Journal of Sugarcane Research (2020) **10**, 121-139
https://doi.org/10.37580/JSR.2020.2.10.121-139

RESEARCH ARTICLE

Wild Sugarcane - *Saccharum* sp. germplasm collection in the states of Punjab and Haryana, India

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(Received 10 July 2020; accepted 30 November 2020)

Abstract
The Indian sub-continent harbours a great wealth of diversity of *Saccharum* and its related genera. Several targeted expeditions were conducted to collect the range of diversity present in *Saccharum* complex from diverse parts of the country. But the north western states of Punjab and Haryana were hitherto unexplored. A targeted exploration was conducted during 2016 to collect the entire range of variability present in *Saccharum* complex. Collections were made from a wide range of habitats and altitudes. The *Saccharum spontaneum* was present mostly along the river banks and river beds and showed considerable variability for morphological characteristics. *S. spontaneum* was distributed throughout both the states and collections could be made from all the districts and union territory of Chandigarh. A total of 104 accessions comprising of 100 *Saccharum spontaneum* and four *Erinathus* sp. have been collected. They were occurring mostly as small population except in the canal bunds where larger population was noticed. Plant height ranged from 0.46 m to 4.5 m. Variation for leaf and stem characters were observed from the collections. Statistical analysis of altitude of the collection sites with plant characters showed no significant association. Correlation analysis among plant characters showed the highest correlation coefficient between plant height and internode thickness. Clustering of Haryana, Chandigarh and Punjab *Saccharum spontaneum* collections through complete linkage clustering resulted in five clear clusters among the collections. Geographic distance and environmental factors including local climates interplay in the formation of the genetic structure and geographic patterns of the native *S. spontaneum* populations. Haryana and Punjab state were not much explored during earlier period and hence the collection would add more diversity to the germplasm maintained. In addition to the 100 *S. spontaneum*, four *Erianthus* sp. were collected during the exploration which will enrich the gene pool.

Keywords: Sugarcane; Germplasm; *Saccharum spontaneum*; *Erianthus* sp.; Exploration; Distribution; Diversity; India

Introduction

Exploration and collection of natural diversity present in the crop gene pool in search of novel genes and alleles has always fascinated the researchers. Wild relative of crop plants constitute a part of crop gene pool which possesses genes that have great potential for their utilization programs. The collection of plant genetic resources primarily aims at tapping germplasm variability in different agri-horticultural plants, their wild relatives and related species. The Indian gene centre harbours about 166 species of native cultivated plants (Vavilov 1949–1950; Zeven and de Wet 1982) and over 320 wild relatives (Arora and Nayar 1984; Arora 1991, 2000). The wild relatives of crop plants by and large occur as components of disturbed habitats within the major vegetation types with distribution in the warm humid tropical, sub-tropical regions and in the Western Himalaya with low representation in the drier parts of north-western region (Arora and Nayar 1984; Arora and Pandey 1996; Arora 2000). Over 100 wild relatives and related taxa and endemic/rare/endangered species occur predominantly in the hot spot / micro-centres of India (Nayar 1996; Pandey and Arora 2004; Pandey et al. 2005). The germplasm so collected reveals the nature and extent of variability in different species, within species,

cultigens, etc. as also their agro-ecological/phyto-geographical distribution. It gives an opportunity to acquire knowledge on diversity in crop plants vis-a-vis its distribution to tap target areas and/or target species and the variability contained thereof. The genetic variability provided by the wild species and weed races is a source of resistance to diseases and insects, controlled mainly by major genes, tolerance for extreme environments such as salinity, desiccation, waterlogging, and frost and high vegetative vigour.

Sugarcane *Saccharum* spp. is the second most important industrial crop of India with a production area of 5m ha and production of 328.09 lakh tons (till June 2018-19). India is also the center of origin of *Saccharum spontaneum* the wild relative of cultivated sugarcane. The wild related species *S. spontaneum* has contributed immensely in the development of modern sugarcane hybrids and are a valuable source of novel alleles for disease resistance and climate resilience. *Saccharum* germplasm is widely distributed across the country across the geographical locations starting from below sea level to the foothills of Himalaya. India is a major centre of diversity for *Saccharum* genetic resources. *Saccharum spontaneum*, *Erianthus*, *Miscanthus*, *Narenga* and *Sclerostachya* species are wild relatives of *Saccharum* and are abundant in the country. Thus, barring *S. officinarum*, *S. barberi* and the New World species of *Erianthus*, most other members of the *Saccharum* complex are present in the Northern parts of India. Plant taxonomists are engaged in collecting information about diversity and distribution of plants throughout the world (Qureshi et al. 2011).

The collection conservation and utilization of *Saccharum* diversity has been the mandate of ICAR sugarcane breeding institute ever since its inception. A number of exploration have been carried out to the endemic regions of sugarcane diversity to collect the range of variation present in *Saccharum* and related genera. An effort was made to explore the hitherto unexplored areas of north western India; the states of Haryana and Punjab during September, 2016 to collect the *Saccharum* diversity.

Exploration activity for germplasm collection was started by the institute as early in 1912. Since then several explorations conducted by the Institute for the collection of *Saccharum* germplasm from different geographical and climatic regimes of the country (Nair et al. 1991, 1993; Nair and Somarajan 2003; Nair and Vigneswaran 2005; Nair et al. 2006; Nair and Sekharan 2009; Sreenivasan et al. 1985, 1986). During the earlier period from this Institute, exploration work was taken up R.R. Panje and S.K. Mukherjee during 1948 to 1957 and as per record seven *Saccharum spontaneum,* one *Erianthus arundinaceous* and one *Erianthus ravennae* were collected and established. During the year 1998, one *Erianthus arundinaceous* and one *Erianthus ravennae* were collected from Haryana and were established here. In order to strengthen the genetic resources collection further, during the recent past efforts are made to collect sufficient samples to represent the entire range of variability in the area during second to fourth week of September 2016 coinciding with the targeted species in the region.

Materials and Methods

Haryana is a landlocked state in northern India. It is located between 27°37’ to 30°35’ N latitude and between 74°28’ and 77°36’ E longitude. The altitude varies between 700 to 3600 ft. (200 meters to 1200 meters) above sea level. The climate is hot in summer and cold in winters. Rainfall varies like the Shiwalik Hills region being the wettest and the Aravalli Hills region being the driest. About 80% of the rainfall occurs in the monsoon season. The geographical area of the state is 44212 sq. km which is 1.3% of India’s geographical area. The

biodiversity activity are dispersed over rugged Shiwalik Hills in north, Aravalli hills in south, sand dunes in west and wastelands, saline-alkaline lands and waterlogged sites in the central part of the state. Haryana is primarily an agriculture state with almost 80% of its land under cultivation. Haryana is agriculturally developed state and the cropping intensity is around 181%. The major food crops cultivated in the state are rice, bajra and maize in kharif season, wheat and pulses in Rabi season. Sugarcane, cotton and oilseed are the other major commercial crops. Yamuna river flows through the state hence the riverbeds, river fed canals and canal fed agricultural fields are the major sources of *S. spontaneum* diversity.

