Agric Res
DOI 10.1007/s40003-017-0264-8

FULL-LENGTH RESEARCH ARTICLE

Island Ecosystems Host Rich Diversity in Coconut (*Cocos nucifera*): Evidences from Minicoy Island, India

B. A. Jerard[1] · M. K. Rajesh[1] · Regi Jacob Thomas[1] · V. Niral[1] · K. Samsudeen[1]

Received: 12 July 2016 / Accepted: 6 April 2017

Abstract Oceanic island ecosystems present immense opportunities for the study of species evolution due to their isolated geographical nature and presence of highly rich species diversity. Assessment of genetic diversity and population structure of four distinct coconut groups, viz. Giant, Ordinary, Micro and Mini Micro types of Minicoy Island, India, was carried out using morphological traits and microsatellite markers. The morphological data set, analysed using principal component analysis, revealed high genetic variability for fruit component traits. The occurrence of Laccadive Mini Micro Tall palms, bearing the smallest coconuts in the world, was observed in the island. The nuts of these palms had a low copra content of 5 g/nut, but high oil content of around 73%. A total of 70 alleles were detected among the four distinct coconut groups of the Island using 19 polymorphic microsatellite markers with a mean of 3.68 alleles per locus. The fixation index ranged from 0.153 to 0.424 indicating highly variable levels of inbreeding in these populations. Pair-wise population matrix formed by Nei's genetic identity showed that Laccadive Mini Micro Tall was genetically distinct from all other groups. The study revealed the presence of rich coconut diversity in islands and highlights the importance of exploration and conservation of such diverse accessions with rare genes for utilizing them in crop improvement.

Keywords Coconut · Island · Diversity · Morphological markers · microsatellite markers

Introduction

Cocos nucifera L. is one of the important palm species widely grown in tropical regions and almost all of its parts from root to terminal bud are used by human and hence called the '*Tree of Life*'. Cultivated in more than 93 countries, it sustains the livelihood of millions of people in these regions and plays a key role in protecting many of the fragile island ecosystems in tropical region. The coconut palms are broadly classified into two major forms, viz. talls and dwarfs. The tall cultivars are predominantly grown for fresh, oil yielding kernel, whereas the dwarf cultivars for their attractive bright-coloured tender fruits having sweet tender nut water. Besides, the fruits also serve as raw materials for many food preparations and industrial products. The roots, shell, tender nuts and male flowers are also used in the preparations of several Indian Ayurvedic and folk medicines.

Conventional breeding approaches in coconut have led to the development of many improved varieties, comprising better performing selections from different tall and dwarf coconut populations and hybrids among them. To satisfy the dynamic needs of crop improvement efforts in coconut, it is imperative to collect and conserve genetic resources from diverse coconut populations. The tropical island ecosystems have been the source of diverse coconut populations and form the bulk of collections conserved and maintained in the National and International Coconut Gene Banks in coconut growing countries. Natural coconut populations, growing in remote regions, are most likely less subjected to human interventions, especially in the selection process. These populations, therefore, offer a good scope for identification of novel and diverse traits which could be utilized for future

✉ B. A. Jerard
jerardba@gmail.com

[1] ICAR-Central Plantation Crops Research Institute, Kasaragod 671124, Kerala, India

Published online: 17 May 2017

crop improvement programmes. Earlier explorations in Andaman and Nicobar Islands, India, have resulted in identification and collection of rare and useful coconut genotypes [25, 26, 41].

The Lakshadweep Islands of Indian Ocean consists of 36 coral islands covering 12 atolls, three reefs and submerged sand banks of which only 11 islands are inhabited [10]. Geologically, the Lakshadweep group of islands are considered to be a continuation of rocks of Rajasthan and Gujarat through the banks of the Gulf of Cambay and through the Agaria banks further south [50]. Lakshadweep Islands rank first among the Indian states and union territories in coconut productivity in terms of number of nuts per hectare area [5], indicating the potential existing in the native coconut population in these islands. The Laccadive Ordinary Tall, Laccadive Micro Tall, Kaithathali Tall and Laccadive Orange Dwarf are the main cultivars in these islands [24]. The coconut population of Lakshadweep Islands consists of palms with large- to medium-sized nuts (commonly known as Lakshadweep Ordinary Talls) and small nuts (Micro Talls). The Laccadive Ordinary Tall is the widely cultivated type, while the Laccadive Micro type is found sporadically amidst the Laccadive Tall populations [12]. The Laccadive Micro Tall has been identified as the genotype with higher oil content in copra [44]. Among the Micro Talls, the palms exhibit high variability for size, shape of fruits, regular or alternate bearing habit and copra content [46].

Minicoy Island, or *Maliku,* is the only inhabited island of the *Maliku* Atoll situated as southernmost island of Lakshadweep archipelago and is separated from the rest of the islands by a 9° Channel about 180 km in width and from the neighbouring Republic of Maldives in the south by an 8° Channel of about 120 km. It is the second largest among the islands of the Lakshadweep archipelago, measuring about 10 km from its northern end to its southernmost point. The island, rising up to 2–5 m above sea level, is mostly flat and is enclosed within coral reefs. There are no natural forests, hillocks, bay creeks, estuaries, rivers, lakes or freshwater tanks, but the area is completely covered with coconut palms. The soil of the islands is thin and quite porous, which retains very little moisture and is formed mostly of fragmented coral limestone and sedimentary rocks with less water holding capacity. The soil is formed of coral debris and the parent material is organogenic calcium carbonate. In order to classify the soils of Minicoy as per soil taxonomy, a new word 'Coral' has also been coined to be prefixed before carbonatic at the family level making them unique [49, 50].

Coconut is the main cultivated crop in Minicoy Island and coconut palms occur in dense clusters or groves, resembling natural forest. During earlier explorations in Lakshadweep during 2002, only a few tall coconut types have been collected from Minicoy Island due to the remoteness of the island and difficulties in collection of nuts. In the present study, the naturally regenerating coconut stand of Minicoy Island were analysed for variations in morphological traits and molecular diversity through SSR markers with the objectives of estimating the genetic diversity within and among the observed coconut groups, determining the degree of genetic differentiation, gene flow and population structure and assessing the potential of coconut populations of Minicoy islands for further collection and conservation efforts.

Materials and Methods

Survey of Coconut Populations and Collection Sites

The dense natural coconut stands in the northern and southern parts of Minicoy Island were explored and the nut component studies conducted during the period from 2007 to 2011 during which the palms in the inhabited regions of the island were excluded (Fig. 1). The location of the study area was between 8° 15′ to 8° 20′N and 73° 01′ to 73° 05′ E, where coconut palms constitute the only vegetation cover (Fig. 2). The width of narrow coral atoll, where the exploration was conducted, ranged from a minimum of 10 m to maximum of 1.2 km with a lagoon on the one side and open sea on the other side. The annual rainfall of the study area ranged from 1000 to 1800 mm and the mean minimum and maximum temperatures ranged from 21 to 32 °C.

Morphological Characterization

A thorough survey was conducted to cover all the coconut palms in fruit-bearing phase in the explored area. Juvenile palms and young seedlings that had grown from fallen nuts were not accounted for while making observations. A total

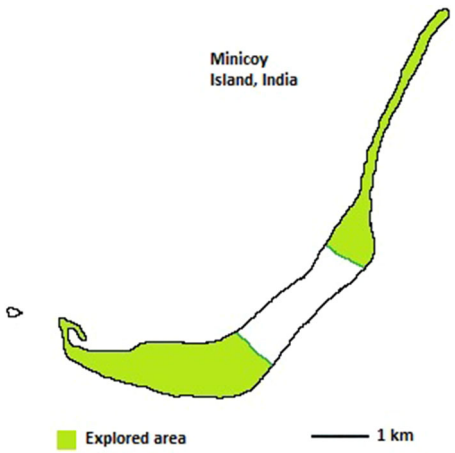

Fig. 1 Explored area of Minicoy Island for coconut diversity

Fig. 2 Coconut on coral atolls at Minicoy Island

of 2672 palms were observed and the population was broadly grouped as Laccadive Giant Tall (LCGT), Laccadive Ordinary Tall (LCT), Laccadive Micro Tall (LMT) and Laccadive Mini Micro Tall (LMMT) based on the visual observations of fruit size and cluster fruit-bearing habit. The frequency of palms under these different groups comprised of 82% LCT palms, 14% LMT palms, 3% LCGT palms and 1% LMMT palms. Wide variations were observed for the crown, fruit shape and fruit size in the Minicoy coconut population (Fig. 3a–h). Thirty-two selected palms of each group, which exhibited regular bearing of fruits, were observed for vegetative and reproductive traits, and the average of the 32 palms was worked out for analysis. Mature fruit samples from the selected palms within the groups were used for fruit component analysis, and the descriptor traits were recorded as per the standard procedures for coconut [23, 44].

