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Research Article

Genetic divergence of selected genotypes in Sunhemp (*Crotalaria juncea* L.)

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Abstract
A total of 40 genotypes of sunhemp maintained at the Centre for Plant Breeding and Genetics, Tamil Nadu Agricultural University, Coimbatore were studied during Rabi 2017 for genetic divergence. The D^2 analysis showed that on 50 DAS maximum contribution to divergence was given by green matter yield followed by leaf breadth and number of nodules per plant. The genotypes were grouped into 10 diverse clusters with farthest being cluster 10 and cluster 9. Cluster 7 had highest intra cluster distance. On 65 DAS, the character that contributed highest to divergence was green matter yield followed by number of nodules per plant and number of pods per inflorescence. 11 different clusters were formed among the 40 genotypes based on critical D^2 value. Inter cluster distance was high between cluster 11 and cluster 6. Cluster 11 had the highest intra cluster distance. Hence, Genotypes from these clusters can be used for hybridization purpose.

Keywords
Crotalaria juncea, D^2 Analysis, Sunhemp, Green manure.

Introduction

Sunhemp (*Crotalaria juncea* L.), belonging to the family *Papilionaceae*, sub-family papilionoideae and the tribe *Crotalariea* of order *Leguminosae* (Van Wyk and Schutte, 1995). It is multipurpose legume mainly for its soft, slightly lignified fibre, in many countries notably India. In India, it is also grown for green manure, as a soil improver. The crop is grown also for legume as well as fodder purpose. The genus name "*Crotalaria*" means 'rattle', which refers to the noise made by the seeds shaken in the mature pods. Species of this genus are wide spread throughout tropical, subtropical and to a lesser extent temperate countries.

Sunhemp is a warm-season annual that grows upright with a height of 4 to 6 feet in 60 to 90 days. It has simple, oblong-shaped leaves that are 2.5 to 5.0 inches long. Branching occurs about 2 feet from the ground or higher, if planted in a thick stand as a green manure crop. The plant has a strong taproot and well-developed lateral roots with branched and lobed nodules.

Crotalaria species are distributed worldwide, but their germplasm has not been well collected. In *Crotalaria* genus, there are approximately 600 species, but there are only 242 accessions in the USDA collection representing about 30 species. More accessions needs to be curated from different regions of the world. In India, a total of 122 sunhemp germplasm are being maintained at Sunhemp Research Station, Pratapgarh in Uttar Pradesh. In National Bureau of Plant Genetic Resources, over 400 accessions of sunhemp and wild species of *Crotalaria* are maintained as seeds and herbarium specimens (Malik and Srivastava, 2006).

Material and Methods

The present investigation was carried out at the Centre for Plant Breeding and Genetics, Tamil Nadu Agricultural University, Coimbatore during 2017-2018. The average annual rainfall received at this location is around 700 mm. A total of 40 germplasms were raised in Randomized Complete Block Design with three replications. Each accession was raised in 1.5m x 1.0m sized plot with 8 x 8 cm dense spacing followed for use as green manure crop. The observations were recorded on 50 DAS from five randomly selected plants from each entry/replication for 13 characters *viz.*, plant height (cm), stem girth (mm), number of primary branches/ plant, number of secondary branches/ plant , number of leaves/ plant, leaf length (cm), leaf breadth (cm), number of flowers/ plant, number of nodules/ plant, green matter yield (g/plant), dry matter yield (g/plant), total nitrogen content (%), N accumulation (g/plant) and on 65 DAS 17 characters *viz.*, days to first flowering, days to 50% flowering, plant height (cm), stem girth (mm), number of primary branches/ plant, number of secondary branches/ plant, number of leaves/plant, leaf length (cm), leaf breadth (cm), number of flowers/ plant, number of nodules/ plant, number of pods/ inflorescence, number of pods/ plant, green matter yield (g/plant), dry matter yield (g/plant), total nitrogen content (%), and N accumulation (g/plant). Mahalanobis D^2 analysis (Mahalanobis, 1936) was used to estimate genetic divergence among the 40 genotypes. Grouping of genotypes into clusters was carried out by Tocher's methods (Rao, 1952).

