International Journal of Advanced Research in Biological Sciences ISSN: 2348-8069 www.ijarbs.com

DOI: 10.22192/ijarbs

www.ijarbs.com Coden: IJARQG(USA)

Volume 6, Issue 12 - 2019

Research Article

2348-8069

DOI: http://dx.doi.org/10.22192/ijarbs.2019.06.12.014

Performance of Advanced Breeding Lines of Soybean [*Glycine max* (L.) Merrill] at Mid Western Terai of Nepal.

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Abstract

An experiment of soybean was conducted at Grain Legumes Research Program Khajura, Banke, Nepal consisting sixteen genotypes during 2015 in randomized complete block design replicated thrice to observe agronomic performance, genetic variability, heritability and genetic advance using six ancillary characters and selection of early maturing, high yielding, disease resistant genotypes for mid western terai of Nepal using released variety Puja as standard check. Statistical analysis revealed all characters: days to fifty percent flowering, days to maturity, plant height, number of pods per plant, hundred seed weight, and grain yield were highly significant differences (<0.01) indicating sufficient genetic variability among the tested genotypes for concerned traits. High broad sense heritability coupled with high expected genetic advance as percent of mean were observed in grain yield (82 and 67.8%), number of pods per plant (83 and 60.3%), plant height (90 and 32.7%), hundred grain weight (87 and 33.4%), and days to fifty percent flowering (91 and 21%). All parameters obtained high phenotypic coefficient of variance indicating role of environment for the expression of the traits. Mean separation and clusturing showed genotype SBO115 is top yielder with highest number of pods and hanged separately in clusture 1, similarly, genotype TGX-311-23D and Puja were high yielder and having high number of pods and were statistically at par for grain yield, and genotype TGX1485-1D also high yielder were separated in same clusture 2. Total four clusters were obtained in eighty percent euclidean similarity clusturing indicating genetic closeness/distances among the genotypes.

Keywords: Genetic Advance, Heritability, Soybean, Variability, Clusturing

Introduction

Soybean (*Glycine max* [L.] Merrill) is a legume crop that grows from tropical, subtropical to temperate climatic regions. It is diploid (2n=2x=40) and is a selfpollinated species with less than 1% cross pollination. Vavilov (1949/50) considered Central Western China as the center of origin for soybean. It is believed that it might have been introduced to Africa in the 19th century by Chinese traders along the east coast of Africa. Soybean is a important grain and oil seed crop that is widely grown throughout the world, and provides more than 50% of the world's oilseed production (Wilson, 2008; Soy Stats, 2011). World's top seven countries are USA, Brazil, Argentina, China, India, Paraguay, and Canada with 123.66, 116.66, 59, 15.9, 11, 9 and 7.3 million metric ton production respectively in 2018/19 (https://www.worldatlas.com/articles/world-leaders-

in-soya-soybean-production-by-country.html).

Soybean ranks second and occupies an average of 7.2% (23563 ha) of the total legume's area and accounts for 7.7% (29061 Mt) of total legume production in Nepal (MOAD, 2018). It is Cultivated from terai to high hills 200-2000 m above the msl, can be grown successfully at an altitude ranging from 500 to 1500 m amsl either under intercropping with maize in upland condition and on paddy bund in lowland conditions (Neupane and Bharati 1990). Hill contributes about 83% of total soybean area and production, while mountain and terai occupies about 10% and 7% respectively (MOAD, 2018). Major production districts of soybean in Nepal are Doti, Kavre, Salyan, Makwanpur, Khotang. Used for food: baby food, dry seed roasted or fried, green pods as vegetable in Nepal and called poor man's meat because it contains >40% protein, around 20% oil.

