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DISTRIBUTION AND DIVERSITY IN *CUCUMIS SATIVUS* L. VAR. *HARDWICKII* GERMPLASM IN WESTERN GHATS OF MAHARASHTRA - A DIVA-GIS APPROACH

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KEYWORDS

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ABSTRACT

Cucumis sativus var. *hardwickii* is a wild relative of cucumber and occurs in foot hills of the Himalayas, North Western states, Western and Eastern Ghats, and adjoining states of India. Germplasm were collected from diverse agro-ecological niches covering Raigadh and Ratnagiri districts. The collected germplasm exhibited variation in qualitative characters of fruits such as shape, luster, blossom end, ridges etc. Wide range of variability were recorded in fruit and seed characters, viz., fruit length (44.88 - 97.54 mm), fruit width (36.31-52.89 mm.), perimeter of the fruits (12.78-17.42 mm), green fruits weight (33.60-152.79 g.), flesh/ rind thickness (0.26–1.24 mm), number of seeds / fruit (199.8- 302.8), seed length (5.16- 8.9 mm), seed width (2.02-2.62mm), number of stripes/placenta (8.4-10.0) present, 100 seed weight (0.53- 1.19 g.), TSS (3.1-13.7 °B), titrable acidity (0.57-0.76 %) and moisture (94.4-97.0 %). Two major clusters were observed in Ward's minimum variance dendrogram generated for quantitative traits. Out of five accessions IC 614594 (ND/DC-16) having high fruit length (97.54mm), fruit weight (152.79g) and number of seeds per fruit (302.8) was identified as promising. DIVA-GIS grid maps generated for specific traits confirms the presence of diversity in *Cucumis sativus* var. *hardwickii* germplasm.

INTRODUCTION

The genus *Cucumis* belongs to the family Cucurbitaceae (Whitaker, Davis 1962), order Cucurbitales. The cucumber is very popular and widely cultivated vegetable in India. It is widely grown for its edible fruits in nearly all the tropical, semitropical and temperate regions of the world. *Cucumis sativus* L. var. *hardwickii* ($2n = 2X = 14$) is a rare, endemic, wild progenitor of cultivated cucumber (*Cucumis sativus* L.). It is an annual, vigorous climbing species, monoecious and highly cross-pollinated having wide genetic variability across the globe. The plant is a natural component of grasslands especially in farm borders trailing to fences, bushes, and other supports. Wild species of *Cucumis* have been found to possess useful morpho-agronomic characters as well as resistance against pest and diseases, therefore, emphasis has been laid on the utilization of the variability in the wild species of *Cucumis* for improvement of cucumber and musk melon (Deakin et al., 1971). *C. sativus* L. var. *hardwickii* possess several desirable characters viz. sequential multiple fruiting habit, multiple lateral branching and total fruit weight per plant (Staub et al., 1993) and potential source for increased yield in pickling cucumbers (Horst and Lower 1978).

The Indian sub-continent is considered to be the centre of origin for *C. sativus* and a centre of diversity for *C. melo* (Zeven and de Wet, 1982). Six species of *Cucumis* with *C. melo* L. and *C. sativus* L. being cultivated and *C. prophetarum* L., *C. callosus* (Rottler) Cogn. et Harms, *C. hystrix* Chakrav and *C. setosus* Cogn occur as truly wild in India. Both melon and cucumber are represented by wild and weedy forms in their distribution range, the former represented by *C. melo* subsp. *Agristis* (Naud.). Pangalo and the latter by *C. sativus* var. *hardwickii* (Royle) Gabaev. (Jeffrey, 2001). The genus *Cucumis* consists of 52 species rather than 33 species reported earlier (Ghebretinsae et al., 2007; Kocyan et al., 2007; Schaefer, 2007; Renner et al., 2007).

In India, *Cucumis sativus* L. var. *hardwickii* (Royle) Alef. has been reported in the higher elevations in the foot hills of the Himalayas of India (Deakin et al., 1971), (Chakravarthy, 1982; Arora and Nayar, 1984), Mount Abu, Rajasthan (Bisht et al., 2004) and distributed in different states of India i.e. Himachal Pradesh, Uttar Pradesh, Uttarakhand, Rajasthan, Madhya Pradesh, Maharashtra, Chhatisgarh and Odisha (Dikshit, 2014a). Diversity is reported to occur in different species of *Cucumis* namely, *C. callosus* (Rottler) Cogn, *C. sativus* var. *hardwickii* (Royle) Alef., *C. hystrix* Chakra, *C. trigonus* Roxb. and *C. prophetarum* L. in India (Arora and Nayar, 1984). Based on the distribution of genetic variability and genetic erosion status the need for collection of *Cucumis* species from India has been emphasized by various workers (Umesh Chandra, 1995; Umesh Chandra and Koppar, 1992). The objective of the present study is to collect diverse germplasm through explorations and study the *in-situ* diversity exists within wild types of *Cucumis sativus* L. var. *hardwickii* and to identify the most useful genotypes with supporting studies on diversity analysis using DIVA-GIS.