The vegetative traits observed on the selected palms were plant height (PH), number of leaf scars in 1 m length of trunk at the bottom (NLS), girth of trunk at 1 m height (GT), number of leaves on the crown (NLC), length of leaf (LL), length of leaflet-bearing portion on the leaf (LLP), number of leaflets on one side of leaf (NLFT), length of leaflet (LLFT), breadth of leaflet (BLFT), number of bunches on crown (NBC), number of female flowers per bunch (NFF), number of fruits per bunch (NF), length of inflorescence (LINF), number of spikelets per inflorescence (NSP) and length of spikelet (LSP). The observations on leaves were recorded on the oldest leaf on the crown of the selected palms of each type, viz. LCGT, LCT, LMT and LMMT (32 palms each; a total of 128 palms). The observations on floral traits were recorded on the same palms on five inflorescences of each group and means were obtained. The actual number of leaves and number of bunches on the crown were recorded on all the palms during the exploration in 2007.

The fruit component traits observed were fruit weight (FW), fruit polar circumference (FPC), fruit equatorial circumference (FEC), husk thickness (HUST), dehusked fruit weight (DFW), dehusked fruit polar circumference (DFPC), dehusked fruit equatorial circumference (DFEC), shell thickness (SHT), kernel thickness (KET), cavity volume (CAV), shell weight (SHW), copra weight (COW) and oil content (OIL). Five 12-month-old mature fruits of each type, viz. LCGT, LCT, LMT and LMMT, were collected from identified palms, stored for 20 days under shade for drying the husk moisture and then used for fruit component analysis. The kernel extracted from the fruit samples was further dried in oven at 40 °C for 72 h, and the oil content was estimated using distillation method with petroleum ether.

The mean data on the 28 vegetative and fruit component observations were analysed for the variations within and between the identified groups. Mean values and standard deviations were calculated for each of the morphological characters in each group. Principal component analysis (PCA) was applied using SAS software package to analyse morphological variation and to assess the differences between and within the identified groups. PCA is particularly relevant to identify variables which most contribute to the value of each principal component.

Genetic Diversity Through SSR Analysis

Total DNA was extracted from spindle leaves of 64 palms (17 each of LCT, LCGT and LMT and 13 palms of LMMT) using a modified SDS method. DNA was extracted from approximately 1 g of fresh young leaves, collected from field grown parental plants and crushed in liquid nitrogen as per the standard procedures [42]. The precipitated DNA was air-dried and dissolved in 0.75 ml TE buffer. The quality and quantity of the extracted DNA was estimated using agarose gel electrophoresis and also using a spectrophotometer. The isolated DNA was then diluted in TE buffer for further analysis. Microsatellite analyses were conducted as described in earlier studies [41] using a set of 19 hyper-polymorphic coconut SSR markers, distributed in different coconut chromosomes.

The alleles were scored individually based on comparison with the molecular ladder. Observed number of alleles, effective number of alleles, Shannon's Information Index and F-Statistics were worked out for the 19 microsatellite loci using the software GenAlEx version 6.5 [36]. The software was also used to calculate the expected and observed homozygosity and heterozygosity across the 19 microsatellite loci, genetic identity, average heterozygosity and Nei's genetic distance [34]. The degree of population structure over all loci was estimated with

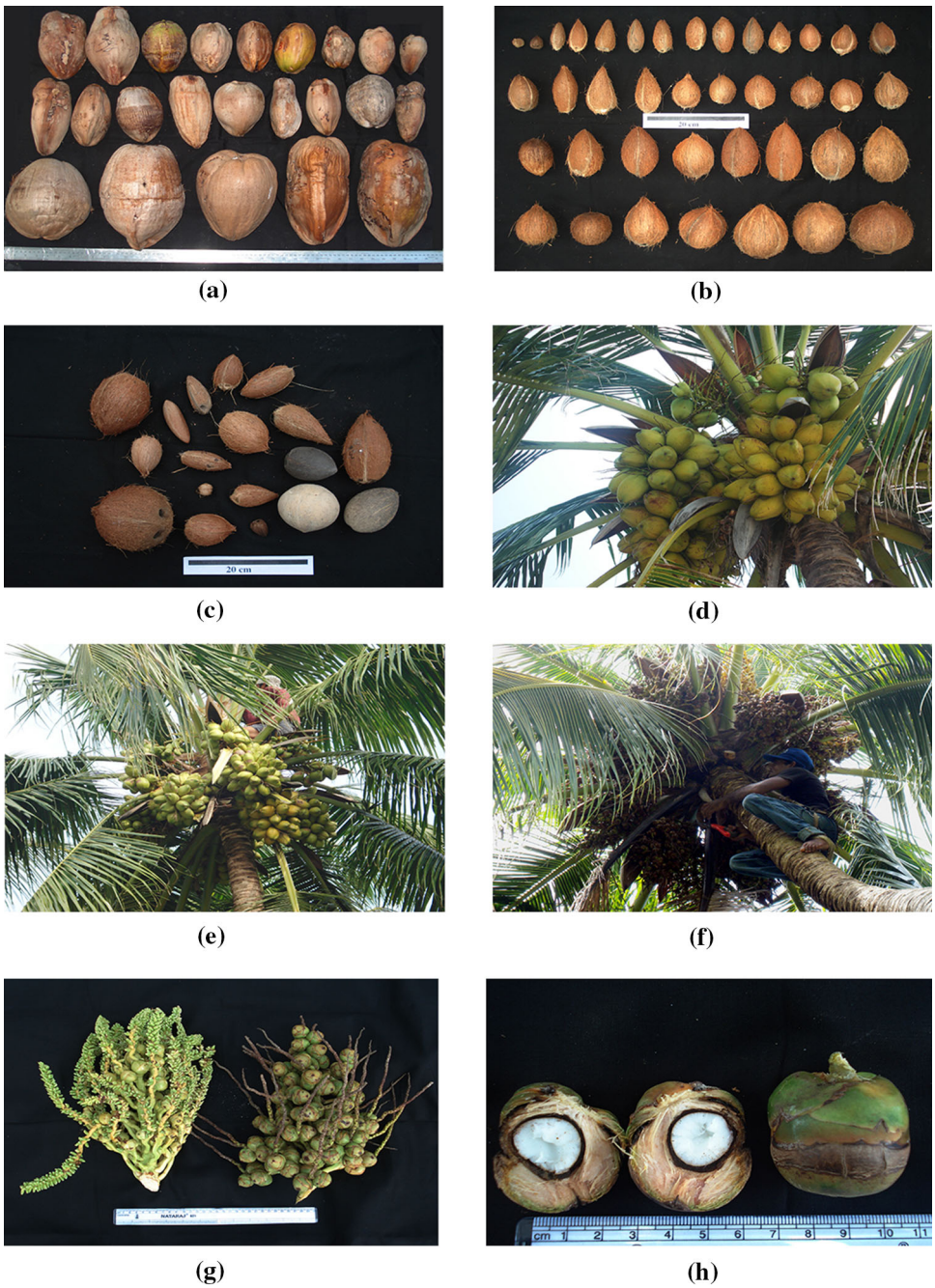

Fig. 3 Morphology of Minicoy coconut population. **a** to **c** Variability for fruit shape and size. **d** Crown of LCT. **e** Crown of LMT. **f** Crown of LMMT. **g** and **h** Bunch and fruits of LMMT

fixation indices (F_{ST}, F_{IT} and F_{IS}), G'_{ST}, G''_{ST} [51] and genetic differentiation (D_{est}) [27, 33], testing the null point by random permutation, and estimating variances via jackknifing and bootstrapping over loci.