It is important for both protein meal and vegetable oil in addition it is rich in lysine and vitamins and is used for both human and animal consumption as well as for industrial purposes, such as bio-fuels (Hartman et al., 2011). Apart from quality protein and oil soybean also has many therapeutic components viz., lactose free fatty acids, antioxidants like vitamins C, K, and D and folic acid, vitamins of B complex group viz., nicotinic acid ($23 \mu g/g$), pantothenic acid ($15 \mu g/g$), thiamine $(12 \mu g/g)$, pyridoxine $(8 \mu g/g)$, riboflavin $(3.5 \mu g)$ /g) and biotin (0.7 μ g /g) and isoflavones like genistein and daidzein (Mathur, 2004). It contains isoflavones which protect human body against the cancer, diabetes, osteoporosis, blood pressure, coronary heart disease etc (Pushpendra et al., 2017). Soybean is hardy crop grown in marginal land and stress environments.

Altogether eight varieties were released for cultivation till the date. Most of the soybean produced in the country is consumed internally either as a roasted beans or green pods as vegetables. Tarkari Bhatmas-1 is suitable as green fresh pod or seed soaked overnight and cooked or boiled or fried. Large quantity of soybean grain, flour, crude/refined oil and cake are imported to meet demand for oil and poultry industries. In 2016/17 imported amount was 212137.325 mt. with value of 1698.5356 crore nepalese rupees (MOAD, 2018).

Prior knowledge of genetic variability and characterization of germplasm available at the station has an important implication for future utilization to identify areas of major priority for conservation and improvement programs (Allard, 1960). Knowledge of level of genetic diversity available in germplasm is prerequisite for any breeding programs that genetic gain could not be limited due to the availability of narrow genetic variability between parental lines involved in crossing programs (Kumar et al., 2004). In the past years, breeders used morphological data and pedigree information to assess genetic diversity among the lentil varieties. Cluster analysis can be a good source to identify the variation in the germplasm and to classify based on similarity and dissimilarity index. This analysis is also useful for the selection of parents for the breeding program and crop modeling (El-Deeb and Mohamed, 1999; Jaynes et al., 2003).

Inheritance of quantitative characters is often influenced by variation in other characters which may be due to pleiotropy or genetic linkage. Knowledge of genetic variability is essential to formulate an effective crop improvement programme.

Main problems of soybean in Nepal are uncertain occurrence of pod blight in hill, yellow mosaic in terai, low priority for input, lack of flood tolerant varieties, lack of disease resistant varieties, low utilization of local germplasm in breeding program, so farmers are searching for more new varieties. The present study was, undertaken to identify a proper plant type for selection so as to improve the seed yield keeping in view the variability, genetic advance, clusturing and heritability of concerned studied parameter of soybean. Selection and advancement of early maturing, high yielding, disease resistant and drought/flood tolerance soybean genotypes for mid western terai of Nepal.

Materials and Methods

A field experiment was conducted at Grain Legumes Research Program (GLRP), Khajura, Banke, Nepal, during summer season of 2014/15 (July IIIrd week 2015 to November Ist week 2015). Geographically Grain Legume Research Program (GLRP) is located at Janaki VDC, Banke district. It lies between at 81^o 37" East longitudes and 28° 06" North lattitude and an altitude of 181 meters above mean sea level. Average annual rainfall of the station is 1000-1500 mm. However, delayed onset and early termination of monsoon rains is a regular feature, causing occasional failure of annual crops in the region. The maximum and minimum temperature at the station is 46°C and 5.4° C respectively, with relative humidity (R.H.) ranging between 27 % to 94 %. Humidity remains low in most parts of the year. Soils of the station have sandy to silty loam, poor in organic carbon and

available N but medium in available P_2O_5 and K_2O , pH varies from 7.2-7.5. Detail of weather parameters is given in Annex 1.