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MATERIALS AND METHODS

An exploration for the collection of wild crops germplasm was organized in Western Ghats region of Maharashtra during November 2014. *Cucumis sativus* L. var. *hardwickii* germplasm were collected during survey from two districts of Maharashtra (Raigadh and Ratnagiri). The collection sites included cultivated fields and natural wild habitats (Table 1). The fruits were collected in random or bulk sampling methods depending upon the site of collection and availability of the material. Detailed passport data on place of collections, longitude, and frequency of occurrence, sample type, sampling method, associated vegetation and habitat/ecology were recorded at each site along with important descriptor traits. Global positioning System (GPS Garmin 12) was used to record the geographical coordinates. Each of the populations collected was assigned a specific collector number and later indigenous collection (IC) number was assigned to each of the targeted wild species *Cucumis sativus* var. *hardwickii* collection. The fruits of *Cucumis sativus* var. *hardwickii* were subjected for qualitative and quantitative analysis. Selective qualitative and quantitative characters of fruits and seeds were recorded using standard descriptors viz., Stripes per fruit (SPF), Fruit length (FLT), Fruit width (FWD), Fruit circumference (FRC), Green fruit weight (FRW), Flesh rind thickness (FRT), Number of seeds per fruit (NSF), Seed length (SLT), Seed width (SWD) and 100 seed weight (100SW). DIVA-GIS version 7.5 (www.diva-gis.org) was used for mapping, modelling and diversity analysis of select traits. SAS Enterprise Guide 4.3 was used for statistical analysis and multivariate analysis. Standard methods for physico-chemical analysis viz. Total soluble solids (TSS°Brix), Titrable acidity (%) and Moisture (%) were adopted (Anonymous, 1975).

Total soluble solids

Pulp was extracted from fruits and total soluble solids (TSS) were determined using Atago make RX 1000 digital refractometer. A drop of juice was extracted and placed on clean prism of refractometer and the lid was closed. Reading was taken directly from the scale at 20°C temperature and recorded as total soluble solids in °Brix.

Titration acidity

The total acidity of *Cucumis hardwickii* fruits was estimated in terms of citric acid. It was determined by titrating the sample against 0.1N Sodium hydroxide (NaOH) solution from colorless to faint pink. A few drops of one percent Phenolphthalein were used as an indicator and the percentage of total acidity were calculated by using following formula.

$$\text{Titration acidity (\%)} = \frac{\text{Titrate} \times \text{normality of alkali} \times \text{equivalent weight of acid}}{\text{Volume of sample taken for estimation} \times 1000} \times 100$$

Table 1: Passport data of *Cucumis sativus* var. *hardwickii* germplasm collected

Accns. No.	Coll. No.	Habitat	Village	Mandal	Mandal	Latitude	Longitude
IC 614592	ND/DC-07	Disturbed	KapadeBudru	Poladpur	Raigarh	17°57' 272	70°30' 138
IC 614593	ND/DC-15	Wild	Kumbawe	Dapoli	Ratnagiri	17°44' 385	73°18' 195
IC 614594	ND/DC-16	Wild	Kumbawe	Dapoli	Ratnagiri	17°44' 385	73°18' 195
IC 614595	ND/DC-24	Wild	Kodouli	Rajapur	Ratnagiri	16°39' 972	73°31' 570
IC 614596	ND/DC-28	Wild	Khingini	Rajapur	Ratnagiri	16°72' 354	73°48' 651