The expected and observed heterozygosity and the fixation index across the four coconut populations were worked out using the Genetic Data Analysis (GDA) software [31]. A cluster analysis was performed on the similarity matrix using the unweighted pair group method with arithmetic averages (UPGMA), and the resultant phenogram was configured.

The data sets were tested for deviations from Hardy–Weinberg equilibrium (HWE) in GENEPOP version 4.0 (http://genepop.curtin.edu.au/), using a Markov chain approximation to exact tests and likelihood ratio tests, respectively. Deviations from HWE were estimated using both the exact test and the F_{IS} statistic estimations, using Markov chain Monte Carlo (MCMC) runs for 1000 batches, each of 2000 iterations, with the first 500 iterations discarded before sampling [17]. Whenever multiple testing was performed, probability values were corrected by using standard Bonferroni corrections [45].

Analysis of Population Structure and Differentiation

The population structure of the coconut populations were explored using the clustering method STRUCTURE 2.3.3 [16]. This method assumes that a sample of individuals comprises of K unknown populations to which individual genotypes or fractional genotypes can be assigned. The admixture model of STRUCTURE and the option of correlated allele frequencies between populations were used. The correct number of clusters (K) was determined by testing K values from 1 to 10 and performing 15 repetitions for each K. The burn-in period consisted of 1×10^5 iterations followed by 1×10^5 MCMC repeats. Finally, estimated log probabilities of data Pr ($X \mid K$) for each value of K were evaluated by calculating ΔK, the rate of change in the log probability of data between successive K values [13]. The STRUCTURE output files were first processed using STRUCTURE HARVESTER version 0.3, which produces an output consisting of a series of files, including graphical files representing, per K and per repeated run, the estimated Ln probability of each run, and three other Ln-based estimates that allow the selection of the most optimal value for K [13]. Samples were analysed without any prior population information, but were sorted by their sampling population once STRUCTURE is completed.

In order to estimate the variance between the groups of populations, pooled sample structuring was estimated using analysis of molecular variance (AMOVA) [14] and 20,000 permutations implemented in Arlequin version 3.5.1.

Results

Morphological Characterization

The observations on coconut population of Minicoy Island revealed wide variability for fruit size, which could be easily differentiated into four major fruit types, based on phenotypic observations of fruit size and fruit-bearing habit. Wide variability was observed for fruit component traits among the dehusked and husked fruits of these four types, viz. LCGT, LCT, LMT and LMMT (Fig. 3). The morphological and fruit component traits recorded among the coconut groups (Table 1) revealed that there was a significant difference among the groups for all the observed traits, except for NLFT. The fruit component traits from FW to OIL and LINF differentiated the groups completely, while the remaining morphological traits exhibited partial differences and grouping. PCA performed for 28 traits revealed that the cumulative contribution of the three components was 75.502% of the total variability among the groups of coconut populations (Table 2). PC 1 was found positively correlated with fruit component traits FW, FPC, FEC, HUST, DFW, DEPC, DFEC and SHW, whereas the morphological traits registered either non-significant positive or negative correction with PC 1, indicating that the axis was determined by fruit component traits. The second axis was positively correlated with morphological traits NLC, LL, LLFT, BLFT, NFB, LINF, NSP and LSP, whereas it registered non-significant positive or negative correlation for fruit component traits. PC 3 recorded a positive and significant correlation with GT, NLC, NBC and CAV. These results indicate that the fruit component traits contribute maximum to the variation among the groups followed by vegetative traits.

Observations obtained from 32 individuals of each group were used to draw a phenogram (figure not shown) based on the unweighted pair group method arithmetic (UPGMA) using a Euclidian distance matrix. The coconut populations were grouped into two major clusters in which the LCGT formed cluster I and LCT, LMT and LMMT formed the second cluster within which LMT and LMMT were grouped together in a sub-cluster. However, the morphological differences among the groups significantly differed and the mean values for the fruit component traits indicated wide range within and between the groups. The LCGT group was characterized by large, heavier nuts with higher cavity volume, shell weight and copra weight. The palms of LCGT recorded greater girth of trunk with more of leaflet-bearing portion on the leaves and broader leaflets, suggesting a robust appearance of palms. They are also characterized by lower number of female flowers and fruits per bunch. The LCT group was characterized with taller plant height, medium-sized fruits with moderate fruit and copra weight. The palms of LCT were observed to be in medium range for most of the traits when compared to LCGT and LMT palms. The LMT group was characterized by the retention of more number of leaves on the crown, lengthy leaves, higher number of female flowers and more number of fruits per bunch. The palms exhibited cluster-bearing habit with more number of fruits per spikelet on longer inflorescences. The nuts of LMT were smaller with lesser cavity volume and copra and had higher oil content compared to LCGT and LCT. The LMMT group was characterized with shorter palms exhibiting more number of leaf scars over the stem, more number of leaflets, higher number of bunches on the crown and higher number of female flowers per inflorescence. The fruits of the LMMT are unique with the lowest values for fruit size, fruit weight, husk thickness, shell thickness, kernel thickness, cavity volume, shell weight and copra weight. However, higher variability was observed within this group also for many traits as like other groups. The oil content in LMMT (73%) was the highest recorded among the Minicoy coconut populations.

Table 1 Morphological and fruit component traits scored in the four groups of Minicoy Island coconut population

