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DUS characterization of linseed (*Linum usitatissimum* L.) germplasm

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ABSTRACT

The present study was carried out during rabi 2020-21 and rabi 2021-22 respectively, where a total of 2612 and 2657 germplasm lines were evaluated for various qualitative and yield and yield attributing traits. Among all these genotypes, the genotypes which have been found common for both the seasons have been categorized. From the results of two seasons, common germplasm lines found for earliness, dwarf stature, and high number of capsules^{-1} plant are IC0118855, IC0498442, IC0498490, IC0342800, EC41466, EC0541213, IC0096701, IC0203213, IC0096533 and IC0525956. Also, common germplasm lines for found for resistance to bud fly infestation and Alternaria blight are IC0096551, IC0342801, IC620658, IC0118859, IC0498634, IC0526105, IC0498795, IC0499155, IC0498843, IC0599415, IC0498763, EC993391, EC993389 and IC633096. These promising genotypes are of the prime candidates for active integration into further breeding initiatives, aimed at cultivated genotypes with desirable characteristics. Furthermore, the strategic utilization of diverse facts found whether the germplasm lines is to play a pivotal role in processes associated with registration, ongoing maintenance & safeguarding of linseed lineages.

Keywords: *Germplasm lines, Linseed, DUS, Characterization, UPOV 2011*

I. INTRODUCTION

Linseed is (*Linum usitatissimum* L.; n = 15), also called as flax, an important cool temperate oilseed crop belonging to the Linaceae family, with 14 genera and over 200 species. This is the only species in the Linaceae family with economic and agronomic values [25]. The cultivated species originated from a single domestication event from *L. bienne* [2] and it is native to the Mediterranean region [25]. The cultivated Linum species have more economic values. It is cultivated commercially for its seed, which is processed into oil and a high protein stock feed after oil extraction [22, 17, 5] and for its fibers, which are made into linen and other cloths [11]. Recently it has gained a new interest in the emerging market of functional food due to its high content of fatty acids, alpha-linolenic acid (ALA), an essential Omega-3 fatty acid, and lignin oligomers which constitute about 57 % of total fatty acids in linseed [21]. In addition, linseed varieties with oils suitable for culinary use are available [15].

Linseed is currently cultivated in Russia, the U.S.A., Argentina, Uruguay, India, Pakistan, China, Japan, Morocco, Australia, Ireland, Scotland, Poland, and a few other European countries. In India, linseed is grown in a total area of about 1727 lakh hectares with a production of about 990 tonnes and a productivity 574 kilograms per hectare. In Maharashtra, linseed is grown in a total area of about 57.48 lakh hectares with a production of about 22.18 tonnes and productivity 385.84 kilograms per hectare, whereas in Vidarbha region of Maharashtra, linseed is grown in a total area of about 51.84 lakh hectares with a production of about 20.38 tonnes and productivity 381.9 kilograms per hectare [3]. It is widely cultivated in Rajasthan, Bihar, Uttar Pradesh, Assam, and Jharkhand followed by other states. Madhya Pradesh has the largest growing area (1.16 lakh ha) and production (0.55 lakh tones) with 474 kg/ha productivity [www.indiastat.in.].

Describing the characteristics of a crop species based on standard descriptors is effective for better utilization and conservation of germplasm [10]. Looking to the above facts study was undertaken for the classification of 2612 and 2657 diverse linseed morphological germplasm based on linseed descriptors or DUS guidelines or DUS descriptor as per UPOV 2011.

II. MATERIALS AND METHODS

The present study was conducted in research field of All India Co-Ordinated Research Project on Linseed at College of Agriculture, Nagpur, Maharashtra (21^{0}8'26 N Latitude and 79^{0}4'22 E Longitude at an altitude of 245.03 meters above mean sea level). The experiment comprised of 2612 and 2657 linseed germplasm lines including exotic and indigenous. These lines were sown subsequently for two years i.e. *rabi* 2020-21 and *rabi* 2021-22. Wilting was observed in several germplasm lines. The seed was availed from the gene bank of ICAR-NBPGR, New Delhi. All these germplasm lines were evaluated for various yield and yield-attributing traits. These germplasm lines were evaluated on the basis of DUS characters. The entries were sown in a single row of 3-meter length with inter and intra-row spacing of 30 cm and 15 cm, respectively. All recommended agronomic practices were adopted in order to raise normal crops. The observations were recorded from five randomly selected competitive plants from each row on seven distinct morphological characters.