RESULTS AND DISCUSSION

C. sativus L. var. *hardwickii* occurs in open grasslands especially in farm borders near fences, bushes and have creeper climbing habit. The area explored is situated between 16°72' 354"- 18°35' 836" latitude and 70°27' 688"- 73°48' 651" longitude. The general topography of the area is hilly to undulating and the climate is warm and humid and annual rainfall varies from 2,500 mm (Raigadh) - 3,713 mm (Ratnagiri). The plants are locally known as (Karate/ Karit) in Raigadh and Ratnagiri districts of Maharashtra. The plants are dark green with light green colour fruits, early maturing and bearing fruit during rainy season. Variability was recorded in fruit shape and size and was mostly medium sized, round and in some cases elongated in shape. Fruit colour ranged from whitish green to greenish white, while the fruit surface was smooth (Fig. 2). Data revealed a wide a range of variation in mean green fruit weight with a range of variation from (33.60g) IC 614596 (ND/DC-28)-152.79 g) IC 614594 (ND/DC-16). Flesh/rind thickness varied from 0.26mm in IC 614595 (ND/DC-24) - 1.24mm IC 614594 (ND/DC-16). Mean fruit length exhibited a range of variation from 44.88 IC 614596 (ND/DC-28) - 97.54 mm IC 614594 (ND/DC-16). Mean fruit width ranged from 36.32 mm IC 614596(ND/DC-28) - 52.89 mm IC 614594 (ND/DC-16) whereas mean fruit perimeter was 14.14 mm with a range of 12.78 mm IC 614595 (ND/DC-24)- (17.42 mm) IC 614594 (ND/DC-16). Variability was also observed in other fruit character such as seeds per fruit (199.8 – 302.8), Seed length (5.16 mm- 8.9 mm), seed width (2.02 mm- 2.62 mm) and 100 seed weight (0.53- 1.19 g). Data on fruit weight and

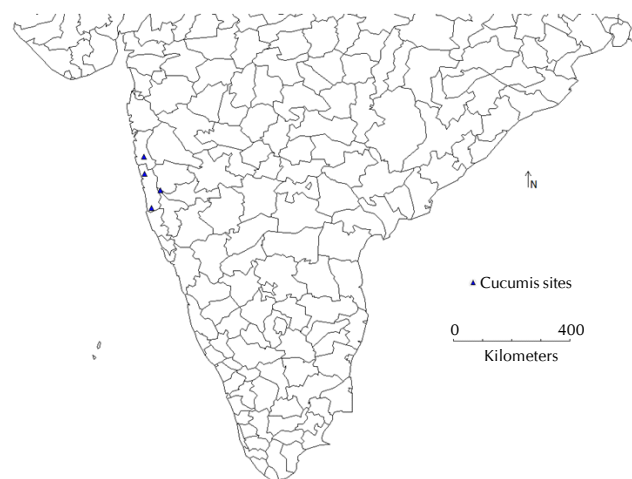


Figure 1: DIVA-GIS map showing the collection sites of *Cucumis sativus* var. *hardwickii* in Maharashtra

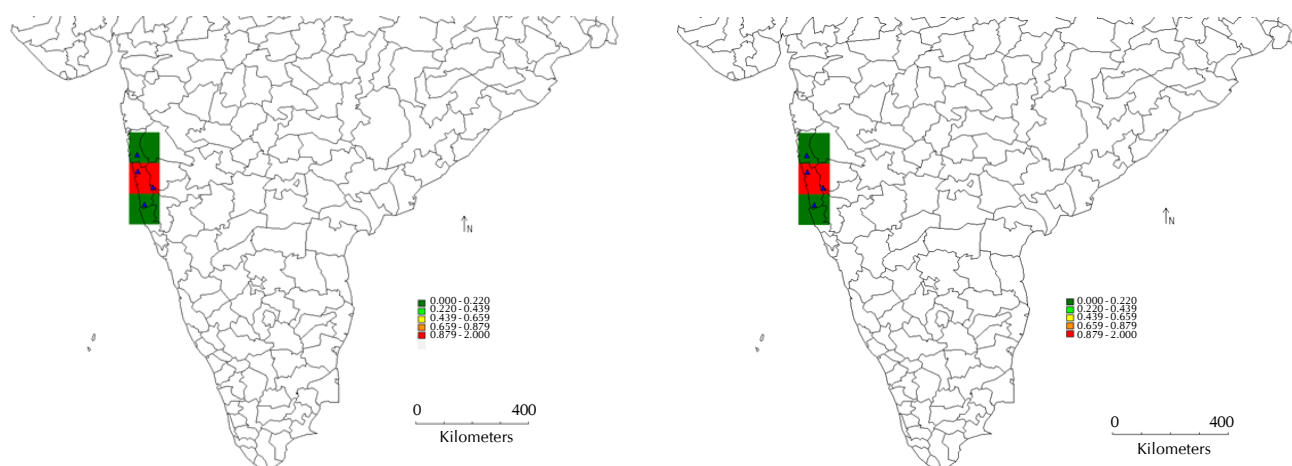


Figure 2: *Cucumis sativus* var. *hardwickii* showing habitat, fruit variability and transverse section of fruits