Traits	LCGT	LCT	LMT	LMMT	Statistical parameters	
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	*F*	CD
PH (m)	13.2 ± 2.0b	15.1 ± 1.8a	12.6 ± 1.3b	9.5 ± 2.4c	48.11	1.02
NLS	14.2 ± 2.0b	14.3 ± 1.9b	12.9 ± 2.1c	19.9 ± 3.7a	46.14	1.48
GT (cm)	102.9 ± 6.8a	85.3 ± 6.5b	89.5 ± 6.6b	87.7 ± 17.2b	18.42	5.65
NLC	33.2 ± 3.2a	30.3 ± 2.6b	34.5 ± 4.5a	27.6 ± 5.7c	17.75	2.24
LL (cm)	433.3 ± 33.0a	416.2 ± 47.4b	443.5 ± 27.0a	366.6 ± 63.9c	18.26	24.28
LLP (cm)	358.3 ± 32.6a	319.7 ± 42.8b	336.6 ± 36.2b	279.7 ± 44.0c	23.00	21.09
NLFT	103.6 ± 8.4a	105.7 ± 7.4a	110.1 ± 5.9a	111.4 ± 8.9a	1.99	7.99
LLFT (cm)	106.8 ± 5.1a	99.3 ± 7.0b	109.0 ± 5.8a	92.1 ± 18.5c	16.77	5.71
BLFT (cm)	6.2 ± 0.6a	4.8 ± 0.8c	5.6 ± 0.4b	4.8 ± 0.8c	33.30	0.36
NBC	11.7 ± 2.3b	11.2 ± 1.8c	13.1 ± 1.5b	18.3 ± 6.4a	26.07	1.93
NFF	24.8 ± 5.7b	35.5 ± 7.6b	117.7 ± 37.3a	105.0 ± 66.0a	49.21	20.60
NFB	6.9 ± 2.5b	11.6 ± 2.5b	69.5 ± 19.0a	9.9 ± 11.6b	227.40	6.07
LINF (cm)	96.4 ± 7.5b	79.3 ± 6.9c	108.6 ± 7.1a	69.6 ± 17.9d	77.79	6.01
NSP	30.9 ± 6.9c	31.7 ± 3.9c	49.2 ± 5.4a	42.6 ± 8.9b	59.05	3.57
LSP (cm)	42.4 ± 3.4a	37.6 ± 3.6b	44.4 ± 3.0a	36.9 ± 9.0b	14.84	2.95
FW (g)	1148.4 ± 127.3a	563.7 ± 136.3b	269.5 ± 69.5c	31.2 ± 5.7d	339.71	85.69
FPC (cm)	71.6 ± 1.9a	54.7 ± 3.5b	42.6 ± 3.7c	13.6 ± 2.1d	2263.26	1.56
FEC (cm)	54.4 ± 1.5a	39.3 ± 2.6b	29.3 ± 3.4c	9.2 ± 0.7d	2211.29	1.23
HUST (cm)	4.7 ± 0.2a	4.2 ± 0.3b	3.0 ± 0.5c	0.8 ± 0.1d	1183.21	0.15
DFW (g)	558.4 ± 36.8a	261.4 ± 71.8b	121.8 ± 43.1c	5.8 ± 1.2d	870.56	24.60
DFPC (cm)	37.6 ± 1.3a	29.5 ± 2.4b	23.4 ± 2.3c	4.2 ± 0.6d	1974.59	0.97
DFEC (cm)	33.5 ± 2.0a	24.2 ± 2.4b	18.8 ± 3.5c	3.5 ± 0.4d	919.89	1.26
SHT (mm)	3.5 ± 0.3b	3.8 ± 0.1a	3.0 ± 0.4c	1.5 ± 0.1d	541.91	0.13
KET (cm)	1.4 ± 0.0b	1.4 ± 0.0a	1.3 ± 0.1c	0.4 ± 0.0d	1331.72	0.04
CAV (ml)	242.0 ± 31.4a	50.3 ± 22.2b	14.8 ± 6.7c	1.1 ± 0.1d	1053.12	10.50
SHW (g)	167.6 ± 10.2a	92.8 ± 17.8b	46.1 ± 17.4c	3.6 ± 0.4d	871.52	7.24
COW (g)	294.0 ± 19.8a	106.1 ± 25.6b	52.1 ± 18.7c	5.0 ± 1.2d	1469.25	10.06
OIL (%)	67.5 ± 0.3d	68.8 ± 0.7c	70.5 ± 0.8b	73.0 ± 0.6a	460.78	0.33

Traits having values with same letters are not significantly different between groups

Allele Richness of SSR Loci

Nineteen polymorphic SSR markers (Table 3) were used to amplify DNA of 64 palms representing the four distinct coconut groups from Minicoy Island. A total of 70 alleles were detected with all the markers revealing three alleles or more with a mean of 3.68 alleles per locus. The effective number of alleles per locus (N_e) ranged from 1.130 (CnCirG4) to 3.016 (CnCirC3′) with a mean of 1.889 (Table 3). Shannon's Information Index ranged from 0.209 (CnCirG4) to 1.245 (CnCirC3′) with a mean of 0.706. The PIC value, a measure of marker diversity, varied from 0.105 (CnCirG4) to 0.673 (CnCirC3′) among the 19 microsatellite loci, the average being 0.024 (Table 3).

F_{IS} for most of the loci was high and greater than zero, with a mean of 0.045. Mean F_{IS} (0.369) and F_{IT} (0.424) were both positive and greater than zero indicating a heterozygote deficit within populations. The mean gene flow (N_m), based on mean F_{ST}, was very high (3.841) indicating an extensive gene flow among the four coconut accessions. Values of G_{ST} and D_{est} were observed to be low (Table 4). Results of the Fisher's exact test for Hardy–Weinberg (HW) equilibrium across loci, considering heterozygote deficit as the alternative hypothesis, showed that 15 of the loci had significant ($p < 0.001$) departures from HW proportions.

Genetic Diversity Within Groups and Population Structure

The fixation index ranged from 0.153 (LMMT) to 0.424 (LMT) with a mean of 0.319, indicating highly variable levels of inbreeding in these populations (Table 5). Among the accessions, expected heterozygosity was almost the

Table 2 Eigenvectors, eigenvalues and per cent variance explained by the first three principal components (PCs) for 28 traits analysed for coconut populations of Minicoy Island

Traits	Eigenvectors		
	PC 1	PC 2	PC 3
PH	0.117	0.047	−0.461*
NLS	−0.193	−0.194	0.187
GT	0.126	0.135	0.282*
NLC	0.037	0.307*	0.253*
LL	0.089	0.297*	0.016
LLP	0.127	0.165	0.129
NLFT	−0.134	0.164	−0.183
LLFT	0.065	0.355*	0.145
BLFT	0.072	0.243*	0.169
NBC	−0.199	−0.068	0.325*
NFF	−0.175	0.096	0.084
NFB	0.017	0.357*	−0.178
LINF	0.096	0.357*	0.091
NSP	−0.153	0.286*	0.012
LSP	0.095	0.265*	0.129
FW	0.244*	−0.127	0.131
FPC	0.260*	−0.025	−0.057
FEC	0.264*	−0.029	0.003
HUST	0.257*	−0.016	−0.079
DFW	0.240*	−0.125	0.149
DFPC	0.263*	0.001	−0.061
DFEC	0.260*	−0.003	0.005
SHT	0.227	−0.015	−0.239
KET	0.233	0.113	−0.231
CAV	0.200	−0.141	0.325*
SHW	0.247*	−0.117	0.118
COW	0.233	−0.124	0.231
OIL	−0.224	0.060	0.063
Eigenvalue	13.874	5.035	2.231
% of variance explained	49.551	17.983	7.968
Cumulative % of variance explained	49.551	67.534	75.502

* These traits contribute mostly to the respective principal components

same for all the accessions. The observed heterozygosity for all the accessions was less than expected indicating a tendency towards inbreeding within the population.

Pair-wise population matrix formed by Nei's genetic identity (Table 6) showed that LMMT was genetically distinct from all the other populations. A dendrogram was constricted using UPGMA clustering. Two major clusters were observed, LCGT being distinct in sub-cluster 1, while LCT and LMT clustered together in sub-cluster 1. LMMT formed a separate, distinct cluster (Fig. 4).

Using STRUCTURE program, the population structure of the coconut populations was investigated by estimating the number of genetically distinct populations (Fig. 5). An *ad hoc* statistical analysis, which was based on the second-order rate of change in the likelihood function with respect to K (ΔK) [13], was used to calculate the most appropriate K value using Structure Harvester version 0.6.92. There was a clear peak in the value of ΔK at $K = 3$.

The log probability of data ($L(K)$) for the admixture and correlated frequencies model under exhaustive sampling (averaged over 15 replicates) obtained in STRUCTURE package (Fig. 6). The highest $L(K)$ averaged over replicates running for each value of K (K from 1 to 10) was observed for $K = 3$ (−1708.58).

The locus by locus AMOVA, performed considering between populations and within populations as sources of variation (Table 7). The highest percentage of variation (92%) correspond to the within population component, while the between population component showed low magnitude (8%). Results from the principal coordinate analysis (PCoA) revealed that the first three coordinates accounted for 100% of the molecular variation. From the two-dimensional plot, the four coconut populations generally dispersed in three centred parts, reinforcing the clustering of accessions from STRUCTURE analysis.

Discussion

The wide diversity for desirable traits existing among different populations forms the basis for most crop improvement programs in coconut. However, the collection, conservation, evaluation and utilization of the coconut genetic resources is a difficult task due to inherent heterozygosity making the population highly variable, long juvenile phase of the crop, lack of viable, reproducible vegetative propagation protocols and requirement of large area and resources for genetic resources management and utilization. A large number of improved varieties for higher yield and quality attributes have been developed in many countries using these genetic resources. However, there is a need for further identification and collection of diverse materials from remote places where coconuts grow without much human selection pressure.