Punjab is the food basket of India which produces nearly 12% of the total food grain production of India. Wheat, rice and maize are the major food crops and sugarcane and cotton are the major commercial crops. The State is a part of two major river basins namely, the Ganga and the Indus. Canals are the most popular and important source of irrigation in the Sub-Region due to easy supply and regular flow of water to the agriculture fields. In the inland drainage basin, the drains excavated are made to outflow in canals through lift pumps. Three major rives viz., Beas, Sutlej and Ravi flows in the states which facilitated the distribution of *Saccharum spontaneum* throughout the state. These major states were not explored fully and during the present exploration all districts of the states were explored and a large number of collections were made. Except some parts of Pathankot district which is in the hilly areas (up to 682 MASL), all other districts are in the plains.

Collections are made randomly to represent the overall variation, while specific clones are also collected selectively if they represent sought after types. For *S. spontaneum*, clumps and 4 - 5 stem cuttings are collected, while for *Erianthus* species only clumps are collected. Passport data on individual clones are recorded at the collection site. Data on plant height, thickness, leaf length, leaf width, canopy and flowering characteristics are recorded. Detailed passport data on location of collection, altitude, longitude, latitude, soil type, topography, soil color, soil texture, sample type, sampling method, associated vegetation and habitat were recorded onsite along with important descriptor traits. Altitude and latitude of the germplasm collection sites were recorded with a 12 channel receiver Gramin GPS12 personal navigator. Collected materials are cleaned, labeled, rolled in several layers of moist corrugated paper sheets to withstand handling during transit packed in polybags and over that slightly moist muslin cloth bag, on the same day and dispatched to ICAR-SBI, Coimbatore, Tamil Nadu by air cargo.

These accessions were planted in single replication on raised beds, with 90 cm spacing. Planting was in a zig-zag manner to prevent spread of vegetative propagules like runners between them. The crop was irrigated till the onset of monsoon upto first week of June. Earthing-up was carried out for each accession to ensure proper support and restrict spreading of runners. The accessions were completely randomized irrespective of their states (regions) of collection and growth habit. There was no apparent variation in the soil fertility in the experimental layout though no soil test was carried out during the experiment and 166:88:90(Kg/ha) NPK was applied in addition to application of lime @ 750 Kg/ha. All the package of practices as recommended for sugarcane cultivation were followed. These collections were characterized for seven quantitative (Table1) descriptors to assess the extent of variability among these clones and also to have reference database for maintaining the identity of clones in the field gene bank. Descriptors as elaborated in the publication by Artschwager and Brandes (1958) and Sreenivasan et al. (2001) were used.

Data were recorded on all collections during the post monsoon period. Individual traits were recorded one trait at a time so that the observations on all clones were completed for a specific trait within one or two days. The matured canes, which were present in the center of the clump were used for data recording, as they represent the maximum growth potential of a particular accession. Data on seven quantitative characters viz., plant height (cm), leaf width (cm), leaf length (cm), internode length (cm), internode thickness (cm), arrow length (cm) and peduncle length (cm) were recorded from each accession as per the standard procedure.

Pearson correlation co-efficient were worked out using OP STAT package developed by HAU, Hissar, Haryana. Diversity analysis presented in this study were performed using R statistical software, version 3.4.1 (R DEVELOPMENT CORE TEAM, 2017).

Results and Discussion

Saccharum spontaneum distribution in Haryana and its variability

The collection route covered most of the districts of Haryana starting from Karnal, Panipat, Sonipat, Rohtak, Jhajjar, Rewari, Palwal, Mewat, Mahendragargh, Bhivani, Hissar, Fatehabad, Sirsa, Kurukshetra, Yamuna nagar, Ambala and Panchkula (Fig. 1). In Haryana *S. spontaneum* was widely distributed from an altitude of 189 m MASL to 384 m MASL. Among the 50 *Saccharum spontaneum* collections (Table 1.), plant height ranged between 51.00 cm (IND 16-1771) to 362 cm (IND 16-1850), internode length with 2.50 cm (IND 16-1776) to 29.00 cm (IND 16-1757), internode thickness 0.2 cm (IND 16-1768, IND 16-1769, IND 16-1776, IND 16-1780, & IND 16-1781) to 1.10 cm (IND 16-1781 IND 16-1757), leaf length 48.00 cm (IND 16-1759) to 144.00 cm (IND 16-1796), leaf width 0.10 cm (IND 16-1752, IND 16-1760, IND 16-1765, IND 16-1775, IND 16-1776 & IND 16-1781) to 1.0 cm (IND 16-1790 & IND 16-1796), arrow length 22.00 cm (IND 16-1774) to 62.00 cm (IND 16-1796) and peduncle length with minimum18.00 cm (IND 16-1776) to 120 cm (IND 16-1850) and three *Erianthus* sp. were collected from 18 districts. Karnal had the maximum of 9 collections followed by Hissar (6 collections). Three clones each were collected from Yamuna nagar, Rohtak, Rewari, Panipat, Mahendragarh, Bhiwani and Ambala districts. Distribution of this species was less in Kaithal, Mewat, Panchkula and Sirsa districts where only single collections were made.

The first collection IND 16-1748 was from an abandoned filed in Karnal district. It was a medium tall plant with short arrow (32 cm) and flowering was completed. A short plant type IND 16-1759 was single isolated clump with early flowering and was collected from Sonipat district. In Rohtak district along the channel bund at Humayunpur locality a large population was located (IND 16-1761). Another clone IND 16-1764 was spotted in the paddy filed bund as large population with medium height, long internode (15 cm) and erect cane in Jhajjar district. At Mewat district, an early flowering type IND 16-1769 was gathered from a waste land. In a relatively dry tract at Mahendragarh district a small population was spotted (IND 16-1772). Under the Water logged condition, a short clone IND 16-1776 was collected at Bhiwani district. Under the saline condition at ICAR – Central Institute for Research on Buffalo, Hissar one tall *S. spontaneum* (IND 16-1778: 252 cm was collected. IND 16-1781 was a narrow leaved (0.1 cm) clone taken from roadside cotton field which occurred as small population. IND 16-1787 had multiple buds and occurred along the canal bunds in Hissar district. One of the tall types IND 16-1795 (312 cm) was spotted in a perennial water pond at Kansapur in Ambala district.