Table 2: Fruit characteristics and Physico-chemical analysis of *Cucumis sativus* var. *hardwickii* of Ratnagiri and Raigadh district of Maharashtra

Collectors Number	No. of stripes fruit ¹	FLT (mm)	FWD (mm)	FRC (mm)	FRW (g)	FRT (mm)	NSF	SLT (mm)	SWD (mm)	100 SWT	TSS (°Brix)	Acidity (%)	Moisture (%)
ND/DC-7	8.4	48.84	39.52	13.56	35.47	0.46	206.8	5.49	2.02	0.58	13.7	0.51	97.0
ND/DC-15	9.8	55.11	42.42	14.1	49.86	0.39	210.6	6.12	2.44	0.80	3.2	0.64	95.42
ND/DC-16	10.0	97.54	52.89	17.42	152.79	1.24	302.8	8.9	2.62	1.19	3.3	0.64	94.41
ND/DC-24	9.2	49.46	37.15	12.78	36.02	0.26	236.0	5.54	2.16	0.53	10.3	0.76	96.87
ND/DC-28	9.4	44.88	36.31	12.86	33.60	0.32	199.8	5.16	2.3	0.59	3.1	0.57	94.92
Descriptive statistical analysis													
Minimum	8.4	44.88	36.31	12.78	33.60	0.26	199.8	5.16	2.02	0.53	3.1	0.57	94.4
Maximum	10.0	97.54	52.89	17.42	152.79	1.24	302.8	8.9	2.62	1.19	13.7	0.76	97.0
Mean	9.36	59.17	41.66	14.14	61.55	0.53	231.2	6.24	2.31	0.74	6.72	0.62	95.72
Standard deviation	0.62	21.76	6.71	1.91	51.42	0.40	42.3	1.53	0.23	0.27	4.97	0.09	1.16
CV %	6.7	36.8	16.1	13.5	83.5	75.2	18.3	22.4	10.2	37.0	73.9	15.0	1.2

No. of Stripes/ fruit, FLT-Fruit Length (mm), FWD-Fruit width (mm), FRC-Fruit circumference(mm), FRW-Fruit ; Weight (g), FRT-Flesh/ rind thickness , NSF-No. of seeds/ fruit, SLT-Seed length, SWD-Seed width, 100 seed wt. ; TSS (°Brix), Acidity (%), Moisture(%)

**Figure 3: Diversity analysis Grid map for the traits total soluble solids and Fruit weight in *Cucumis sativus* var. *hardwickii***

fruit length of IC 614594 (ND/DC-16) expressed higher value than the earlier findings (Munshi *et al.*, 2007, Dwivedi *et al.*, 2010 and Dikshit, 2014a). Ward's minimum variance dendrogram generated based on quantitative traits for the *Cucumis sativus* var. *hardwickii* revealed two major clusters. Out of five accessions IC 614594 (ND/DC-16) collected from Kumbawe village, Dapoli Mandal of Ratnagiri stand alone as unique and other four accessions clustered together with 2 sub-clusters within (Fig. 5).

DIVA-GIS map generated for the collection sites of *Cucumis* germplasm is depicted in Fig. 1. The collected germplasm showed immense variation in fruit shape and seed morphological values (Table 2). Distribution pattern of different wild species of *Cucumis* were observed by Dwivedi *et al.* (2010) in parts of Aravali ranges of Rajasthan during different exploration trips. DIVA-GIS grid maps generated for the specific traits (TSS and fruit weight) revealed the presence of diversity among the *Cucumis sativus* var. *hardwickii* germplasm (Fig. 3). The results for total fruit weight, acidity of fruit and total soluble solids should be taken into consideration for increasing the yield potential (Jat *et al.*, 2014). Besides the general variability for morphological characters, the collection of *Cucumis sativus* var. *hardwickii* showed considerable