The experimental material was planted in augmented design. Observations were recorded as per DUS UPOV 2011, in which five random plants from each line were taken for recording of data. Quantitative and qualitative characters were examined using measurements from a single plant or its part or from

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groups of plants or their parts, visual assessments from single plant or its part or from groups of plants or their parts, depending on the element used to characterize the accession and analysis carried out as per DUS, UPOV 2011. To assess distinctness (D), uniformity (U) and stability (S), the characteristics and their states as given for the characteristics were used at the optimum plant growth stage.

III. RESULTS AND DISCUSSION

Descriptors for linseed germplasm (Table 1 and Table 2) details the nature and range of agronomic and morphological descriptors and classifiers that we defined for the linseed germplasm grown in field plots. Ranges reported below refer to the 198 accessions of Ethiopian material except where noted.

Characterization defines the expression of highly heritable characters ranging from morphological or agronomical features. The importance of characterization is that it allows to know the information on various characteristics of plants so that maximum utilization of germplasm lines for plant breeders. Through proper evaluation of germplasm for its various traits a breeder can have it's better practical utilization. In the process of evaluation, a number of activities have been involved starting from receiving new samples from the curator and growing these for seed increase, characterization, and preliminary evaluation, and also for further or detailed evaluation and documentation. As per the breeding objectives and program of evaluation, close organizational and personal contact between the curator and breeder is needed.

In the present study, out of 2612 and 2657 diverse lines every accession was repeatedly observed during the season at various growth stages. The volunteer and off-type plants were roughed out. Measurement of qualitative traits is done by measuring a single plant or group of plants with the visual assessment of the same. The observation on single plant or group of plants is decided based on the particular element for the characterization of that particular accession. All morphological descriptors showed remarkable differences in their distribution and the amount of variations within them.

Flower characters: Floral characters were highly polymorphic and provided a useful group of seven descriptors. As flower characters are qualitative in nature, it remains the same for generation to generation. All the traits are mentioned in Table 1.

Corolla/petal colour (CC): It scored in fully opened flowers, it was measured at 50% flowering. It was ranged from blue to white including five various classes *viz*., blue, light blue, red, red-violet, violet and white. A total of 102 accessions had blue colour corolla, 1137 accession had light blue, only one accession had red colour. Seven genotypes with Red-violet, 1020 violet and white coloured corolla with 123 accessions (Table 1).

Flower/Petal aestivation (PA): It describes overlapping of petals ranging from twisted to valvate flowers. This observation of individual plants at 50% flowering stage was recorded before noon. Twisted trait was found in 858 accessions, 1052 had semi-twisted aestivation and 533 germplasm lines showed valvate accessions (Table 1).

Flower shape(FS): Included disk shaped and funnel shape. 50 accessions had disc-shaped flowers and a funnel shape was found in 2407 accessions (Table 1).

Anther colour (AC)*:* It was showed a continuous range of colour variation *viz*., black, blue, cream, grey and violet. Black-coloured anthers were found in a single germplasm line. Blue colour was found in 1733 accessions. Cream, grey and violet anther colour was found in 494, 46 and 116 accessions, respectively (Table 1).

Filament/ stamen colour (FC): It had a narrower range of colour than anthers and petals. The range of stamen colours was varied from blue, violet and white. Blue coloured stamens are found in 620 accessions, although violet and white coloured stamen are found in 1097 and 726 accessions, respectively (Table 1).

Flower Venation Colour (FVC): The trait had showed more polymorphism ranging from blue to violet. A total of six colours were found for flower venation colour *viz*., blue, light violet, red, red violet, violet and white. Red and red violet flower venation was found in a single genotype each. However, blue venation colour was found in 1808, light violet colour was in 128 germplasm lines, violet colour was observed in 358 and white was observed in 93 entries (Table 1).

Corolla size: It was measured from fully opened flowers at 50% flowering stage. It was divided into two categories as small (<5 mm) and large (>5 mm). A total of 99 lines had small corolla size and 2369 germplasm lines showed large corolla size. In the presence study, it was observed that almost all white petal flowers with valvate aestivation accessions had a large corolla size. Over half of the studied accessions were characterized by large size of corolla (Table 1).