Year Month	8:45 NST Temperature (degree celcius)		Temp	5 NST perature e celcius)	Rain (mm)	Relative humidity (%)
	Max	Min	Max	Min		
Jul-15	35	27	35	29	27	73
Aug-15	33	26	34	28	18	83
Sep-15	34	25	34	29	5	78
Oct-15	32	20	32	25	15	81
Nov-15	29	14	29	20	2	85
Dec-15	24	9	23	12	-	96
Jan-16	22	7	22	10	2	96
Feb-16	27	11	28	15	2	85
Mar-16	33	15	33	23	3	54
Apr-16	40	18	40	29	-	29
May-16	37	24	37	29	12	47
Jun-16	35	26	35	28	39	81

Annex: 1 : Monthly agro-Meteorological Data of the Station during the period of research

Sixteen advanced breeding lines of soybean were used including one standard check released variety Puja for this study. The experiment was carried out in randomized complete block design in three replications. Each entry were planted having row-row distance of 50 cm with 4 x 3 m (12 m2) plot area. All recommended dose of manure and fertilizer (20:40:20 kg N:P₂O₅:K₂O and 6 ton FYM/ha) were applied as basal in soybean. All the recommended cultural practices were followed to raise the healthy crop. Days to 50% flowering and days to 90% maturity were recorded on the plot basis. Five random plants in each plot were used to record the plant height, number of pods per plant. Grain yield was taken from net harvested area of 12 m2 and 12% moisture percent was maintained while weighing and thousands grain weight was taken from same harvested lot.

Analysis of variance and mean separation among the lines was done with statistical software ADEL-R and clusuring of the genotypes was done with statistical software MINITAB. Estimation and categorization of expected genetic advance as percent of mean was done according to (Johnson et al., 1955). The heritability and genetic advance of the traits were calculated by the formula given by (Falconer, 1960). The Genetic advance as percent of mean (% mean GA) were classified as low, moderate, and high as follows: low <10%, moderate 10–20%, and high >20% and broad sense heritability values were ranked follows: low <30%, moderate 30–60%, and high >60% according to (Robinson et al., 1949).

Results and Discussion

Analysis of variance

Analysis of variance revealed that all characters days to 50% flowering (DFF), days to maturity (DM), number of pods per plant (NPPP), Pht (plant height), hundred seed weight (HSW) and grain yield kg/ha (Yld) were significant at (<0.01) indicating sufficient variation among the tested genotypes at genotypic level for concerned traits (Table 1). Breeder can utilize the variability for selection and may use in hybridization for trait improvement, gene transfer to the other genotypes. Similar result was reported by Baraskar et al., (2014) the analysis of variance revealed that mean squares due to genotypes were significant for all the 15 characters indicating varietal differences for all the characters studied. Aditya et al., 2011. reported similar findings in soybean as all studied characters were highly significant at (<0.01). Ibrahim et al 2018 reported similar findings as Number of Days to 50% flowering, Plan height. Chlorophyll content, Number of days to 95% maturity, No of pods per plot, No of pods per hectare were significant at (<0.01) level. Hossain et al., 2004. reported similar findings as Number of Days to 50% flowering, Plan height, NPPP, HSW and vld significant at (<0.01) level. Chandrawat et al., 2017 reported variation due to genotypes was significant for all the characters under study, similar to our finding. Sulistyo et al., 2018 showed similar result as DFF,

DM, Pht, NPPP, HSW and Yld all characters were highly significant differences for genotypic effect. Malek et al., 2014. also reported similar findings. Grain yield resembled highest variability (15.3) followed by number of pods per plant (13.3) followed by followed by hundred seed weight (6.3) and followed by plant height (5.3). Lowest variability was observed by days to maturity, then days to fifty percent flowering (Table 1).

Table 1. Mean squares, CV and F statistic of different agronomic traits of lentil genotypes tested at mid western terai of Nepal in 2015

Traits	Rep	Treat	Error	Fcal	CV	P value
DFF	1.0	85.7	2.6	32.7	3.1	**
DM	2.3	58.2	3.0	19.6	1.6	**
Pht	2.1	715.3	25.3	28.2	5.3	**
NPPP	73.1	1841.3	118.7	15.5	13.3	**
HSW	0.2	8.7	0.4	20.9	6.3	**
Yld	3438.5	293444.7	19799.6	14.8	15.3	**

Note; DFF: Days to 50% flowering, DM: Days to 90% maturity, NPPP: Number of pods per plant, Pht: Plant height (cm), HSW: Hundred seed wt (gm) and Yld: Grain yield (kg/ha)