Table 1. Passport information on list of *Saccharum spontaneum* collections from Haryana state

S. No.	Collection No.	Plant height (cm)	Internode length (cm)	Internode thickness (cm)	Leaf length (cm)	Leaf width (cm)	Arrow length (cm)	Peduncle length (cm)
1	IND 16-1748	151.00	10.20	0.30	82.00	0.40	32.00	42.00
2	IND 16-1749	275.00	14.00	0.62	132.00	0.20	32.00	87.00
3	IND 16-1750	171.00	8.40	0.38	101.00	0.32	27.00	37.00
4	IND 16-1752	292.00	11.80	0.40	116.00	0.10	38.00	76.00
5	IND 16-1753	242.00	10.00	0.40	93.00	0.70	52.00	90.00
6	IND 16-1754	334.00	9.50	0.70	86.00	0.90	31.00	81.00
7	IND 16-1755	256.00	20.00	0.80	87.00	0.90	46.00	111.00
8	IND 16-1756	162.00	9.50	0.50	78.00	0.20	42.00	74.00
9	IND 16-1757	227.00	29.00	1.10	92.00	0.20	62.00	47.00
10	IND 16-1758	186.00	16.00	0.70	84.00	0.60	42.00	90.00
11	IND 16-1759	98.00	8.00	0.30	48.00	0.20	29.00	36.00
12	IND 16-1760	162.00	9.00	0.40	66.00	0.10	41.00	59.00
13	IND 16-1761	171.00	10.00	0.30	71.00	0.30	27.00	57.00
14	IND 16-1762	281.00	18.00	0.70	132.00	0.30	33.00	75.00
15	IND 16-1763	182.00	19.00	0.40	67.00	0.20	31.00	48.00
16	IND 16-1764	182.00	15.00	0.40	58.00	0.30	37.00	64.00
17	IND 16-1765	153.00	9.00	0.30	68.00	0.10	27.00	42.00
18	IND 16-1766	200.00	8.50	0.30	76.00	0.20	33.00	41.00
19	IND 16-1767	235.00	9.00	0.40	136.00	0.30	37.00	35.00
20	IND 16-1768	142.00	6.50	0.20	63.00	0.20	28.00	52.00
21	IND 16-1769	112.00	8.00	0.20	56.00	0.20	28.00	46.00
22	IND 16-1770	162.00	13.00	0.30	78.00	0.20	25.00	36.00
23	IND 16-1771	51.00	7.00	0.20	83.00	0.20	23.00	32.00
24	IND 16-1772	172.00	7.00	0.30	97.00	0.60	34.00	54.00
25	IND 16-1773	146.00	10.00	0.30	96.00	0.20	33.00	54.00
26	IND 16-1774	174.00	13.00	0.50	72.00	0.30	22.00	40.00
27	IND 16-1775	195.00	14.00	0.40	87.00	0.10	42.00	64.00
28	IND 16-1776	115.00	2.50	0.20	88.00	0.10	31.00	18.00
29	IND 16-1777	240.00	7.10	0.60	71.00	0.70	45.00	52.00
30	IND 16-1778	252.00	23.00	0.60	77.00	0.30	27.00	57.00
31	IND 16-1780	182.00	10.00	0.20	97.00	0.20	59.00	31.00
32	IND 16-1781	140.00	6.00	0.20	66.00	0.10	42.00	19.00
33	IND 16-1782	201.00	8.00	0.50	104.00	0.30	34.00	74.00
34	IND 16-1783	265.00	10.00	0.50	62.00	0.20	34.00	64.00

Table 1 Contd...

S. No.	Collection No.	Plant height (cm)	Internode length (cm)	Internode thickness (cm)	Leaf length (cm)	Leaf width (cm)	Arrow length (cm)	Peduncle length (cm)
35	IND 16-1784	284.00	11.00	0.70	97.00	0.50	37.00	57.00
36	IND 16-1785	262.00	14.00	0.50	79.00	0.70	34.00	60.00
37	IND 16-1786	352.00	12.50	0.70	112.00	0.80	56.00	110.00
38	IND 16-1787	286.00	10.00	0.90	122.00	0.40	31.00	82.00
39	IND 16-1788	175.00	14.00	0.40	94.00	0.40	28.00	30.00
40	IND 16-1789	264.00	11.00	0.50	110.00	0.40	42.00	72.00
41	IND 16-1790	282.00	14.00	0.70	89.00	1.00	42.00	80.00
42	IND 16-1791	172.00	8.50	0.20	93.00	0.30	26.00	40.00
43	IND 16-1792	304.00	10.00	0.60	132.00	0.60	28.00	75.00
44	IND 16-1793	336.00	19.00	0.60	79.00	0.50	42.00	95.00
45	IND 16-1794	195.00	7.00	0.40	85.00	0.30	58.00	94.00
46	IND 16-1795	312.00	22.00	0.70	66.00	0.50	36.00	62.00
47	IND 16-1796	340.00	9.00	0.90	144.00	1.00	62.00	93.00
48	IND 16-1797	252.00	11.00	0.40	120.00	0.50	52.00	45.00
49	IND 16-1798	240.00	7.50	0.40	94.00	0.30	60.00	75.00
50	IND 16-1850	362.00	16.00	0.60	92.00	0.70	34.00	120.00
	Average	218.54	11.71	0.48	89.56	0.39	37.48	61.50
	Maximum	362.00	29.00	1.10	144.00	1.00	62.00	120.00
	Minimum	51.00	2.50	0.20	48.00	0.10	22.00	18.00
	SD	71.88	4.97	0.21	22.46	0.25	10.69	24.11

Among the collections from Haryana state, plant height ranged between 362 cm and 51 cm with the standard deviation of 71.16 cm. Variation for leaf length (144 cm to 48 cm), leaf width (1 cm to 0.1 cm), peduncle length (120 cm to 18 cm), arrow length (62 cm to 22 cm), stalk diameter (1.1 cm to 0.2 cm) and internode length (29 cm to 2.5 cm) were observed. *S. spontaneum* was distributed throughout the state and showed high variability for different agronomical traits.

Saccharum spontaneum distribution in Chandigarh and its variability

On the way from Haryana to Punjab two *S. spontaneum* were collected from the union territory of Chandigarh . Both the clones IND 16-1799 and IND 16-1800 were medium in height, narrowed leaf (0.2 cm) with average leaf length of 112 cm and were found in abandoned lands.