Plant characteristics

Early Plant Vigour: The observation on plant vigour was recorded after 25 days of sowing and it was categorized into three classes *viz*., poor, good and very good. During *rabi* 2020-21, poor plant vigour was observed in 154 accessions, good vigour was observed in 2228 accessions and very good vigour was seen in 204 accession lines. Also, there was much variation for early plant vigour during *rabi* 2021-22 like, poor plant vigour was observed in 154 accessions, whereas 2243 and 204 accessions showed good and very good accessions, respectively.

Plant height (cm): The height of plant was recorded in centimeters from the base to the tip of main stem. It has divided into three classes namely, dwarf (<50 cm), medium (50-70 cm) and tall (>70 cm). Plant height was measured in these many germplasm lines and ranged from 20.2 to 146.6 cm in the *rabi* 2020-21, whereas it was ranged from 10 to 57 cm for *rabi* 2021-22. During *rabi* 2020-21, out of 2286 germplasm lines, 1918 germplasm lines are categorized as dwarf whereas, 359 were categorized as medium in height and nine genotypes found to be tall in height (Table 2). Whereas during *rabi* 2021-22, out of 2626 genotypes dwarf genotypes found were 2625 and a single genotype was found as of medium height. The common genotypes which found as dwarf in both the seasons are 1875 and no genotype found common under the medium and tall categories. During both the seasons, a total of 1875 accessions were described as dwarf and no common genotype found for medium and tall plant height.

Days to 50% flowering: Days to 50% flowering (DFF) showed continuous variation from 43 to 78 days during *rabi* 2020-21, whereas it was ranged from 46 to 73 cm for *rabi* 2021-22. For

each genotype, number of days taken from the day of sowing to the day on which 50% of the plants showed flowering was recorded as the number of days taken for 50% flowering. It was also divided into three classes *viz*., early (<50 days), medium(50-60 days), and late (>60 days). During *rabi* 2020-21 the genotypes which found early are 299, medium duration are 1181 and late are 806. Whereas, during *rabi* 2021-22, actual sowing of 2657 was done but again due to wilt disease some of the genotypes wilted till the plants bear 50% flowering. The genotypes which found in early category during this season are 60, medium duration is found in 1558 genotypes and a total of 1035 genotypes were found as late in duration (Table 2). During both the seasons, common germplasm found for early duration are 31 in number, for medium duration 968 and common germplasm lines for late duration are 598 in number.

Total capsules per plant: The trait has been alienated into three categories, *viz*., low (<30 capsules), medium (30-60 capsules) and high (>60 capsules). During *rabi* 2020-21, less number of total capsules was found in 61 genotypes, a medium number of capsules were in 884 genotypes and high number of capsules were found in 1341 genotypes. During *rabi* 2021-22, out of 2626 genotypes low number of capsules were found in 25 genotypes, medium in 763 and high number of total capsules were found in 1838 genotypes. Common genotypes found during both the season for less number of capsule is one, medium number of capsules are 250 genotypes and high number of capsules found in 644 genotypes (Table 2).

Primary branches per plant: The trait primary branches per plant was categorized as low (<5 branches) and high (>5 branches). During *rabi* 2020-21, less number of primary branches were found in 1821 genotypes and 465 genotypes had a high number of branches. During *rabi* 2021-22, 2128 genotypes had less primary branches and 498 genotypes had with high number of branches. Common genotypes with low number of branches in 1165 genotypes and high number of branches are found in 125 genotypes (Table 2).

Seed yield per plant: Seed yield per plant was divided into three categories *viz*., low (<2 gm), medium (2-5 gm) and high (>5 gm). During *rabi* 2020-21, 1249 genotypes found are low yielder, 1005 genotypes were with medium yield, and 32 genotypes are yielded high. During *rabi* 2021-22, 282 genotypes found are low yielders, 2149 genotypes were with medium yield and 195 genotypes are yielded high. Common genotypes found during both the season are 147 for low yield, 819 genotypes for medium yield and no common genotype found for high yield (Table 2).

% Bud fly Infestation: It is grouped into five classes as, Resistant (0-10), Moderately resistant (10.01-25), Moderately Susceptible (25.01-50), Susceptible (50.01-75) and Highly Susceptible (>75). During *rabi* 2020-21, the 168 genotypes are resistant, 1426 genotypes are moderately resistant, 676 germplasm lines are moderately susceptible and 16 genotypes are susceptible, whereas no genotypes were found as highly susceptible. During *rabi* 2021-22, resistant genotypes found are 434, 2110 are moderately resistant, 81 germplasm lines are moderately susceptible, one genotype was susceptible and no genotype was found as highly susceptible. The number of common genotypes found during both the season are 37 for the resistant category, 1200 for moderately resistant, 24 genotypes are moderately susceptible. No genotype was found common for susceptible and highly susceptible (Table 2).