Mean Separation

Separation of mean values of quantitative traits are given in Table 2. It was observed that standard check puja were earlier for days to 50% flowering (44 days) and genotype AGS-371 was earlier for maturity (106 days) followed by puja (106.3 days). While genotype G-4508 was late for both days to flowering (64.7 days) and AGS-87-4 was late for days to maturity (120 days). Obtained mean value of flowering and maturity were 51.5 and 110.4 days respectively. Genotypes

SBO115, TGX-311-23D, Puja and TGX1485-ID were high yielder with more number of pods 1520.7, 1315, 1321 and 1275.7 kg/ha, and 123.7, 109.7, 100.7 and 91 numbers respectively (Table 2). Genotypes Puja and IARS87-1 were with high hundred seed weight (13.2 and 12.5 gm), breeder can use these genotypes for respective traits for genetic improvement. Obtained mean value for grain yield, number of pods per plant and hundred seed weight were 918 kg/ha, 81.9 and 10.2 gm respectively.

Table 2. Mean separation of different agronomic traits of 16 soybean genotypes tested at mid western terai of Nepal in 2015

TN	Genotypes	DFF	DM	Pht	NPPP	HSW	Yld
1	SBO115	55.0 ^{bc}	106.0 ^e	111.3 ^{bc}	123.7 ^a	8.6 ^h	1520.7 ^a
2	TGX1903-1F	53.0 ^{cdef}	106.7 ^e	81.3 ^{hi}	102.0 bc	9.9 ^{efg}	1073.3 ^{cd}
3	G-4508	64.7 ^a	113.0 bc	113.7 ^b	113.7 ^{ab}	9.9 ^{efg}	$1008.7^{\ de}$
4	TGX1485-1D	51.7 defg	111.3 ^{cd}	98.3 ^{de}	91.0 ^{cd}	10.8 cde	1275.7 ^{bc}
5	G-1871	55.7 ^{bc}	108.7 ^{de}	104.0 ^{cd}	91.0 ^{cd}	10.0 ef	950.3 def
6	SBO 122	50.7 ^{efg}	108.7 ^{de}	93.0 ^{ef}	52.3 fg	9.5 ^{fgh}	529.7 $^{\rm hi}$
7	G-1873	49.7 ^{gh}	114.7 ^b	91.3 efg	48.7 ^g	11.9 ^{bc}	766.3 $^{\rm fg}$
8	TGX-311-23D	53.3 ^{cde}	107.7 ^e	108.0^{bc}	109.7^{ab}	8.7 ^h	1315.0 ^{ab}
9	IARS87-1	47.0 ^{hi}	115.7 ^b	78.0^{i}	47.7 ^g	12.5 ^{ab}	624.3 ^{ghi}
10	Puja	44.0 ^j	106.3 ^e	83.7 ^{ghi}	100.7 bc	13.2 ^a	1321.0 ^{ab}
11	AGS-371	50.3 fg	106.0 ^e	$87.0^{\text{ fgh}}$	87.3 ^{cd}	6.5 ⁱ	783.0 ^{efg}
12	AGS-376	49.7 ^{gh}	113.7 ^{bc}	96.3 ^{de}	73.0 ^{de}	11.6^{bcd}	759.3 $^{\rm fgh}$
13	KAVRE	57.0 ^b	107.0 ^e	131.0 ^a	80.0 ^{de}	8.9 ^{gh}	966.0 ^{def}

14	AGS-87-4	54.0 ^{cd}	120.0 ^a	98.3 ^{de}	75.7 ^{de}	9.5 ^{fgh}	652.7 ^{ghi}
15	LS77-16-16	44.3 ^{ij}	106.0 ^e	76.0 ⁱ	68.0 ^{ef}	$10.5 ^{\text{def}}$	625.7 ^{ghi}
16	PI94159	44.7 ^{ij}	115.0 ^b	76.3 ⁱ	46.3 ^g	11.9 ^b	516.7 ⁱ
Mean		51.5	110.4	95.5	81.9	10.2	918.0
Std MSE		1.6	1.7	5.0	10.9	0.6	140.711
LSD(5%)		2.7	2.9	8.4	18.2	1.1	234.637
Min		44.0	106.0	76.0	46.3	6.5	516.7
Max		64.7	120.0	131.0	123.7	13.2	1520.7