Saccharum spontaneum distribution in Punjab and its variability

In Punjab the river banks of Ravi, Beas and Sutlej were extensively covered and 48 *S. spontaneum* and two *Erianthus* sp. distributed in 19 districts (Fig. 3) were collected (Table 2). Maximum number of collections were from Pathankot district (7) which included hilly regions followed by Sangur (5), Ludhiana (4). Three germplasm each were collected form Faridkot, Gurdaspur, Hoshiapur, and Patiala districts. *S. spontaneum* was distributed throughout the Punjab states from

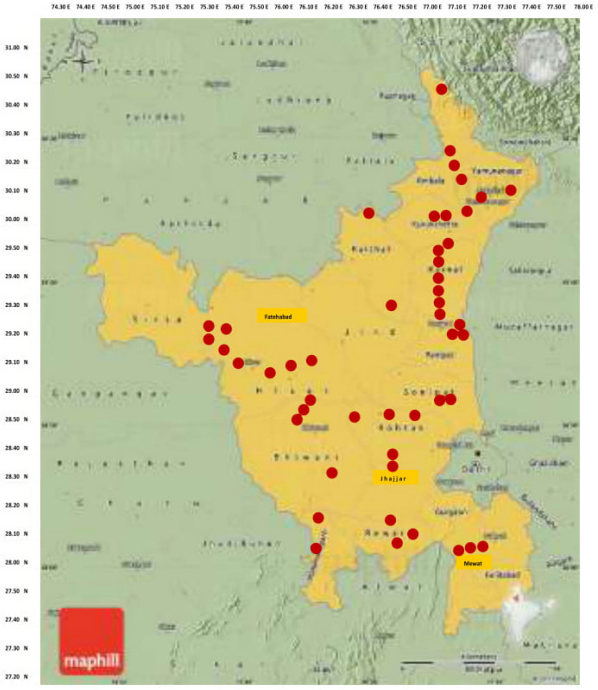

Figure 1. Map showing *Saccharum spontaneum* collection sites (red dots) in Haryana

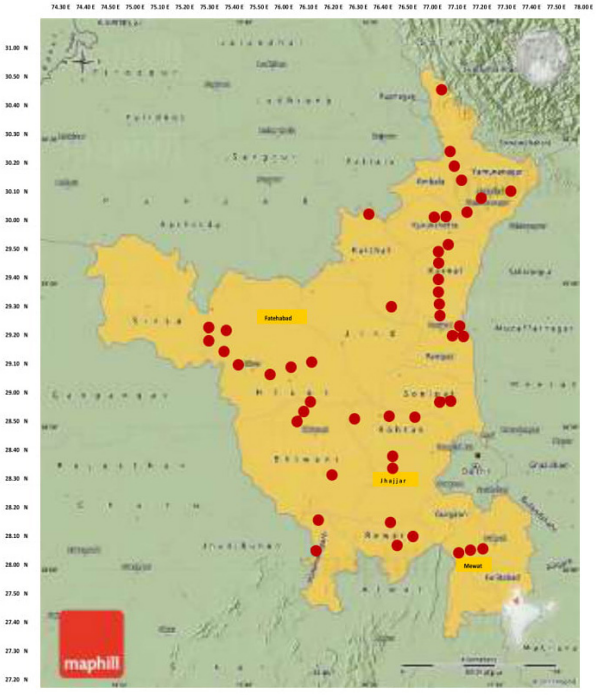

Figure 2. Map showing *Saccharum spontaneum* collection sites (pink and blue dots) in Chandigarh and Punjab

197 m MSL (IND 16-1828) to 682 m MSL (IND 16-1815) and mostly occur as small population and individual clumps (Fig. 2).

The first collection in Punjab was from Rupnagar district in a waste land and it was tall (242 cm), heavy tillering and profusely flowering. IND 16-1802 was a very tall (352 cm) clone with long and broader leaves and moderate tillering. Another collection IND 16-1804 was collected from a paddy field bund and measured 254 cm height. One collection was made from Nawansahar (IND 16-1805) which had more number of internodes (21) and medium tall in stature (216 cm). In Gurdaspur district, a broad leaved clone (IND 16-1817) was spotted which was tall (332 cm), heavy tillering and profusely flowering. An early flowering type IND 16-1836 was located in Moga district where the flowering was completed and almost all the fluff had flown off. IND 16-1841 was a short form collected from Patiala branch canal bund. In addition one *E. bengalense* (IND 16-1820) was also collected from Amritsar.

High variability was observed among the collection form Punjab state for various quantitative characters. While plant height varied between 46 cm (IND 16-1813) and 450 cm (IND 16-1809), arrow length ranged from 14 cm IND 16-1839) to 66 cm (IND 16-1809). Leaf width varied greatly from 0.1 cm (in 9 clones - IND 16-1807, IND 16-1808, IND 16-1812, IND 16-

Figure 3. *S. spontaneum* (IND 16-1759), early flowering collection

1813, IND 16-1815 , IND 16-1823, IND 16-1828, IND 16-1829 and IND 16-1830) to 1.3 cm (IND 16-1831) with standard deviation of 0.29 cm. The shortest internode length was observed with IND 16-1813 (4 cm) and the longest internode length was recorded by IND 16-1827 (24 cm).

Erianthus sp. distribution in Haryana and Punjab

Totally, four *Erianthus* sp. were collected from these two states (Table 3). The first collection (IND 16-1751) of *Erianthus* sp**.** a medium tall type (238 cm) was collected from Karnal. This collection was having long leaves (196.00 cm) and broader leaf lamina (2.70 cm). Peduncle length was also maximum in this collection. Under the saline condition at ICAR – Central Institute for Research on Buffalo, Hissar one *Erianthus* clones (IND 16-1779: 270 cm) was collected. Internode length was maximum (25.00 cm) in this entry, however arrow length and peduncle length was found to be minimum (45.00 cm & 52.00 cm respectively). IND 16-1851, the third collection was found in Kaithal at an altitude of 261 MASL with reduced internode thickness (1.30 cm) leaf length (120 cm) and leaf width (1.40 cm). This was the only collection in which flowering was not noticed. Only one short (245.00 cm) collection of *Erianthus* sp. (IND 16-1820) was from Amritsar, Punjab. It had thick internode (1.30 cm), narrow leaf (1.40 cm) and long peduncle (111.00 cm).

Correlation analysis of Saccharum spontaneum collections from Haryana

Altitudinal gradients can negatively affect plant distribution and limit plant growth and reproduction. In natural environments, plants must overcome many abiotic stresses, such as drought, salt, cold, ultraviolet radiation (UV) and altitude, to complete their life cycles (Ahmad et al. 2018). Altitudinal gradients cause the climate and environment to differ greatly within a short

Figure 4. *Saccharum spontaneum* in canal bunds

vertical distance by decreasing air temperature, total atmospheric pressure and partial pressure of all atmospheric gasses and by increasing radiation in the forms of incoming solar radiation, outgoing nighttime thermal radiation, and UV radiation (Barry 1981; Korner 2007). In the present study, *S. spontaneum* collections of Haryana & Punjab indicated that altitude had no significant influence on any of the plant traits in Haryana collections (Table. 4) but showed non-significant negative trend in leaf length, leaf width, internode length, plant height and internode thickness. The non-significant association may be due to the minimal variation in climatic factors among the collection sites whose altitude ranged from 189 m MSL to 384 m MSL in Haryana and from 197 m MSL to 682 m MSL in Punjab. However, Liu *et al*., (2012) reported that altitude was negatively related to yield and sugar related traits in Chinese decaploids *S. spontaneum*. Correlation analysis among plant characters showed the highest correlation coefficient between plant height and internode thickness (0.722). Besides, plant height had significant positive association with all other characters studied *viz*., peduncle length (0.716), leaf width (0.635), leaf length (0.505), internode length (0.403) and arrow length (0.355). Internode length had highly significant positive correlation with internode thickness (0.614) and peduncle length (0.295). Internode thickness had positive correlation with peduncle length (0.606),

leaf width (0.560), leaf length (0.383) and arrow length (0.364). Leaf length recorded significant correlation with peduncle length (0.312), leaf width (0.284) and arrow length (0.268). Leaf width had a positive significant correlation with peduncle length (0.593) and arrow length (0.301). Arrow length recorded highly significant positive correlation with peduncle length (0.386) (Table 4).