Morphological traits in coconut are considered important for selection of parents for hybridization and have also been extensively utilized to assess the extent of genetic diversity. Morphological variability has been reported among different coconut populations of Mexico [53–55], Papua New Guinea [35], South Pacific Islands [1], Cocos Islands [30], Indian Ocean Islands [29], Andaman and Nicobar Islands [25]. The pattern of morphological variability observed in the Minicoy coconut populations is similar to those from other places; however, the level of occurrence of exceptional types such as Micro (LMT) and

Table 3 Locus-wise data on number of alleles, effective alleles (N_e), Shannon's Information Index (I), polymorphism information content (PIC), probability of deviation from Hardy–Weinberg Equilibrium (P) are reported

Sl. No.	Locus	Number of alleles	N_e	I	PIC	P
1.	CnCirG4	3	1.130	0.209	0.105	0.0040
2.	CnCir73	4	1.171	0.282	0.148	0.0000
3.	CnCirG11	3	1.417	0.415	0.257	0.0000
4.	CnCir86	4	1.989	0.780	0.469	0.0000
5.	CnCirE10	4	1.781	0.732	0.452	0.0000
6.	CnCirC3′	6	3.016	1.245	0.673	0.0000
7.	CnCir74	4	1.823	0.805	0.455	0.0000
8.	CnCir87	4	1.764	0.747	0.435	0.1224*
9.	CnCir56	3	1.966	0.801	0.507	0.0011
10.	CnCirH7	3	1.694	0.659	0.398	0.0011
11.	CnCirC7	3	2.821	1.063	0.668	0.0019
12.	CnCirB6	5	2.606	1.063	0.622	0.0000
13.	CnCirH9′	4	2.426	0.998	0.589	0.0013
14.	CnCirK1	3	1.765	0.608	0.437	0.0246*
15.	CnCir2	3	1.230	0.321	0.172	0.1075*
16.	CnCirE4′	4	2.325	0.974	0.575	0.0000
17.	CnCirE11	4	2.300	0.922	0.532	0.0011
18.	CnCirA3	3	1.518	0.547	0.330	0.0025*
19.	CnCirE7	3	1.153	0.242	0.132	0.0093*
	Mean		1.889	0.706	0.419	

* No significant departures from HW equilibrium

Table 4 Locus-wise data on F statistics (F_{ST}, F_{IT}, F_{IS}), gene flow (N_m), G_{ST}, D_{est}

Locus	Number of alleles	F_{IS}	F_{IT}	F_{ST}	N_m	G_{IS}	G_{ST}	$G'_{ST}N$	$G'_{ST}H$	G''_{ST}	D_{est}
CnCirG4	3	0.512	0.538	0.053	4.465	0.536	0.016	0.021	0.018	0.024	0.003
CnCir73	4	0.757	0.763	0.025	9.829	0.771	−0.020	−0.026	−0.024	−0.031	−0.005
CnCirG11	3	0.564	0.607	0.098	2.294	0.588	0.058	0.076	0.086	0.104	0.030
CnCir86	4	0.394	0.458	0.105	2.139	0.427	0.065	0.084	0.144	0.162	0.085
CnCirE10	4	0.764	0.812	0.203	0.982	0.778	0.164	0.207	0.353	0.386	0.226
CnCirC3′	6	0.338	0.358	0.029	8.307	0.368	−0.006	−0.008	−0.022	−0.024	−0.016
CnCir74	4	0.482	0.611	0.250	0.750	0.507	0.218	0.271	0.469	0.505	0.321
CnCir87	4	0.028	0.073	0.046	5.173	0.061	0.020	0.027	0.041	0.048	0.021
CnCir56	3	0.275	0.310	0.048	4.914	0.307	0.015	0.020	0.036	0.041	0.022
CnCirH7	3	0.345	0.476	0.199	1.004	0.376	0.168	0.212	0.319	0.355	0.181
CnCirC7	3	0.278	0.296	0.025	9.676	0.310	−0.009	−0.012	−0.035	−0.038	−0.025
CnCirB6	5	0.473	0.537	0.121	1.815	0.500	0.085	0.110	0.279	0.299	0.213
CnCirH9′	4	0.173	0.254	0.099	2.287	0.208	0.068	0.088	0.199	0.217	0.141
CnCirK1	3	0.248	0.335	0.116	1.907	0.281	0.084	0.109	0.173	0.195	0.097
CnCir2	3	0.132	0.177	0.052	4.584	0.167	0.022	0.029	0.028	0.035	0.006
CnCirE4′	4	0.364	0.415	0.080	2.882	0.395	0.044	0.058	0.126	0.139	0.086
CnCirE11	4	0.145	0.220	0.087	2.619	0.181	0.057	0.074	0.144	0.160	0.092
CnCirA3	3	0.358	0.403	0.069	3.351	0.390	0.032	0.043	0.054	0.064	0.022
CnCirE7	3	0.388	0.407	0.031	7.837	0.419	−0.007	−0.010	−0.009	−0.012	−0.002
Mean		0.369	0.424	0.091	3.841	0.376	0.066	0.086	0.131	0.150	0.070

Table 5 Population-wise data on Number of different alleles (N_a), effective alleles (N_e) and private alleles, observed (H_o) and expected (H_e) heterozygosity, fixation index (F) and percentage of polymorphic loci (% P)

Group	N_a	N_e	No. of private alleles	H_o	H_e	F	% P
LCGT	3.00	1.835	0.15	0.269	0.399	0.287	95
LCT	2.60	1.863	0.10	0.242	0.413	0.408	90
LMT	2.95	1.760	0.10	0.225	0.401	0.424	95
LMMT	2.80	1.921	0.10	0.323	0.404	0.153	90
Mean	2.838	1.845	0.1125	0.265	0.404	0.319	92.50
SE	0.179	0.077	0.078	0.021	0.023	0.041	1.44

Table 6 Pair-wise population matrix of coconut groups of Minicoy Island, Lakshadweep (based on Nei's genetic identity)

Group	LCGT	LCT	LMT	LMMT
LCGT	1.000			
LCT	0.931	1.000		
LMT	0.929	0.942	1.000	
LMMT	0.866	0.898	0.879	1.000

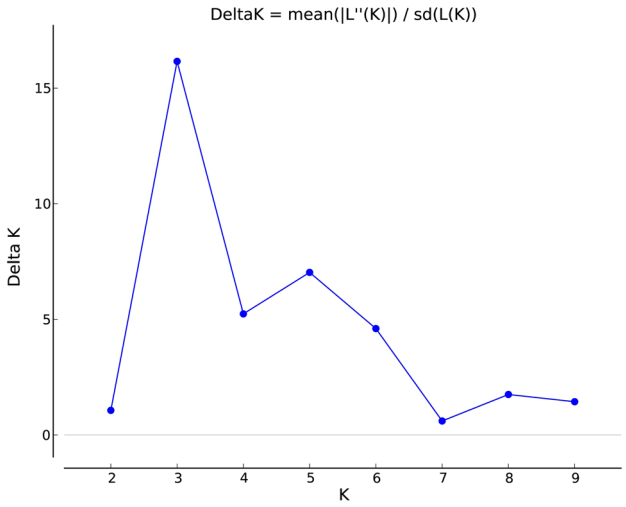

Fig. 5 Estimated population structure of coconut groups from Minicoy Island using STRUCTURE

Mini Micro (LMMT) palms is unique. Although a few LMMT palms were earlier reported from Minicoy islands [9], the present survey of the area indicated the presence of more number of LMMT palms with higher variability within the population and the LMMT palms are distinct from other palms for most fruit component traits. The occurrence of LMT palms in other Islands of Lakshadweep archipelago has been studied earlier [46], and it was reported that they are closely related to LCT population and might have developed through introgression between LCT variants having different fruit size and husk content. The sporadic occurrence of LMT in Agatti and Kavaratti Islands of Lakshadweep was earlier reported [12] and the morphological observations reported earlier are in line with the present results. However, the presence of Mini Micro palms (LMMT), which bears the smallest coconuts in the world [8], was observed to be occurring only in Minicoy Island and has not been reported from elsewhere. The fruits of LMMT palms recorded very low fruit weight and size with only a few drops of nut water, but the embryos were seen to be normally sized like in other groups. These fruits do not germinate naturally due to very less nut water and endosperm to support the growth and therefore require the embryo culture technique for its rescue. A few embryo-cultured plantlets of this type have been conserved in the National field gene banks for coconut at ICAR-CPCRI [6].