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Note; DFF: Days to 50% flowering, DM: Days to 90% maturity, NPPP: Number of pods per plant, Pht: Plant height (cm), HSW: Hundred seed wt (gm) and Yld: Grain yield (kg/ha)

Heritability, coefficients of variation and genetic advance

The genotypic, phenotypic variances, broad sense heritability and genetic advance for all the traits are given in Table 3. The observed phenotypic coefficient variance (PCV) was higher than the genotypic coefficient of variance (GCV) for all six quantitative traits indicating role of environment for the expression of characters. Higher the differences between the GCV and PCV higher the role of environment for character expression. Chandrawat et al., 2017 reported PCV was higher than GCV for all the studied characters in support to our finding. Our result resembled higher role of environment for number of pods per plant and grain yield.

The high broad sense heritability coupled with high expected genetic advance as percent of mean was observed for the traits, number of pods per plant (83 and 60.3), grain yield (82 and 67.8) hundred seed weight (87 and 33.4), plant height (90 and 32.7) and days to flowering (91 and 21) percentage respectively (Table 3). These characters can be considered as favorable attributes for the improvement through selection and this may be due to additive gene action could be improved upon by adapting selection without progeny testing. High heritability and low expected genetic advance as percent of mean was obtained in days to maturity 86 and 8 percentage respectively. This trait may be governed by additive and non additive gene action and selection for improvement may be difficult. Baraskar et al., (2014) reported similar findings, the magnitudes of genetic advance were observed to be high (> 20%) for number of clusters per plant, plant height, seed yield per plant, number of primary branches per plant, biological yield per plant and number of pods per plant. Besufikad (2018) reported high heritability coupled with high genetic advance as percent of mean for DFF (99.16 and 47%), Pht (99.26 and 58.48), NPPP (97.66 and 53.29%), HSW (91.86 and 37.48%) and Yld (64.66 and 67%), he also reported high heritability and low genetic advance as percent of mean for DM (99.4 and 15.28%) which was in full support to our findings in soybean. Ibrahim et al 2018 also reported high heritability for DFF, Pht, DM, and NPPP. Hossain et al., 2004. reported high heritability and high genetic advance as percent of mean for Number of Days to 50% flowering, Plan height, NPPP, HSW and yld which was in full support to our findings. Chandrawat et al., 2017 reported high heritability coupled with high genetic advance as percent of mean for Pht (90.9 and 44%), NPPP (91.71 and 67.05), HSW (83.85 and 29.08%), and Yld (85.88 and 37.77%), he also reported high heritability and low genetic advance as percent of mean for DM (61.78 and 5.51%) which was in full support to our findings in soybean. His reporting have only one contradict that medium genetic advance as percent of mean for DFF. Sulistyo et al., 2018 reported similar findings as high heritability for DFF, DM, Pht, NPPP, HSW and Yld in soybean. Malek et al., 2014. also reported similar findings.

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Traits	Mean	Std	Vg	Vp	Н	GCV	PCV	GA	%mean GA
DFF	51.5	5.4	27.7	30.3	0.91	10.2	10.7	10.8	21.0
DM	110.4	4.5	18.4	21.4	0.86	3.9	4.2	8.8	8.0
Pht	95.5	15.6	230.0	255.3	0.90	15.9	16.7	31.2	32.7
NPPP	81.9	25.8	574.2	692.9	0.83	29.3	32.1	49.4	60.3
HSW	10.2	1.7	2.8	3.2	0.87	16.2	17.4	3.4	33.4
Yld	918.0	326.2	91215.1	111014.6	0.82	32.9	36.3	622.2	67.8