Results and Discussion

Genetic improvement in the crop plant depends on variability present in that crop and thereafter selection of parental line for hybridization programme. In this context, the study on genetic divergence is of vital importance for any plant breeding programme, which aims at genetic improvement and productivity of that plant species. The analysis of variance indicated that all the characters showed significant difference. (Table 1 & 2). Based on D^2 analysis, the 40 genotypes were grouped into 10 clusters at 50 DAS (Table 3) and 11 clusters on 65 DAS (Table 4) for the characters studied (Praveen *et al.,* 2013). The clustering pattern indicated the absence of relationship between genetic diversity and geographical origin of genotypes on both 50 DAS and 65 DAS (Badhe *et al.* 2015).

Among the 13 characters studied on genotypes during 50 DAS, higher divergence was contributed by green matter yield (Bedassa and Eshete, 2013) followed by leaf breadth and number of nodules per plant and number of leaves per plant, whereas plant height and stem girth contributed least to divergence (Table 5). Out of 10 clusters, cluster 7 had the highest intra cluster distance suggesting that the genotypes, within these clusters were divergent. While inter cluster distance was higher between cluster 10 and cluster 9, followed by cluster 7 and cluster 3. Hence, genotypes under these clusters are diverse (Table 5). Similar results were given by Srinivas *et al*. (2016). Hence, crosses can be made between the genotypes present in these clusters for the improvement of green matter yield. Regarding cluster means (Table 6), Cluster 6 exhibited the highest mean value for green matter yield (14.8g), secondary branches per plant (4.25), leaf length (8.94cm) and leaf breadth (2.14cm). cluster 8 exhibited the highest mean value for plant height (101.82cm), number of primary branches per plant (4.01), total nitrogen content (3.6%), nitrogen accumulation (0.097g/plant), Cluster 10 showed the highest mean value for dry matter yield (3.107g). Cluster 3 exhibited low mean values for number of primary branches (2.40), leaf breadth (1.063cm), stem girth (2.78mm), green matter yield (10.48g) and number of leaves per plant (16.85).

At the ages of both 50^{th} & 65^{th} day of the 40 sunhemp genotypes studied, green matter yield (Bedassa and Eshete, 2013) contributed to high divergence followed by number of nodules per plant and number of pods per inflorescence (Table 6). Out of 11 clusters, cluster 11 had the highest intra cluster distance suggesting that genotypes, within this cluster were more divergent. Inter cluster distance was high between cluster 11 and cluster 6 followed by cluster 6 and cluster 9. The genotypes of these clusters were inferred to be diverse (Table 7). Hence, crosses can be made between genotypes present in these clusters. Similar results were furnished by Sidhu *et al.* (2004). On 65 DAS cluster 6 exhibited the highest mean value for days to fifty per cent flowering (53.15), days to first flowering (46.76), number of leaves per plant (42.86), green matter yield (29.27g), stem girth (5.28mm), number of primary branches (4.46), leaf breadth (2.99cm) and nitrogen accumulation (0.213g/plant). Cluster 1 exhibited highest value for number of flowers per plant (23.94) and leaf length (9.06cm), total nitrogen content (2.43%). Cluster 3 was found to be highest value for number of pods per plant (36.28), number of secondary branches per plant (6.23) (Table 8-10). Results of overall cluster means suggested that the selection of genotypes from the clusters 6, 7, 8 and 10 on 50 DAS and from clusters 1, 3, 6, 9 and 11 from 65 DAS for further breeding work may yield better recombinants.