Correlation analysis of Saccharum spontaneum collections from Chandigarh and Punjab

Similar trend as that of Haryana *S. spontaneum* collections was observed in Chandigarh and Punjab collections also. Plant height had a significant positive association with internode thickness (0.880) and all other characters studied *viz*., peduncle length (0.823), arrow length (0.774), leaf width (0.736), leaf length (0.463), and internode length (0.424). Internode length had positive correlation with leaf length (0.405), peduncle length (0.339), arrow length (0.319) and leaf width (0.315). Internode thickness had highly significant positive correlation with arrow length (0.788), peduncle length (0.753), leaf width (0.656) and leaf length (0.413). Leaf length recorded significant correlation with peduncle length (0.454) and arrow length (0.452). Leaf width recorded significant correlation with arrow length (0.555) and peduncle length (0.515). Arrow

Table 2. Passport information on list of *Saccharum spontaneum* collections from Chandigarh and Punjab state

S. No.	Collection No.	Plant height (cm)	Internode length (cm)	Internode thickness (cm)	Leaf length (cm)	Leaf width (cm)	Arrow length (cm)	Peduncle length (cm)
1	IND 16-1799	162.00	9.00	0.30	102.00	0.20	28.00	65.00
2	IND 16-1800	303.00	12.00	0.70	122.00	0.20	41.00	98.00
3	IND 16-1801	242.00	12.00	0.50	152.00	0.20	62.00	72.00
4	IND 16-1802	352.00	12.00	0.90	135.00	0.40	58.00	88.00
5	IND 16-1803	304.00	13.00	0.80	131.00	0.50	51.00	91.00
6	IND 16-1804	254.00	14.00	0.60	132.00	0.70	63.00	108.00
7	IND 16-1805	216.00	21.00	0.30	92.00	0.50	42.00	71.00
8	IND 16-1806	125.00	12.00	0.30	121.00	0.20	23.00	28.00
9	IND 16-1807	101.00	5.00	0.20	91.00	0.10	22.00	43.00
10	IND 16-1808	110.00	6.00	0.30	125.00	0.10	31.00	27.00
11	IND 16-1809	450.00	13.00	1.00	133.00	0.80	66.00	123.00
12	IND 16-1810	215.00	9.00	0.30	86.00	0.70	46.00	72.00
13	IND 16-1811	122.00	8.50	0.40	76.00	0.20	38.00	35.00
14	IND 16-1812	136.00	8.00	0.30	45.00	0.10	46.00	41.00
15	IND 16-1813	46.00	4.00	0.20	40.00	0.10	24.00	26.00
16	IND 16-1814	232.00	8.00	0.50	115.00	0.20	56.00	94.00
17	IND 16-1815	160.00	8.00	0.20	94.00	0.10	26.00	45.00
18	IND 16-1816	182.00	16.00	0.40	96.00	0.60	35.00	60.00
19	IND 16-1817	332.00	12.00	0.80	108.00	1.30	43.00	39.00

Table 2 contd...,

20	IND 16-1818	282.00	10.00	0.70	86.00	0.80	45.00	76.00
21	IND 16-1819	172.00	7.00	0.40	103.00	0.60	36.00	45.00
22	IND 16-1821	242.00	6.00	0.60	81.00	0.40	36.00	62.00
23	IND 16-1822	250.00	18.00	0.60	163.00	0.60	47.00	92.00
24	IND 16-1823	135.00	6.00	0.20	98.00	0.10	26.00	42.00
25	IND 16-1824	275.00	8.00	0.90	143.00	0.30	62.00	80.00
26	IND 16-1825	225.00	4.50	0.50	78.00	0.40	26.00	73.00
27	IND 16-1826	151.00	8.40	0.40	58.00	0.20	22.00	49.00
28	IND 16-1827	265.00	24.00	0.40	166.00	0.20	37.00	60.00
29	IND 16-1828	105.00	9.00	0.20	88.00	0.10	29.00	26.00
30	IND 16-1829	75.00	4.00	0.30	94.00	0.10	28.00	35.00
31	IND 16-1830	118.00	6.00	0.20	112.00	0.10	25.00	42.00
32	IND 16-1831	412.00	11.00	0.80	132.00	1.30	58.00	94.00
33	IND 16-1832	327.00	9.50	0.70	132.00	0.80	64.00	85.00
34	IND 16-1833	150.00	13.50	0.40	96.00	0.40	28.00	37.00
35	IND 16-1834	117.00	7.00	0.30	75.00	0.20	28.00	21.00
36	IND 16-1835	182.00	8.50	0.30	101.00	0.20	19.00	38.00
37	IND 16-1836	176.00	9.50	0.30	102.00	0.20	21.00	49.00
38	IND 16-1837	161.00	8.00	0.20	90.00	0.20	28.00	47.00
39	IND 16-1838	215.00	14.00	0.60	102.00	0.60	45.00	70.00
40	IND 16-1839	96.00	5.00	0.30	93.00	0.20	14.00	47.00
41	IND 16-1840	156.00	9.00	0.30	75.00	0.20	26.00	36.00
42	IND 16-1841	141.00	10.00	0.20	114.00	0.20	27.00	45.00
43	IND 16-1842	152.00	7.00	0.20	80.00	0.50	22.00	40.00
44	IND 16-1843	263.00	15.00	0.50	81.00	0.60	36.00	70.00
45	IND 16-1844	207.00	9.50	0.20	90.00	0.30	32.00	58.00
46	IND 16-1845	262.00	11.00	0.60	68.00	0.60	36.00	88.00
47	IND 16-1846	138.00	7.00	0.30	134.00	0.20	28.00	68.00
48	IND 16-1847	174.00	7.00	0.30	96.00	0.20	27.00	50.00
49	IND 16-1848	181.00	23.00	0.30	93.00	0.30	29.00	45.00
50	IND 16-1849	161.00	12.00	0.30	162.00	0.20	23.00	45.00
	Average	200.20	10.20	0.43	103.64	0.37	36.22	58.82
	Maximum	450.00	24.00	1.00	166.00	1.30	66.00	123.00
	Minimum	46.00	4.00	0.20	40.00	0.10	14.00	21.00
	SD	85.43	4.50	0.22	28.47	0.29	13.72	24.07