Fruit component analysis of Lakshadweep coconut populations has earlier been reported from other islands of Lakshadweep [4, 28]. The studies classified the tall forms from the Lakshadweep Islands as Niu Kafa types characterized by high husk and endosperm proportions of the total fruit weight in contrast to low husk and endosperm proportions of Niu Vai types [20]. Based on a study of fruit components of Laccadive Ordinary Tall and Laccadive Micro Tall coconut populations from Kadmat and Amini

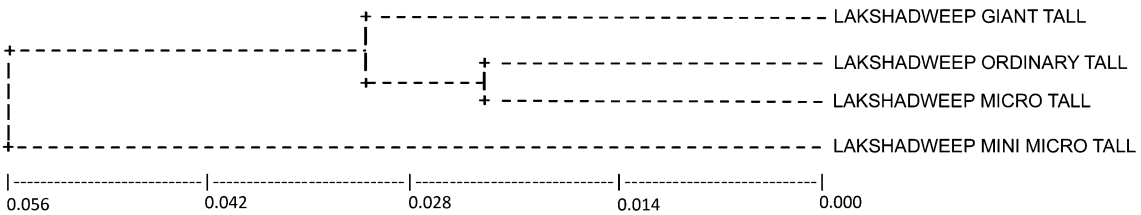

Fig. 4 UPGMA dendrogram showing the relationship among the coconut groups from Minicoy Island

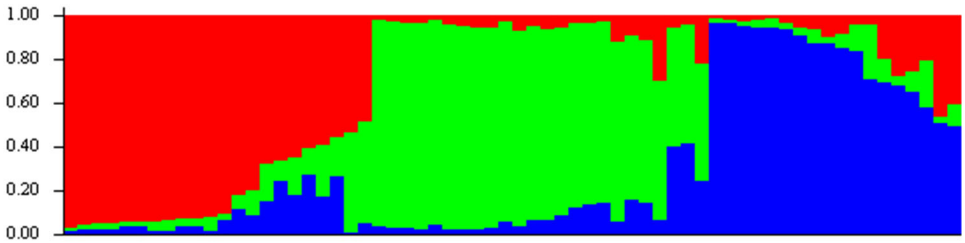

Fig. 6 Log probability for the admixture and corrected frequencies of observed coconut groups from Minicoy Island using STRUCTURE [13]

Table 7 AMOVA [14] among populations and within populations

Source	*df*	SS	MS	Est. var.	%
Among populations	3	103.004	34.335	1.281	8
Within populations	60	835.167	13.919	13.919	92
Total	63	938.172		15.200	100

Islands, a high level of intra-population variability among the coconut accessions and occurrence of both Niu Vai and Niu Kafa types has been reported [42, 47]. The present study is the first report of coconut population structure of Minicoy Island which is distant from the other Islands of the Lakshadweep group and the coconut populations of this island is considered as less influenced with human selection as evidenced through wide variability for all the observed traits and presence of unique types such as LMMT. It was postulated that the Andaman and Nicobar Islands in Bay of Bengal could be a part of centre of diversity of coconut based on the observation of wild diverse coconut types from these islands [2]. Minicoy Island had been on trade and cultural relations for centuries with the Nicobar Islands and this would have facilitated movement of coconut to these islands and lead to further introgression with local types. The Minicoy Island has served as a halting point for many seafarers from Southeast Asia to Africa before the opening of Suez Canal [48]. All these might have contributed towards the migration of coconut populations from Nicobar Islands and other SE Asian Islands to Minicoy and then to other places in Lakshadweep and from there to mainland India. Hence, the Minicoy coconut population could be basically a combination of local types and introgressed types with Nicobar coconut types and their segregating progenies. The LMMT palms could be a transgressive segregant for fruit component traits, which would have occurred due to less interference of human selection. The LMMT type, if propagated true to type, could be useful for ornamental purpose as avenue planting as the falling nuts would not be a problem for visitors. Although some islanders consider the water from LMMT for medicinal uses, there is no scientific evidence for the same. However, the very high oil content in the LMMT make this type important in breeding varieties for higher oil content. The inheritance of this small nut trait needs further studies to be used in breeding programmes as most of these traits are quantitative and governed by polygenes.

The LMT, in general, can be assumed to possess the inherent variability existing in LCT with the distinction of producing smaller-sized nuts of varying size and bearing habit [12]. The importance of selection among the LMT types was highlighted for successful utilization of this type for desirable traits despite its irregular bearing nature and variation in nut size over seasons as the conserved LMT in the gene bank was reported to be suitable for ball copra production [7], which is a premium type of copra fetching more price in the market. The low germination rate of LMT nuts under storage makes them suitable for ball copra production. In larger nuts, owing to the larger cavity and high nut water, the germination and subsequent spoilage of kernel during storage was high, whereas in LMT, the germination and spoilage was found to be less [7]. Individual palms of LMT from Minicoy were reported to be yielding more than 700 fruits per annum with estimated annual copra out turn of over 50 kg per palm [9]. Hence, variable LMT types in Minicoy Island have potential to be utilized in developing not only superior varieties for ball copra production, but also development of superior genetic stock with more copra output.

According to a theory proposed on origin, domestication and dissemination of coconuts around the world [19], coconuts disseminated by floating between offshore islands. Introgressive hybridization between the domesticated type (large, spherical coconuts having desirable attributes for human consumption and cultivation) and the wild type (long angular coconuts, evolved by natural selection for large distance dissemination by sea) gave rise to the present diversity of coconuts around the world. The wild-type coconuts evolved as long angular fruit by natural selection, capable of long distance dissemination by sea, in both Indian and Pacific oceans. It was reported that domestic type coconuts, selected by early cultivators in Southeast Asian countries for large nut volume as a refreshing drinking water source, were subsequently carried during long voyages by ancestors of the Polynesians, who reached Madagascar in the west and Samoa in the east. Minicoy Island is situated in the path of this route and archaeological evidences point to the transport of goods through this Island [48]. Hence, the occurrence of large

fruit size in Minicoy (LCGT) may be due to the introgression of large fruited types from SE Asian countries or Nicobar Islands with the local types.

The fruit component and molecular analysis of coconut populations around the world distinguished two main groups of coconuts, viz. the Southeast Asian and South Pacific coconut comprising the Dwarfs and the Indian Ocean types. While addressing the questions about the evolutionary and genetic advantages of occurrence of small-sized fruits in the Lakshadweep Islands, contribution of possible extreme forms of pollination was suggested towards the production of Micro Tall types [12]. Micro nut types are also reported from Andaman and Nicobar Islands and have been collected and conserved from those islands [25]. Similarly, Micro forms of coconut types have been reported from southern part of Indian mainland and popularly called in vernacular name as '*Ayiramkachi*' (meaning 1000 nuts bearer) Tall [43]. Hence, the LMT types can be categorized as the type developed by the introgressive hybridization with other coconut types and the LMMT is another segregant from the population.

Microsatellite analysis using a set of 19 polymorphic SSR markers revealed higher allelic diversities, which is in accord with previous studies of tall coconut populations from islands, viz. Sri Lanka [37], Andaman and Nicobar Islands [41], Dominican Republic [32], Hainan [52] and studies on Indian coconut accessions [11]. Fourteen of the loci showed significant departures from HW equilibrium which may be due to heterozygote deficit. The observed heterozygosity for all the accessions was less than expected indicating a tendency towards inbreeding within the population, which could be due to the result of a positive assortative mating between the individuals, an artificial sub-grouping of individuals from populations or selection which favours homozygotes.