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Table 1. Analysis of variance for different characters at 50th day observation in Sunhemp crop

	PH	SG	NBP	NBS	NLP	LL	LB	NFP	NNP	TNC	NA	DMY	GMY
GENOTYPE	795.85**	1.19	1.72**	1.50**	145.80**	2.17**	0.39**	121.55**	50.42**	1.65**	0.00**	1.12**	17.66**
ERROR	36.74	0.07	0.06	0.08	4.48	0.32	0.02	2.01	0.78	0.06	0.00	0.04	0.83

Table 2. Analysis of variance for different characters at 65th day observation in Sunhemp crop

	DF	DFF	PH	SG	NBP	NBS	NLP	LL	LB	NFP	NNP	NPI	NPP
GENOTYPE	115.71**	124.98**	636.93**	1.37**	1.48**	1.93**	8.08	2.99**	0.56**	592.98**	47.05**	3.56**	326.48**
ERROR	10.48	12.75	72.02	0.10	0.07	0.11	0.44	0.40	0.02	2.15	0.41	0.04	6.66

Table 2. continued.

	TNC	NA	DMY	GMY
GENOTYPE	0.44**	0.01**	5.71**	79.89**
ERROR	0.02	0.00	0.15	1.74

** = Significant at 1% level * = Significant at 5% level

DF – Days to first flowering
DFF – Days to 50% flowering
PH – Plant height
SG- Stem girth
NPB – Number of primary branches/plant
NSB – Number of secondary/plant
NLP – Number of leaves/plant
LL – Leaf length
LB – Leaf breadth
NFP – Number of flowers/plant
NNP – Number of nodules/plant
NPI – Number of pods/infloresence
NPP – Number of pods/plant
TNC – Total nitrogen content
NA – Nitrogen accumulation
DMY – Dry matter yield
GMY – Green matter yield

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Table 3. Distribution of 40 genotypes into different clusters on 50th day observation based on D^2 analysis

Cluster No.	Number of Accessions	Genotypes
I	8	TNCJ 1, TNCJ 2, TNCJ 3, TNCJ 4, TNCJ 5, TNCJ 6, TNCJ 22, TNCJ 33
II	2	TNCJ 36, TNCJ 38
III	2	TNCJ 17, TNCJ 19
IV	2	TNCJ 21, TNCJ 34
V	2	TNCJ 35, TNCJ 39
VI	2	TNCJ 32, TNCJ 37
VII	8	TNCJ 7, TNCJ 8, TNCJ 9, TNCJ 10, TNCJ 11, TNCJ 12, TNCJ 13, TNCJ 14
VIII	3	TNCJ 15, TNCJ 26, TNCJ 31
IX	10	TNCJ 16, TNCJ 18, TNCJ 20, TNCJ 23, TNCJ 24, TNCJ 25, TNCJ 27, TNCJ 28, TNCJ 29, TNCJ 30
X	1	TNCJ 40

Table 4. Distribution of 40 genotypes into different clusters on 65th day observation based on D^2 analysis

Cluster No.	Number of Accessions	Genotypes
I	4	TNCJ 1, TNCJ 2 , TNCJ 26, TNCJ 27
II	2	TNCJ 21 , TNCJ 30
III	2	TNCJ 10, TNCJ 31
IV	2	TNCJ 36, TNCJ 40
V	2	TNCJ 34, TNCJ 39
VI	2	TNCJ 12, TNCJ 13
VII	2	TNCJ 24, TNCJ 32
VIII	2	TNCJ 17, TNCJ 33
IX	16	TNCJ 3, TNCJ 4, TNCJ 5, TNCJ 6, TNCJ 7, TNCJ 8, TNCJ 9, TNCJ 11, TNCJ 14 , TNCJ 15, TNCJ 16, TNCJ 18, TNCJ 19, TNCJ 20, TNCJ 22, TNCJ 23
X	2	TNCJ 35, TNCJ 38
XI	4	TNCJ 25, TNCJ 28, TNCJ 29, TNCJ 37

Table 5. Percent contribution of morphological characters and biochemical characters on 50th day towards divergence