Table 3. Passport information on list of *Erianthus* sp. collections from Haryana; Punjab state

S. No.	Collection No.	Location District	Alti tude (m)	Plant height (cm)	Inter node length (cm)	Internode thickness (cm)	Leaf length (cm)	Leaf width (cm)	Arrow length (cm)	Peduncle length (cm)
1	IND 16-1751	Karnal	238	322	4.8	1.20	196.0	2.70	75.00	111.00
2	IND 16-1779	Hisar	208	270	25.0	0.90	162.0	1.50	45.00	52.00
3	IND 16-1851	Kaithal	261	298	21.0	0.90	120.0	1.40	NF	NF
4	IND 16-1820	Amritsar	245	245	11.0	1.30	123.0	1.40	121.00	105.00
		Average	238	283.8	15.5	1.08	150.3	1.75	80.33	89.33
		Maximum	261	322.0	25.0	1.30	196.0	2.70	121.00	111.00
		Minimum	208	245.0	4.8	0.90	120.0	1.40	45.00	52.00
		SD	22.2	33.45	9.22	0.21	36.0	0.63	38.28	32.47

* NF – Not Flowered

length has a highly significant positive correlation with peduncle length (0.766) (Table 5).

Diversity analysis

Crop wild relatives or species have rich and variable genetic integrity explorable for breeding programmes. The first and foremost important step of any successful breeding programme is to identify crop plants that exhibit exploitable variation for the trait(s) of interest. Preservation of genetic diversity is possible when genetic or germplasm resource is realized as the most precious asset in conserving genetic diversity (Brown 1989). Some grouping methods in genetic diversity studies identified origin and geographical diversity not important in measuring genetic diversity. Diversity in sugarcane is not to be associated with origin (Nair et al. 1999). Earlier studies by (Stevenson 1965), (Skinner et al. (1987), and Govindraj et al. (2014) utilized morphological and agronomical traits for assessment of variability. Genetically diverse germplasm of *S. spontaneum* can effectively contribute to disease resistance, cold tolerance, insect resistance, and tillering potential to new breeding products. Clustering of Haryana *Saccharum spontaneum* collections (Fig.

Table 4. Correlation among different quantitative traits of Saccharum spontaneum collections from Haryana state

Parameter	Plant height	Internode length	Internode thickness	Leaf length	Leaf width	Arrow length	Peduncle length
Altitude	0.064^{NS}	-0.012^{NS}	0.003^{NS}	0.030^{NS}	0.122^{NS}	0.101^{NS}	0.065^{NS}
Plant height		0.403^{**}	0.722^{**}	0.505^{**}	0.635^{**}	0.355^{*}	0.716^{**}
Internode length			0.614^{**}	-0.006^{NS}	0.161^{NS}	0.129^{NS}	0.295^{*}
Internode thickness				0.383^{**}	0.560^{**}	0.364^{**}	0.606^{**}
Leaf length					0.284^{*}	0.268^{NS}	0.312^{*}
Leaf width						0.301^{*}	0.593^{**}
Arrow length							0.386^{**}

* $P<0.05$; ** $P<0.01$

Table 5. Correlation among different quantitative traits of *Saccharum spontaneum* collections from Chandigarh and Punjab state

Parameter	Plant height	Internode length	Internode thickness	Leaf length	Leaf width	Arrow length	Peduncle length
Altitude	-0.168^{NS}	-0.181^{NS}	-0.141^{NS}	-0.273^{NS}	-0.235^{NS}	0.075^{NS}	-0.046^{NS}
Plant height		0.424**	0.880**	0.463**	0.736**	0.774**	0.823**
Internode length			0.262^{NS}	0.405**	0.315*	0.319*	0.339*
Internode thickness				0.413**	0.656**	0.788**	0.753**
Leaf length					0.176^{NS}	0.452**	0.454**
Leaf width						0.555**	0.515**
Arrow length							0.766**

* P<0.05; ** P<0.01

5) as well as Chandigarh and Punjab collections (Fig. 6) through complete linkage clustering using Euclidean distances (GGE Biplot package of R statistics) resulted in five clear clusters among the collections respectively (Table 6 and Table 7).

The I cluster from Haryana was with single collection (IND 16-1757) having a long internode length (29.00 cm) and internode thickness (1.10 cm). It was collected from the Yamuna river bank of Panipat located at 29.23°N longitude 77.08°E latitude and altitude of 223 MASL. The II cluster consists of two collections viz., IND 16-1786 from Hissar, 218 MASL and IND 16-1796, 227 MASL. IND 16-1786 is the second tallest collection and

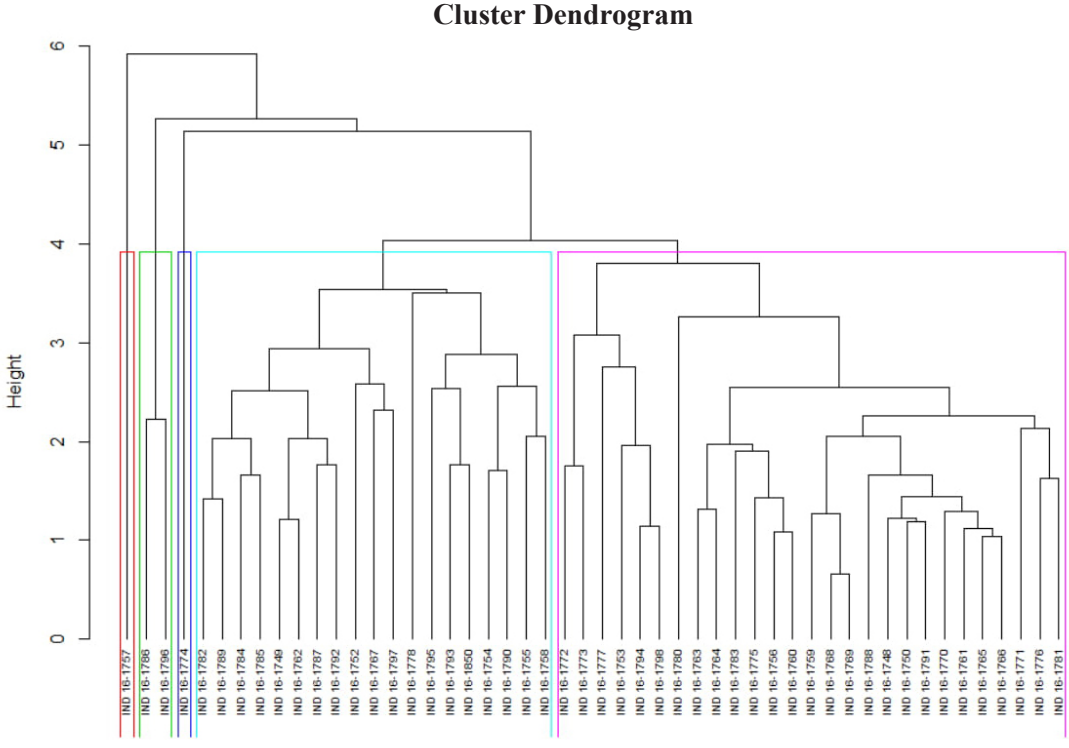

Figure 5. Dendrogram of *Saccharum spontaneum* collections from Haryana based on Euclidean distances