An important part of population genetics is the determination of genetic structure of natural populations and this estimation has varied applications in evolutionary biology [33]. For devising effective conservation management strategies, it is necessary to assess genetic variation present and partition it within and between populations. F_{ST}, G_{ST} and D_{est} estimates all indicated only a low level of genetic differentiation among coconut populations of Minicoy Island, well below those expected for outcrossing species [18]. Mean F_{IS} (0.369) and F_{IT} (0.424) were both positive and greater than zero indicating a heterozygote deficit within populations. The mean gene flow (N_m), based on mean F_{ST}, was very high (3.841) indicating an extensive gene flow among the four coconut accessions. Estimates of genetic differentiation based on heterozygosity (F_{ST}) can underestimate levels of differentiation for markers such as SSRs that have high allelic diversity [22, 27]. Our estimates of differentiation for the SSR data based on Jost's D_{est} and G'_{ST} and G''_{ST} are substantially lower than those based on F_{ST}, suggesting these corrected estimates reveal the true levels of differentiation than those based on heterozygosity (F_{ST}).

The results obtained in the present study are an indication that genetic drift in coconut populations, in addition to outcrossing behaviour, might play a major role in the determination of the amount of genetic variation between populations and also genetic differentiation among populations. From SSR analysis, a higher level of genetic variation was observed among individual palms within populations, compared to variation among populations in the Minicoy Island coconut populations, suggesting the existence of a high genetic overlap as a result of gene flow through pollen. In spite of the use of a large number of polymorphic markers, analyses conducted using the Bayesian clustering method [39] similarly produced little evidence of clear or pronounced population structure. It might be possible that because the average levels of differentiation estimates was low ($F_{ST} = 0.091$), STRUCTURE could not detect evidence of true genotypic clustering. UPGMA-based clustering revealed LMMT to be a distinct population, whereas LCGT, LCT and LMT clustered together.

The studies highlight the diverse nature of Minicoy Island coconut populations and the need for further explorations for ex situ conservation and utilization. High degree of coconut diversity losses are reported worldwide due to the lethal and debilitating diseases such as lethal yellowing of pacific region, root (wilt) of India, Weligama leaf wilt of Sri Lanka, several pests and climate change adversity which may lead to deterioration of livelihood among different coconut communities. Recent studies using worldwide samples of coconuts for genetic diversity and population structure indicated the possible independent origins of coconut cultivation from Southeast Asia and southern margin of Indian subcontinent [3]. It is suggested that genetic structure may be useful in targeting source populations for disease resistance and other crop improvement traits. In this background, further efforts are required for conservation of natural coconut populations available in remote places such as the one in Minicoy Island as they may offer the required resistance gene sources for breeding programmes by way of natural recombinants. The coconut palms on islands within atoll ecosystem are considered to be members of one open pollinated population [21]. Considering the natural selection pressure for survival on these populations, it is generally expected that coconut population has to be less diverse. However, significant variation in fruit shape, size and morphological features of palms was also reported from Amini and Kadmat Islands of Lakshadweep [42].

Hence, we propose in situ conservation efforts on these populations through which observations on continuous

natural evolution in coconut are possible as the progenies are established with natural selection rather than with human intervention for selection. Such efforts will aid in identifying the adaptive traits in coconut for establishment under natural conditions. The geographical ecology of palms was highlighted as determinants of diversity and distribution [15], whereas such detailed studies or information on coconut palm is limited. Therefore, studies on the adaptive traits and differentiation of coconut types under Island ecosystems such as Minicoy will be very much helpful in initiating research on mitigation of climate change effects in coconut farming considering the fact that Lakshadweep coconut types were reported to possess traits contributing to drought tolerance [40]. The Lakshadweep coconuts have also been reported to be unique for their nectar production and compositions [38] when compared to other coconut types which need further studies for exploitation. In the past, three improved varieties, viz. Chandrakalpa, Chandra Laksha, Laksha Ganga, have been developed and released for commercial cultivation in different parts of India using Lakshadweep Ordinary Tall selection [12] as parent. Hence, careful selection of individual palms among LMT, LCGT and LMMT groups through morphological and molecular markers would help in development of superior genetic stock with desirable traits for use in improvement programmes. In the light of the studies, it is imperative that further investigations on remote island coconut populations would yield useful information on rich diversity of coconut genetic resources and offer scope for expanding genetic base of coconut for the benefit of coconut-breeders.

Acknowledgements The authors acknowledge their gratitude to ICAR for the financial support.

Author Contributions BAJ carried out field exploration and in situ morphological characterization. MKR carried out microsatellite analysis. RJT carried out collection of identified samples for molecular analysis. VN was involved in documentation of identified types. KS carried out the statistical analysis of morphological traits. All the authors were involved in preparation of manuscript.

Compliance with Ethical Standards

Conflict of interest The authors declare that they have no conflict of interest.

References

1. Ashburner GR, Thompson WK, Halloran GM, Foale MA (1997) Fruit component analysis of South Pacific coconut palm populations. Genet Resour Crop Evol 44:327–335
2. Balakrishnan NP, Nair VJ (1976) Wild populations of *Areca* and *Cocos* in Andaman and Nicobar Islands. Indian J For 2:350–363
3. Bee FG, Baudouin L, Olsen KM (2011) Independent origins of cultivated coconut (*Cocos nucifera* L.) in the old world tropics. PLoS ONE 6:e21143
4. Bhaskara Rao EVV, Pillai RV (1982) Characterization of coconut germplasm based on fruit component analysis. In: Bavappa KVA, Iyer RD, Prafulla KD, Sethuraj MR, Ranganathan V, Nambiar KKN, Chacko MJ (eds) Proceedings of PLACROSYM V, CPCRI. Kasaragod, 15–18 Dec 1982, pp 112–124
5. CDB (2013) All India Final Estimates of area and production of Coconut. Coconut Development Board 2012–2013, India. http://coconutboard.nic.in/stat.htm
6. CPCRI (2002) Annual Report 2001–02. Central Plantation Crops Research Institute, Kasaragod, 671124, Kerala, India, p 120
7. CPCRI (2008) Annual Report 2007–08. Central Plantation Crops Research Institute, Kasaragod, 671124, Kerala, India, p 102
8. CPCRI (2009) Annual Report 2008–09. Central Plantation Crops Research Institute, Kasaragod, 671124, Kerala, India, p 145
9. CPCRI (2012) Annual Report 2011–12. Central Plantation Crops Research Institute, Kasaragod, 671124, Kerala, India, p 128
10. Das AK (2002) Islands. In: Alfred JRB, Das AK, Sanyal AK (eds) Ecosystems of India. ENVIS Zoo Surv India, Kolkata, pp 1–410
11. Devakumar K, Jayadev K, Rajesh MK, Chandrasekar A, Manimekalai R, Kumaran PM, Parthasarathy VA (2005) Assessment of the genetic diversity of Indian coconut accessions and their relationship to other cultivars, using microsatellite markers. Plant Genet Resour Newsl 145:38–45
12. Devakumar K, Niral V, Jerard BA, Jayabose C, Chandramohanan R, Jacob PM (2010) Microsatellite analysis of distinct coconut accessions from Agatti and Kavaratti Islands, Lakshadweep, India. Sci Hortic 125:309–315
13. Evanno G, Regnaut S, Goudet J (2005) Detecting the number of clusters of individuals using the software STRUCTURE: a simulation study. Mol Ecol 14:2611–2620
14. Excoffier L, Smouse P, Quattro J (1992) Analysis of molecular variance inferred from metric distances among DNA haplotypes: application to human mitochondrial DNA restriction data. Genetics 131:479–491
15. Eiserhardt WL, Svenning JC, Kissling WD, Balslev H (2011) Geographical ecology of the palms (Arecaceae): determinants of diversity and distributions across spatial scales. Ann Bot 108:1391–1416
16. Falush D, Stephens M, Pritchard JK (2003) Inference of population structure using multilocus genotype data: linked loci and correlated allele frequencies. Genetics 164:1567–1587
17. Guo S, Thompson EA (1992) Performing the exact test of Hardy-Weinberg proportion for multiple alleles. Biometrics 48:361–372
18. Hamrick JL, Godt MJW (1989) Allozyme diversity in plant species. In: Brown AHD, Clegg MT, Kahler AL, Weir BS (eds) Plant population genetics, breeding and germplasm resources. Sinauer, Sunderland, pp 43–63
19. Harries HC (1978) The evolution, dissemination and classification of *Cocos nucifera*. Bot Rev 44:265–320
20. Harries HC (1982) Coconut varieties. Indian Cocon J 12:3–10
21. Harries HC, Clement RC (2014) Long-distance dispersal of the coconut palm by migration within the coral atoll ecosystem. Ann Bot 113:565–570
22. Hedrick PW (2005) A standardized genetic differentiation measure. Evolution 59:1633–1638
23. IPGRI (1995) Descriptors for Coconut (*Cocos nucifera* L.) International Plant Genetic Resources Institute, Rome, Italy
24. Jacob PM, Krishnamoorthy B (1981) Observations on the coconut gene pool in Lakshadweep islands. In: Vishveshwara S, Sreenivasan MS, Krishnamurthy RW, Srinivasan CS, Purushotham K, Iyer RD, Nair MK, Sharma VS, Panickar AON (eds)