% Alternaria blight: Like bud fly infestation,% Alternaria blight is also grouped into five classes as, Resistant (0-10), Moderately resistant (10.01-25), Moderately Susceptible (25.01-50), Susceptible (50.01-75) and Highly Susceptible (>75). During *rabi* 2020-21, the 730 genotypes are resistant, 1445 genotypes are moderately resistant, 102 germplasm lines are moderately susceptible and nine genotypes are susceptible, whereas no genotypes are found as highly susceptible. During *rabi* 2021-22, resistant genotypes found for alternaria blight are 1250, 1360 are moderately resistant genotypes, 16 genotypes are moderately susceptible and no common genotype found for susceptible and highly susceptible. The number of common genotypes found during both the seasons are 344 for the resistant category, 785 for moderately resistant, only one genotype is moderately susceptible. No genotype found common for susceptible and highly susceptible (Table 2).

In short from table 2, One group of ten accessions, which was characterized for early maturity, dwarf stature, and a high number of capsules are IC0096533, IC0525956, IC0118855, IC0498442, IC0498490, IC0342800, EC41466, EC0541213, IC0096701 and IC0203213. The second group including fourteen germplasm lines was characterized for resistance against the two major biotic stresses in linseed *viz*., bud fly infestation and alternaria blight. The genotypes which included in second group are IC0096551, IC0342801, IC620658, IC0118859, IC0498634, IC0526105, IC0498795, IC0499155, IC0498843, IC0599415, IC0498763, EC993391, EC993389 and IC633096 (Table 2).

IV. DISCUSSION

In order to meet various objectives of plant breeding in terms of wider adaptation, enhanced yield, insect pest and disease resistance, and desirable quality, germplasm diversity is of utmost importance [16, 4]. In earlier studies, various researchers have worked on the DUS characterization of linseed germplasm [6, 7, 20]. Recently [26] also evaluated 109 linseed germplasm lines. [1, 13, 23 and 24] reported a wide range of genetic diversity in linseed. Results from the present study shows that, many genes interact for the development of different characters. Morphological variation is very important in the selection of distinguishable, uniform, and stable traits. This is very useful at the time of seed production, and monitoring program of linseed.

Agronomic and phenotypic characters of linseed: [12] considered that there was limited diversity in linseed and concluded that serious genetic erosion had occurred [19].

All the floral structures had different subclasses of color as colour trait characters. [14] studied number and type of petal colour in linseed and the findings of present study are similar to it. Also [14] reported linseed anther only as blue and yellow, different from the range of anther colours found here, although in wild species of *Linum grandiflorum*, yellow anther has been observed. Color petals scored from world core collections by [8]. [9] reported three anther colours: white, blue and yellow. In the study of gene interaction, [14] found that there are eight gene interacting with other for the development of colours in anther, stamen and petal.

V. CONCLUSION

From the results of two seasons, common germplasm lines found for earliness, dwarf stature and high number of capsules^{-1} plant are IC0118855, IC0498442, IC0498490, IC0342800, EC41466, EC0541213, IC0096701, IC0203213, IC0096533 and IC0525956. Also, common germplasm lines for found for resistance to bud fly infestation and Alternaria blight are IC0096551, IC0342801, IC620658, IC0118859, IC0498634, IC0526105, IC0498795, IC0499155, IC0498843, IC0599415, IC0498763, EC993391, EC993389 and IC633096. Also, the significant morphological variation in studied germplasm can be utilized for making the diallel crosses amongst diverse parents. These crosses may result into good transgressive segregants for desirable traits.

Drawing for the data gathered across two seasons, the genotypes exhibiting traits of early maturity, a notable abundance of capsules and dwarfed plant stature holds significant promise. The genotypes offering resistance to two major biotic stresses of linseed i.e. Alternaria blight & linseed bud fly are of utmost significance. These promising genotypes are the prime candidates for active integration into further breeding initiatives, aimed at cultivated genotypes with desirable characteristics.

Furthermore, the strategic utilization of diverse facts found whether the germplasm lines to play a pivotal role in processes associated with registration, ongoing maintenance & safeguarding of linseed lineages. This underscores the importance of lapping into wade away of traits to drive advancements in linseed breeding ensuring its continued growth and protection for the future.