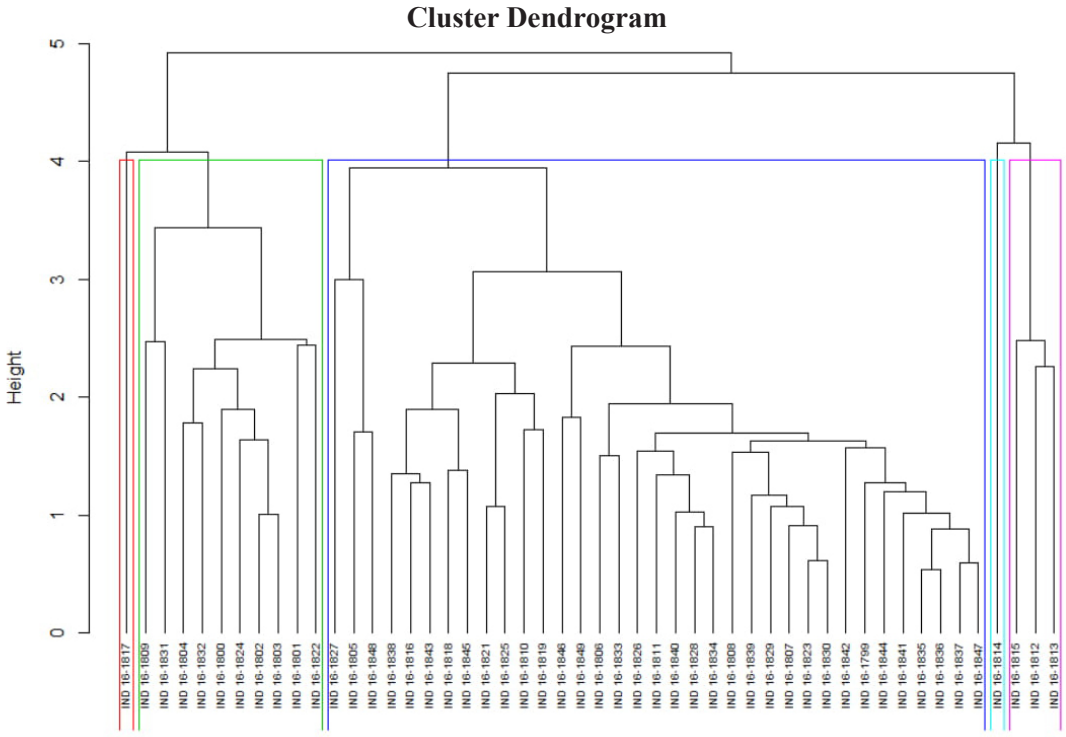

Figure 6. Dendrogram of *Saccharum spontaneum* collections from Chandigarh & Punjab based on Euclidean distances

has the longest arrow among the collections from Haryana. Only one collection (IND 16-1774) from Mahendragarh forms the cluster III from the highest altitude (384 MASL) in Haryana, with a shortest arrow among the collections. In cluster IV nineteen clones from Karnal (5 collections), Hissar (3 collections), Fatehabad, Yamuna nagar and Ambala, two collections each and single collection from Rohtak, Palwal, Kaithal, Kurukshetra and Panipat. The average attitude in the cluster was 232.53 MASL and the average height was recorded to be 276.63 cm. This cluster recorded average internode length of 14.07 cm, internode thickness (0.61 cm), leaf length (102.11 cm), leaf width (0.51 cm), arrow length (36.74 cm) and peduncle length (75.47 cm). Cluster V forms the largest collection of *Saccharum spontaneum* from Haryana with 27 collections among which four are from Karnal, three each are from Bhiwani and Rewari, two each from Mahendragarh, Hissar, Rohtak, Jhajjar and Sonipat and on each from Yamuna nagar, Panchkula, Sirsa, Panipat, Pulwal, Mewat and Kurukshetra. The average altitude was 235.30 MASL and these were having average short internode length (9.43 cm), internode thickness (0.33 cm) and reduced lead width (0.27 cm) known to contain drought adaptive mechanism.

In Chandigarh and Punjab *Saccharum spontaneum* collections, IND 16-1817 was with single collection in the I cluster at an average altitude of 246 MASL. This was the tallest (332 cm height) among all with broad leaf (1.30 cm) and heavy tillering. The II cluster with average altitude of 254.50 MASL consists of ten collections viz., three from Ludhiana, and two from Mansa and one each from Pathankot, Firozpur, Rupnagar and Tarantaran and one among the two of Chandigarh. This cluster recorded the average longest internode length (12.25 cm, average medium thick internode (0.75 cm), average longest leaf length (137.50)

Table. 6. Grouping of new *Saccharum spontaneum* collections of Haryana based on Euclidean distances

Cluster	Collection No.	Average values of the group							
		Altitude (m)	Plant height (cm)	Internode length (cm)	Internode thickness (cm)	Leaf length (cm)	Leaf width (cm)	Arrow length (cm)	Peduncle length (cm)
I	IND 16-1757	223.00	227.00	29.00	1.10	92.00	0.20	62.00	47.00
II	IND 16-1786 IND 16-1796	222.50	346.00	10.75	0.80	128.00	0.90	59.00	101.50
III	IND 16-1774	384.00	174.00	13.00	0.50	72.00	0.30	22.00	40.00
IV	IND 16-1782,IND 16-1789, IND 16-1784, IND 16-1785, IND 16-1749, IND 16-1762, IND 16-1787, IND 16-1792, IND 16-1752, IND 16-1767, IND 16-1797, IND 16-1778, IND 16-1795, IND 16-1793, IND 16-1850, IND 16-1754, IND 16-1790, IND 16-1755, IND 16-1758	232.53	276.63	14.07	0.61	102.11	0.51	36.74	75.47
V	IND 16-1772, IND 16-1773, IND 16-1777, IND 16-1753, IND 16-1794, IND 16-1798, IND 16-1780, IND 16-1763, IND 16-1764, IND 16-1783, IND 16-1775, IND 16-1756, IND 16-1760, IND 16-1759, IND 16-1768, IND 16-1769, IND 16-1788, IND 16-1748, IND 16-1750, IND 16-1791, IND 16-1770, IND 16-1761, IND 16-1765, IND 16-1766, IND 16-1771, IND 16-1776, IND 16-1781	235.30	169.56	9.43	0.33	78.44	0.27	36.07	50.04