S.No	Character	Times ranked 1st	% Contribution
1	Plant height	0	0.00
2	Stem girth	0	0.00
3	No. of primary branches	28	3.59
4	No. of secondary branches	44	5.64
5	No. of leaves per plant	73	9.36
6	Leaf length	42	5.38
7	Leaf breadth	77	9.87
8	No. of nodules per plant	75	9.62
9	No. of flowers per plant	64	8.21
10	Total nitrogen content	39	5.00
11	Nitrogen accumulation	12	1.54
12	Dry matter yield	68	8.72
13	Green matter yield	258	33.08
14	Total	780	100

Table 6. Percent contribution of morphological characters and biochemical characters on 65th day towards divergence

S.No	Character	Number of times ranked first	% Contribution
1	Days to first flowering	0	0.00
2	Days to 50% flowering	29	3.72
3	Plant height	4	0.51
4	Stem girth	14	1.79
5	No. of leaves per plant	0	0.00
6	Leaf length	31	3.97
7	Leaf breadth	62	7.95
8	No. of primary branches	63	8.08
9	No. of secondary branches	36	4.62
10	No. of nodules per plant	70	8.97
11	No. of pods/ inflorescence	69	8.85
12	No. of flowers per plant	62	7.95
13	No. of pods per plant	2	0.26
14	Total nitrogen content	50	6.41
15	Nitrogen accumulation	46	5.90
16	Dry matter yield	61	7.82
17	Green matter yield	181	23.21
18	Total	780	100

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Table 7. Inter and intra (bold) cluster D^2 values of 50th day observation.

CLUSTERS	I	II	III	IV	V	VI	VII	VIII	IX	X
I	**52.74**	54.55	55.60	48.33	57.78	60.40	59.79	58.62	61.58	65.53
II		**28.60**	51.51	42.09	48.64	54.75	63.71	60.07	58.81	61.14
III			**29.89**	45.96	51.31	69.69	70.75	55.75	66.16	64.25
IV				**31.33**	46.17	53.99	55.49	55.31	55.10	53.83
V					**31.33**	45.61	57.52	53.96	58.73	57.55
VI						**32.77**	60.30	65.38	61.54	69.77
VII							**60.74**	61.97	65.30	62.96
VIII								**45.85**	62.82	68.40
IX									**60.54**	70.81
X										**0.00**

Table 8. Inter and intra (bold) cluster D^2 values of 65th day observation.

CLUSTERS	I	II	III	IV	V	VI	VII	VIII	IX	X	XI
I	**59.32**	55.82	66.06	64.06	67.28	69.44	65.91	57.50	69.97	64.71	74.08
II		**30.47**	57.05	50.20	43.82	60.19	53.65	49.94	60.65	44.05	70.75
III			**31.58**	59.76	61.21	66.24	66.83	57.78	69.97	52.11	72.58
IV				**35.76**	44.20	65.33	62.85	60.52	66.04	55.35	65.12
V					**35.81**	64.71	57.53	56.37	67.23	48.05	64.11
VI						**41.50**	75.03	68.89	73.16	69.98	78.62
VII							**42.27**	60.70	68.00	66.54	74.05
VIII								**44.17**	69.37	56.74	67.66
IX									**73.57**	66.45	77.94
X										**47.20**	72.97
XI											**74.85**

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Table 9. Cluster means of 50th day morphological observation based on D^2 analysis

	PH	SG	NPB	NSB	NLP	LL	LB	NFP	NNP	TNC	NA	DMY	GMY
I	70.94	3.13	3.04	4.03	28.87	7.16	1.81	22.39	13.87	3.49	0.09	2.41	11.28
II	89.75	3.87	3.63	3.61	19.75	6.90	1.47	17.32	8.00	2.51	0.08	2.89	11.28
III	76.98	2.79	2.40	3.74	16.86	7.08	1.06	26.46	7.70	2.64	0.08	2.47	10.05
IV	86.40	3.17	2.80	2.83	20.65	6.94	1.41	19.20	8.30	2.19	0.06	2.43	10.87
V	97.69	3.64	3.78	4.19	21.69	8.07	1.53	21.65	5.99	2.06	0.05	2.95	12.96
VI	93.34	3.67	3.94	4.25	22.33	8.95	2.15	19.88	7.60	2.52	0.09	3.63	14.80
VII	79.69	3.78	3.77	4.15	33.30	7.94	2.10	17.34	11.86	3.12	0.08	2.62	13.33
VIII	101.82	3.86	4.02	4.15	32.42	8.22	1.87	24.09	10.49	3.61	0.10	2.84	11.74
IX	94.05	3.29	3.23	3.47	28.89	7.47	1.64	13.92	12.02	3.33	0.09	2.71	11.06
X	86.42	3.70	3.33	2.47	17.88	6.81	1.56	11.01	6.87	2.17	0.08	3.11	17.10