Table 1. Agronomic and morphological descriptors, descriptor scales or states, and frequency distribution for linseed accessions

Sr. No	Trait/Descriptor	Descriptor state	Class or scale of descriptor	Distribution by classes of descriptor
1	Early Plant Vigour (EPV)	It was recorded after 25 days of sowing	Poor	154
			Good	2228
			Very good	204
2	Corolla Colour (CC)	Observation of individual plants at 50% flowering stage was recorded	White	123
			Light blue	1137
			Blue	102
			Violet	1020
			Red-violet	7
			Red	1
3	Flower/Petal Aestivation (PA)	Observation of individual plants at 50% flowering stage was recorded before noon	Twisted	858
			Semi-Twisted	1052
			Valvate	533
4	Flower Shape (FS)	Observation of individual plants at 50% flowering stage was recorded. It was recorded before noon	Funnel	2407
			Star	-
			Disk	50
			Tubular	-
5	Anther Colour (AC)	Anthers of individual flowers at 50% flowering stage was recorded.	Cream	494
			Grey	46
			Violet	116
			Blue	1733
			Black	1
			Dull	-
6	Stamen Colour (SC)	Colour of distal part of filament of individual flowers at 50% flowering stage was recorded	White	726
			Violet	1097
			Blue	620
7	Flower Venation Colour (FVC)	Observation of individual plants at 50% flowering stage was recorded	White	93
			Light-violet	128
			Violet	358
			Blue	1808
			Red	1
			Red violet	1
8	Size of Corolla (mm)	It was measured as distance from petal to petal in five randomly selected individual plants at 50% flowering stage	Small (< 5cm)	99
			Large (>5 cm)	2369

Table 2. Characterization of linseed germplasm based on Distinctness (D),Uniformity (U) and Stability (S) as per DUS, UPOV 2011 during rabi 2020-21 and rabi 2021

Sr. No	Trait	Descriptor state	Class or scale	Distribution by classes of descriptor		Common germplasm for both the seasons	Trait specific Common germplasm lines
				2020-21	2021-22		
1	Days to 50% flowering	Number of days taken from the day of sowing to the day on which 50 % of the plants showed flowering was recorded	Early (<50 days)	299	60	31	Common germplasm lines for Earliness, Dwarf stature and High number of capsules^{-1} plant :IC0118855,IC0498442, IC0498490,IC0342800, EC41466,EC0541213, IC0096701,IC0203213, IC0096533, IC0525956
			Medium (50-60 days)	1181	1558	968	
			Late (>60 days)	806	1035	598	
2	Plant height	The height of plant from the base, to the tip of the main stem was recorded in centimetres	Dwarf (<50 cm)	1918	2625	1875	
			Medium (50-70 cm)	359	01	-	
			Tall (>70 cm)	9	-	-	
3	Total capsules per plant	It was recorded as total number of capsules	Low (<30)	61	25	1	
			Medium (30-60)	884	763	250	
			High (>60)	1341	1838	644	
4	No. of primary branches per plant	It was recorded visually by counting the number of primary branches of individual plants	Less (<5)	1821	2128	1165	-
			High (>5)	465	498	125	
5	Seed yield per plant	Recorded as seed yield of an individual plant in gram after harvest	Low (<2 gm)	1249	282	147	
			Medium (2-5 gm)	1005	2149	819	
			High (>5 gm)	32	195	-	
6	% Bud fly infestation	The bud fly infestation was recorded at dough stage by counting total number of floral buds and infested buds which was converted into % bud fly infestation	Resistant (0-10)	168	434	37	Common germplasm lines for Bud fly infestation and Alternaria blight resistance: IC0096551, IC0342801,IC620658, IC0118859, IC0498634, IC0526105,IC0498795, IC0499155,IC0498843, IC0599415,IC0498763, EC993391,EC993389, IC633096
			Moderately resistant (10.01-25)	1426	2110	1200	
			Moderately Susceptible (25.01-50)	676	81	24	
			Susceptible (50.01-75)	16	1	-	
			Highly Susceptible (>75)	-	-	-	
7	% Alternaria Blight	The alternaria blight infestation was recorded at dough stage by counting total number of floral buds and infected buds which was converted into % alternaria blight infestation	Resistant (0-10)	730	1250	344	
			Moderately resistant (10.01-25)	1445	1360	785	
			Moderately Susceptible (25.01-50)	102	16	1	
			Susceptible (50.01-75)	9	-	-	
			Highly Susceptible (>75)	-	-	-	

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