Table. 7. Grouping of new Saccharum spontaneum collections of Chandigarh and Punjab based on Euclidean distances

Cluster	Collection No.	Altitude (m)	Plant height (cm)	Inter-node length (cm)	Inter-node thick-ness (cm)	Leaf length (cm)	Leaf width (cm)	Arrow length (cm)	Pe-duncle length (cm)
I	IND 16-1817	246.00	332.00	12.00	0.80	108.00	1.30	43.00	39.00
II	IND 16-1809, IND 16-1831, IND 16-1804, IND 16-1832, IND 16-1800, IND 16-1824, IND 16-1802, IND 16-1803, IND 16-1801, IND 16-1822	254.50	316.90	12.25	0.75	137.50	0.58	57.20	93.10
III	IND 16-1827, IND 16-1805, IND 16-1848, IND 16-1838, IND 16-1816, IND 16-1843, IND 16-1818, IND 16-1845, IND 16-1821, IND 16-1825, IND 16-1810, IND 16-1819, IND 16-1846, IND 16-1849, IND 16-1806, IND 16-1833, IND 16-1826, IND 16-1811, IND 16-1840, IND 16-1828, IND 16-1834, IND 16-1808, IND 16-1839, IND 16-1829, IND 16-1807, IND 16-1823, IND 16-1830, IND 16-1842, IND 16-1799, IND 16-1844, IND 16-1841, IND 16-1835, IND 16-1836, IND 16-1837, IND 16-1847	238.17	169.57	9.93	0.34	97.29	0.31	29.83	50.43
IV	IND 16-1814	638.00	232.00	8.00	0.50	115.00	0.20	56.00	94.00
V	IND 16-1815, IND 16-1812, IND 16-1813	664.67	114.00	6.67	0.23	59.67	0.10	32.00	37.33

and average longest arrow length (57.20 cm). It was observed to have second longest average peduncle length (93.10 cm). Among the fifty collections, thirty five collections come under the III cluster with the recorded average altitude of 238.17 MASL. This cluster had an average plant height of 169.57 cm, average internode length of 9.93 cm, average internode thickness of 0.34, average leaf length of 97.29 cm, average leaf width of0.31 cm, average arrow length of 29.83 cm and average peduncle length of 50.43 cm. Among the thirty five collections from this group, five collections were from Sangrur, three each from Faridkot, Patiala and Hoshiarpur, two each from Gurdaspur, Fatehgarh Sahib, Amritsar, Pathankot, Barnala, Mukstar & Moga and single collections from Nawanshahr, Ludhiana, Firozpur, Bathinda, Tarantaran, SAS Nagar and one was from Chandigarh. The IV cluster consists of one clone IND 16-1814 from Pathankot collected at an average attitude of 638.00 MASL and has the average longest peduncle length (94.00 cm). This collection recorded the second longest leaf length (115.00 cm) and narrow average leaf width of 0.20 cm. The V cluster consists of three collections from Pathankot and maximum average altitude recorded was 664.67 MASL. This cluster was observed to have shortest average plant height (114.00 cm), shortest average internode length (6.67 cm), highly reduced average internode thickness, shortest average leaf length (59.67 cm) completely reduced leaf lamina width (0.10 cm) and shortest average peduncle length (37.33 cm), indicated that they may possess drought tolerant characteristics.

Davis et al. (2002), Govindaraj et al. (2016) and Lopez et al. (2008) are of the opinion of the opinion that these characteristics were known to be drought adaptation mechanism in Sugarcane. Tena et al. (2016) also reported the same by grouping of genotypes based on qualitative characters among collection of local and country of introduction of exotic genotypes of sugarcane germplasm studied and considerable phenotypic diversity was observed. The variation within locations was larger than the diversity observed between locations. However Chang et al. (2012) has stated that, the native *Saccharum spontaneum* collected in Sichuan, China was genetically diverse and the clusters were closely related to their geographic origins and surrounding geographic environments. Zhang et al. (2010) was also of the opinion that the genetic diversity of eighty nine *Saccharum spontaneum* accessions collected from five different ecological regions of Guangxi, China were found be genetically diverse. The populations collected along the river Ghaggar basins and its tributaries in the western part of Haryana, river Yamuna and its tributaries in the eastern part of Haryana, river Ravi, Sutlej and Beas of Punjab and canal banks forms appears that geographic distance and environmental factors including local climates interplay in the formation of the genetic structure and geographic patterns of the native *S. spontaneum* populations. Grouping of the collections into five distinct clusters clearly indicated that fuller sampling of the genetic diversity of temperature-tolerant local germplasm in the species requires comprehensive germplasm collection in heterogeneous environments along the northern regions of the species distribution. The germplasm should be a valuable source in *Saccharum* improvement program in near future.

Conclusion

According to the conservation of biodiversity (1992), Conservation of biodiversity and sustainable use of its components are sovereign rights of each nation. Natural calamities, intensive agriculture, habitat destruction and climate change are pausing great threat to the biological diversity. Conservation of the biological diversity and preservation for them for posterity is our

duty. Haryana and Punjab were not fully explored earlier. In the present attempt to tap the *Saccharum* sp. diversity, a total of 50 and 50 *S. spontaneum* were collected from Haryana and Punjab states respectively along with four *Erianthus* sp. They were occurring mostly as small population except in the canal bunds where larger population was noticed (Fig. 4). In the exploration route, two *S. spontaneum* were collected in the union territory of Chandigarh. *S. spontaneum* was well distributed throughout both the states and collections could be made from all the districts and rivers surveyed. Variation for leaf and stem characters were observed from the collections. Plant height varied from 450 cm to 46 cm and stalk diameter varied from 0.2 cm to 1.1 cm. Leaf width ranged between 0.1 cm and 1.3 cm. Collections from Haryana *viz.,* IND 16-1850 recorded highest for plant height and peduncle length, IND 16-1757 for internode length and internode thickness and IND 16-1796 for leaf length, leaf width and arrow length. Similarly among Punjab collections, IND 16-1809 is good for plant height, internode thickness, arrow length and peduncle length, IND 16-1827 for internode length and leaf length and IND 16-1831 for leaf width. *Saccharum spontaneum* accessions collected from the river Ghaggar river Yamuna in Haryana, river Ravi, Sutlej and Beas of Punjab and canal banks forms into five distinct clusters clearly indicated that fuller sampling of the genetic diversity of temperature-tolerant local germplasm in the species. Hence the collection would add more diversity to the germplasm maintained in our institute. In addition to the 100 *S. spontaneum,* four *Erianthus* sp. were collected during the exploration which will enrich our gene pool. These are being conserved at the field gene bank at ICAR Sugarcane Breeding Institute, Coimbatore. These collections may harbor novel genes for biotic stress resistance and climate resilience. Characterization of them and utilization of them in the future pre breeding programmes would help to broaden the genetic base of sugarcane cultivars.

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