PH – Plant height (cm)
SG - Stem girth (mm)
NPB – Number of primary branches/plant
NSB – Number of secondary/plant
NLP – Number of leaves/plant
LL – Leaf length (cm)
LB – Leaf breadth (cm)
NFP – Number of flowers/plant
NNP – Number of nodules/plant
TNC – Total nitrogen content (%)
NA – Nitrogen accumulation (g/plant)
DMY – Dry matter yield (g)
GMY – Green matter yield (g)

Table 10. Cluster means of 65th day morphological observation based on D^2 analysis

	DF	DFF	PH	SG	NPB	NSB	NLP	LL	LB	NFP	NNP	NPI	NPP	TNC	NA	DMY	GMY
I	44.77	49.71	102.12	4.39	4.14	4.97	35.12	9.07	2.16	23.95	10.40	1.57	13.49	2.43	0.18	6.10	21.38
II	37.73	42.20	110.38	3.91	3.13	3.57	26.97	7.55	1.73	0.00	4.69	3.53	29.76	1.79	0.10	3.92	12.75
III	46.19	50.27	119.51	4.87	4.18	6.23	24.81	9.05	2.47	0.00	8.28	3.10	36.28	2.19	0.16	5.08	17.52
IV	37.37	41.24	102.84	4.36	2.23	3.58	35.74	7.42	1.77	13.36	4.74	2.35	16.92	1.84	0.09	5.52	17.25
V	38.76	43.14	115.85	4.36	2.63	3.92	24.61	7.87	1.80	0.00	3.78	2.20	17.04	1.37	0.06	3.69	11.63
VI	46.77	53.15	112.93	5.28	4.47	4.85	42.87	8.99	3.00	19.50	9.95	2.14	21.59	2.12	0.21	7.74	29.27
VII	41.01	45.20	100.70	3.34	2.78	3.94	28.35	8.75	2.20	0.00	4.64	2.68	20.15	2.09	0.14	3.92	12.77
VIII	35.89	41.28	113.83	3.77	3.74	4.69	26.23	7.60	1.77	0.00	5.20	2.08	20.76	1.98	0.08	4.15	11.70
IX	43.66	48.00	113.87	4.12	3.74	4.62	27.86	8.51	2.35	18.85	7.95	2.88	26.35	2.19	0.14	5.10	17.28
X	40.35	44.24	124.86	4.92	3.71	4.95	32.10	8.42	1.91	0.00	2.63	3.31	31.79	1.61	0.09	3.67	11.59
XI	41.10	45.83	124.58	4.42	3.42	4.74	19.80	9.03	2.29	12.70	10.99	2.15	19.68	2.29	0.10	4.95	14.50

DF – Days to first flowering
DFF – Days to 50% flowering
PH – Plant height (cm)
SG- Stem girth (mm)
NPB – Number of primary branches/plant
NSB – Number of secondary/plant
NLP – Number of leaves/plant
LL – Leaf length (cm)
LB – Leaf breadth (cm)
NFP – Number of flowers/plant
NNP – Number of nodules/plant
NPI – Number of pods/infloresence
NPP – Number of pods/plant
TNC – Total nitrogen content (%)
NA – Nitrogen accumuation (g/plant)
DMY – Dry matter yield (g)
GMY – Green matter yield (g)