Table 3. Standard deviation, variance, broad sence heritability, PCV, GCV and percent mean genetic advance for quantitative traits of soybean genotypes

Note; DFF: Days to 50% flowering, DM: Days to 90% maturity, NPPP: Number of pods per plant, Pht: Plant height (cm), HSW: Hundred seed wt (gm) and Yld: Grain yield (kg/ha)

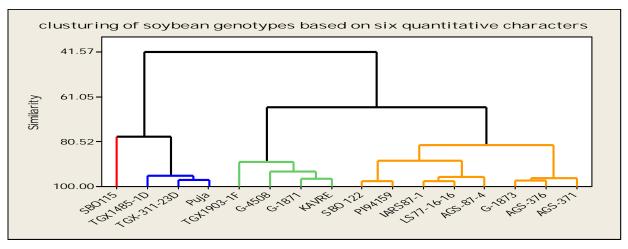
Clusturing

Dendrogram generated based on Unwaited Pair Group Method with Arithmetic Mean (UPGMA) clustering method and Euclidean similarity coefficient among sixteen soybean genotypes is given in table 4 and figure 1. Clusturing was done with UPGMA clustering method with 80% eucdian similarity coefficient and four clustures were generated. Clusture 1 contains single genotype SBO115 which is highest yielder and with maximum number of pods per plant. Breeber can use earliness and seed weight from the genotypes of this clusture. Clusture II carries 4 genotypes which have slight higher values for grain yield and NPPP than grand centroid. Clusture III represents 3 genotypes including puja which are superior than grand centroid for grain yield, HSW and NPPP. Similarly, clusture IV carries remaining eight genotypes they are inferior grain yield and NPPP. We can select clusture 1 and 3 for large plot demonstration and/or hybridization program.

Table 4. The average of traits for each cluster obtained from UPGMA cluster analysis

Variables	Cluster1	Cluster2	Cluster3	Cluster4	Grand Centroid
DFF	55	57.6	49.67	48.8	51.55
DM	106	108.85	108.43	112.475	110.406
Pht	111.3	107.5	96.67	87.025	95.469
NPPP	123.7	99.175	100.47	62.375	81.925
HSW	8.6	9.683	10.91	10.488	10.248
Yld	1520.7	999.575	1303.9	657.213	918.025

Note; DFF: Days to 50% flowering, DM: Days to 90% maturity, NPPP: Number of pods per plant, Pht: Plant height (cm), HSW: Hundred seed wt (gm) and Yld: Grain yield (kg/ha)





Conclusion

Obtained results indicates presence of sufficient genetic variability for the studied traits and soybean genotypes are suitable for breeding purpose. Result resembled high broad sense heritability coupled with high genetic advance for grain yield, number of pods per plant, plant height, hundred grain weight, and days to fifty percent flowering shows these traits maybe governed by additive gene effects and selection of these traits would be more effective for genetic improvement. Mean separation and clusturing showed genotypes SBO115, TGX-311-23D, Puja and TGX1485-ID are superior among the tested genotypes will be selected for further evaluation in advance trial for mid western terai of Nepal. Selection for grain improvement should be conducted vield bv simultaneous selection of many traits such as number of pods per plant, and higher grain weight.

Acknowledgments

The authors would like to thank NARC management for managing budget to conduct research. We would like to thank technical assistant staffs of Grain Legumes Research Program (GLRP), Khajura, Banke for their support on conducting experiment on the field.

Conflict of Interest

The authors declare that there is no conflicts of interest regarding publication of this manuscript.

Authors contribution

N. H. Ghimire: Designed and performed experiment, analyzed data and wrote the paper.

R. A. Sah: Performed experiment, data recorded

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How to cite this article:

Netra Hari Ghimire, Rabendra Prasad Sah. (2019). Performance of Advanced Breeding Lines of Soybean [*Glycine max* (L.) Merrill] at Mid Western Terai of Nepal. Int. J. Adv. Res. Biol. Sci. 6(12): 118-125. DOI: http://dx.doi.org/10.22192/ijarbs.2019.06.12.014