Proceedings-PLACROSYM IV, CCRI, Mysore, 3–5 Dec, 1981, pp 3–8
25. Jerard BA, Dhanapal R, Arunachalam V (2000) Biometric clustering of field-collected coconut germplasm of Andaman and Nicobar Islands, India. In: Engels JMM, Ramanatha Rao V, Brown AHD, Jacksn MT (eds) Proceedings of the international conference on science and technology for managing plant diversity in the 21st century, Kuala Lumpur, Malaysia, Abstract, p 77
26. Jerard BA, Damodaran V, Niral V, Samsudeen K, Rajesh MK, Sankaran M (2013) Conservation and utilization of soft endosperm coconut accession from Andaman islands. J Plant Crops 41:14–21
27. Jost L (2008) G_{st} and its relatives do not measure differentiation. Mol Ecol 17:4015–4026
28. Krishnamoorthy B, Jacob PM (1982) Fruit component analysis in Lakshadweep coconuts. In: Bavappa KVA, Iyer RD, Prafulla KD, Sethuraj MR, Ranganathan V, Nambiar KKN, Chacko MJ (eds) Proceedings of PLACROSYM V, CPCRI. Kasaragod, 15–18 Dec, 1982, pp 180–183
29. Kumaran PM, Koshy PK, Arunachalam V, Niral V, Parthasarathy VA (2000) Biometric clustering of coconut populations of Indian Ocean Islands. In: Muraleedharan N, Rajkumar R (eds) Recent advances in plantation crops. Allied Publishers, New Delhi, pp 73–81
30. Leach BJ, Foale MA, Ashburner GR (2003) Some characteristics of wild and managed coconut palm populations and their environment in the Cocos (Keeling) Islands, Indian Ocean. Genet Resour Crop Evol 50:627–638
31. Lewis PO, Zaykin D (2002) Genetic data analysis: computer program for the analysis of allelic data. Version 1.1. Free program distributed by the authors over the internet from http://hydrodictyon.eeb.uconn.edu/people/plewis/software.php
32. Martinez RT, Baudouin L, Berger A, Dollet M (2009) Characterization of the genetic diversity of the Tall coconut (*Cocos nucifera* L.) in the Dominican Republic using microsatellite (SSR) markers. Tree Genet Genomes 6:73–81
33. Meirmans PG, Hedrick PW (2011) Assessing population structure: F_{ST} and related measures. Mol Ecol Resour 11:5–18
34. Nei M (1978) Estimation of average heterozygosity and genetic distance from a small number of individuals. Genetics 89:583–590
35. Ovasuru T, Tan GY, Bridgland LA (1993) Coconut germplasm collection in Papua New Guinea. In: Nair MK, Khan HH, Gopalasundaram P, Rao EVVB (eds) Advances in coconut research and development. Oxford and IBH Publishing Co. Pvt. Ltd., New Delhi, India, pp 33–42
36. Peakall R, Smouse PE (2012) GenAlEx 6.5: genetic analysis in Excel. Population genetic software for teaching and research-an update. Bioinformatics 28:2537–2539
37. Perera L, Russell JR, Provan J, Powell W (2000) Use of microsatellite DNA markers to investigate the level of genetic diversity and population genetic structure of coconut (*Cocos nucifera* L.). Genome 43:15–21
38. Prasannakumari TO (1963) Sugar composition of nectar in coconut. Curr Sci 32:460–461
39. Pritchard J, Stephens M, Donnelly P (2000) Inference of population structure using multi-locus genotype data. Genetics 155:945–959
40. Rajagopal V, Shivashankar S, Kasturibai KV, Voletti SR (1988) Leaf water potential as an index of drought tolerance in coconut (*Cocos nucifera* L.). Plant Physiol Biochem 15:80–86
41. Rajesh MK, Nagarajan P, Jerard BA, Arunachalam V, Dhanapal R (2008) Microsatellite variability of coconut (*Cocos nucifera* L.) accessions from Andaman and Nicobar islands. Curr Sci 94:1627–1631
42. Rajesh MK, Samsudeen K, Jerard BA, Rejusha P, Karun Anitha (2014) Genetic and phylogenetic relationships of coconut populations from Amini and Kadamat islands, Lakshadweep (India). Emir J Food Agric 26:898–906
43. Ramachandran M, Muralidharan VN, Balasubramanian K (1977) A note on the new coconut variety-Ayiramkachi. Indian Cocon J 8:4–6
44. Ratnambal MJ, Nair MK, Muralidharan K, Kumaran PM, Rao EVVB, Pillai RV (1995) Coconut Descriptors Part-I. CPCRI, Kasaragod, pp 168–175
45. Rice WR (1989) Analyzing tables of statistical tests. Evolution 43:223–225
46. Samsudeen K, Jacob PM, Rajesh MK, Jerard BA, Kumaran PM (2006) Origin and evolution of Laccadive Micro Tall, a coconut cultivar from Lakshadweep islands of India. J Plant Crops 34:220–225
47. Samsudeen K, Jacob PM, Niral V, Kumaran PM, Salooja R, Moosa H (2006) Exploration and collection of coconut germplasm in Kadmat and Amini Islands of Lakshadweep, India. Genet Resour Crop Evol 53:1721–1728
48. Tripati S (2009) Stone anchors from Minicoy Island, Lakshadweep, India. Int J Naut Archaeol 38:406–412
49. Vadivelu S, Bandyopadhyay AK (1997) Characteristics, genesis and classification of soils of Minicoy Island, Lakshadweep. J Ind Soc Soil Sci 45:797–801
50. Vadivelu S, Muralidharan, Bandyopadhyay AK (1993) Soils of Lakshadweep Islands. Research Bulletin No. 9. Central Agricultural Research Institute, Port Blair, Andamans
51. Wright S (1951) The genetical structure of populations. Ann Eugen 15:323–354
52. Xiaolei L, Hua T, Dongdong L, Liheng H (2011) Genetic diversity of coconut cultivars in China by microsatellite (SSR) markers. Mol Plant Breed 2:83–91
53. Zizumbo-Villarreal D, Piñero D (1998) Pattern of morphological variation and diversity of *Cocos nucifera* (Arecaceae) in Mexico. Am J Bot 85:855–865
54. Zizumbo-Villarreal D, Colunga-García Marín P (2001) Morpho-physiological variation and phenotypic plasticity in Mexican populations of coconut (*Cocos nucifera* L.). Genet Resour Crop Evol 48:547–554
55. Zizumbo-Villarreal D, Fernández-Barrera M, Torres-Hernández N, Colunga-García Marín Patricia (2005) Morphological variation of fruit in Mexican populations of *Cocos nucifera* L. (Arecaceae) under in situ and ex situ Conditions. Genet Resour Crop Evol 52:421–434