variability in specific characters. The utilization/knowledge on the extent of variability in the morphological and biological characters serve as a base for use of germplasm in modern breeding (Kristkova *et al.*, 2003). The observations recorded on morphological characters of the germplasm collections showed a wide range of variability. The Ecocrop model for assessing the suitable cultivation areas for the cultivated species *Cucumis sativus* in India is provided in Fig. 4. Majority of the Western Ghats region are suitable for the cultivation of *C. sativus*. However, crop modeling needs to be performed for ascertaining the potential distribution of *Cucumis sativus* var. *hardwickii* species. GIS mapping may be effectively used for documentation, diversity analysis, identifying gaps in collection, assessment of loss of diversity, developing new strategies for conservation, and sustainable utilization, particularly in the wake of recent international developments related to food and nutritional security. GIS mapping has been successfully used in assessing biodiversity and in identifying areas of high diversity in *Phaseolus* bean (Jones *et al.*, 1997); wild potatoes (Hijmans and David, 2001); horsegram (Sunil *et al.*, 2008); *Jatropha curcas* (Sunil *et al.*, 2009); linseed (Sivaraj *et al.*, 2009); blackgram (Babu Abraham *et al.*, 2010); *Canavalia* fatty acids (Sivaraj *et al.*, 2010); medicinal plants (Varaprasad

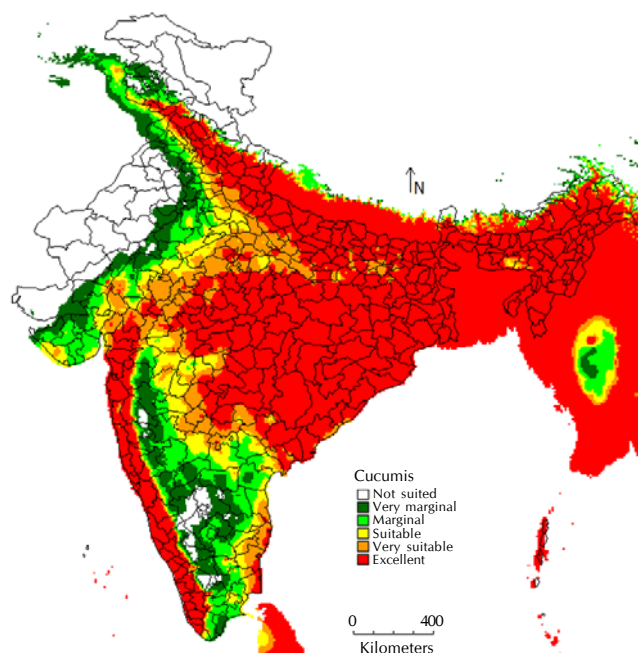


Figure 4: Ecocrop Model showing *Cucumis sativus* (cultivated species) suitability in India

et al., 2007), wild pigeon pea (Dikshit *et al.*, 2014b), brinjal (Rameash *et al.*, 2015) and agrobiodiversity (Varaprasad *et al.*, 2008). In the present research work, it is inferred that fruit and seeds specific traits emerged as the most apparent and distinguishing morphological traits. Study shows that *Cucumis sativus* var. *hardwickii* in India is very divergent and rich in variability.

Augmentation of *Cucumis* genetic resources should be taken up in a consortium mode and a wide range of samples from different regions of India be collected, characterized, the changes in spatial and temporal variation could be associated with geographical origin for its distribution, variability and diversity. In order to discover distribution of cultivated and wild forms in the current climate change regime, modelling (e.g. Maximum entropy (MaxEnt), Eco crop) should be performed for wild and cultivated forms.

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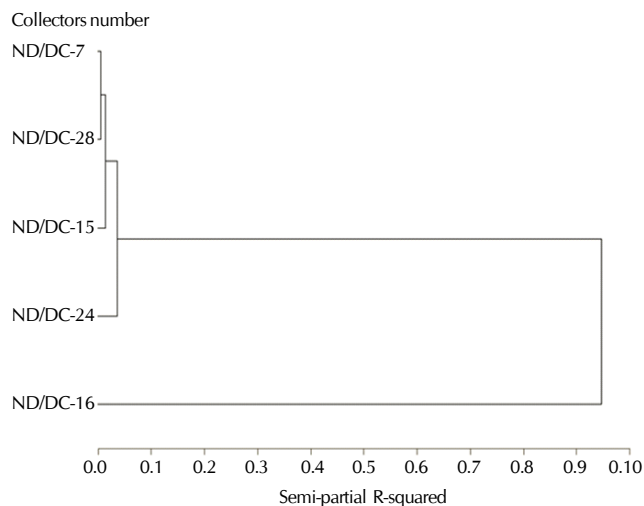


Figure 5: Ward's Minimum variance dendrogram generated for *Cucumis sativus* var. *hardwickii*

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