74	EC267569-B	3	Small leaflet size; stem anthocyanin colouration present and erect plant growth habit
75	EC267569-A	2	Violet colour standard petal and brown colour seed testa
76	EC267563	2	Large leaflet size and seed testa mottling present
77	EC267557-D	3	Semi erect plant growth habit; orange colour cotyledon and seed testa mottling present
78	EC267554	2	Medium seed size and erect plant growth habit
79	EC267545-D	2	Pod anthocyanin colouration absent and violet colour standard petal
80	EC225501	3	Medium leaflet size; seed testa mottling present and orange colour cotyledon
81	EC223242	2	Violet colour standard petal and stem anthocyanin colouration absent
82	EC223188	2	Light foliage intensity and semi-erect plant growth habit
83	EC78551-A	3	Violet colour standard petal; medium leaflet size and seed testa mottling present
84	EC267540	2	Light foliage intensity and orange colour cotyledon

85	EC223397	2	Black colour seed testa and pod anthocyanin colouration present
86	EC267539	3	Erect plant growth habit; violet colour of standard petal and very large seed size
87	EC267536	2	Semi-erect plant growth habit and seed testa mottling present
88	EC267533	2	Large leaflet size and orange colour cotyledon
89	EC267529	3	Semi-erect plant growth habit; medium leaf let size and brown colour seed testa
90	EC78488	2	Large leaflet size and stem anthocyanin colouration absent
91	EC78474	2	Light foliage intensity and large seed size
92	EC78472	2	Very large seed size and violet colour standard petal
93	EC78542-A	2	Small leaflet size and stem anthocyanin colouration present
94	EC78477-A	3	White colour standard petal; medium seed size and semi-erect plant growth habit
95	EC78476	2	Pod anthocyanin colouration absent and violet colour standard petal
96	EC78425	2	violet colour standard petal and orange colour cotyledon
97	EC78423	3	Light foliage intensity; medium leaflet size and seed testa mottling present
98	E C78421	2	White colour standard petal and brown colour seed testa
99	EC241476	2	Large seed size and white colour standard petal
100	EC78524-A	2	White colour standard petal and pod anthocyanin colouration absent
101	EC78520	3	Semi-erect plant growth habit and white colour standard petal
102	EC78519	2	Stem anthocyanin colouration absent and white colour standard petal
103	EC78515	3	Light foliage intensity; large seed size and white colour standard petal
104	EC78487	3	White colour standard petal, brown colour seed testa and orange colour cotyledon
105	EC78495	3	Erect plant growth habit; medium leaflet size and white colour standard petal



Large Medium Small

Fig 1: Variation in leaflet size of lentil



Semi erect Erect

Fig 2: Variation in plant growth habit of



White



Violet

Fig 3: Variation in flower colour of standard petal of lentil



Absent



Present

Fig 4. Variation in pod anthocyanin colouration of lentil

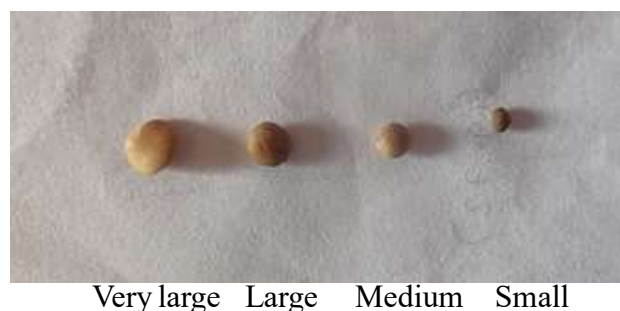


Fig 5: Variation in seed size of lentil



Fig 7: Variation in cotyledon colour of lentil



Fig 6: Variation in seed testa colour of lentil



Fig 8: Variation in seed testa mottling of lentil

and protection. The characterization of mini core set opened up opportunities for their use in selection, molecular breeding, and trait discovery.

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