



Genetic diversity and population structure of the upriver orange mangrove *Bruguiera sexangula* along the coastlines of Thailand

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ABSTRACT: Upriver orange mangrove *Bruguiera sexangula* is a member of the Rhizophoraceae family and is usually distributed in the intertidal zones of the tropical and subtropical coastal areas. The analyses of genetic diversity and population structure of *B. sexangula* are essential for their conservation and management. In the present study, the genetic diversity and structure of 101 *B. sexangula* individuals from mangrove forests in Thailand were evaluated using single nucleotide polymorphisms (SNPs) through restriction site-associated DNA sequencing (RAD-seq). A total of 3482 high-quality SNPs were obtained after filtration for genetic study. All 3 genetic structure analyses (Bayesian analysis, principal components analysis, and maximum likelihood tree) revealed that these individuals can be clustered into 2 groups corresponding to geographical regions, namely the Andaman Sea and Gulf of Thailand. The level of genetic differentiation between the clusters was moderate ($F_{ST} = 0.122$, $p < 0.001$), and the analysis of molecular variance (AMOVA) indicated that the individual differences within groups were greater than the differences between the 2 groups. A moderate level of genetic diversity (Shannon's information index, $I = 0.458$; observed heterozygosity, $H_o = 0.416$; expected heterozygosity, $H_e = 0.295$) of *B. sexangula* was observed. These results give us a better understanding of the genetic variations and will provide a framework for the conservation of *B. sexangula*.

KEY WORDS: Genetic diversity · Population structure · Mangrove · *Bruguiera sexangula* · RAD-seq · Single nucleotide polymorphisms · SNPs

1. INTRODUCTION

Mangrove forests are widely distributed in the intertidal zones of the tropical and subtropical coastal areas. They are found in over 123 countries and comprise around 73 species (Spalding 2010, Giri et al. 2011). Mangrove forests are one of the most ecologically valuable ecosystems in the world (Costanza et al. 2014) and serve as nurseries for several marine and terrestrial species that support coastal livelihoods (Nagelkerken et al. 2008, Alongi 2012). Man-

groves also provide several critical services such as fisheries support, water quality maintenance, flood protection, mitigating coastal erosion, and storm protection (Walters et al. 2008, Zhang et al. 2012, Marois & Mitsch 2015, Carrasquilla-Henao & Juanes 2017, Menéndez et al. 2020). Despite their importance, mangrove forests continue to decline globally due to both natural and anthropogenic causes, and these ecosystems have become more fragmented (Binks et al. 2019, Bryan-Brown et al. 2020). The estimated global area of mangrove forests was 152 604 km² in

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1996 and 147 359 km² in 2020 and—although there was an overall decline in mangrove cover—on the positive side, deforestation rates in this 24 yr period had also decreased (Bunting et al. 2022). In Thailand, mangroves are found on the coastlines of both the Gulf of Thailand and Andaman Sea and cover approximately 1761 km² of the Andaman coastline and 693 km² of the Gulf of Thailand (Lange et al. 2019). Mangrove forests in the country have suffered destruction by conversion to agriculture, aquaculture, industrial expansion, and urban area extension (Pumijumnong 2014). They decreased drastically from 3679 km² in 1961 to 2296 km² in 2007 (Aksomkoe 1993, Pumijumnong 2014).

Upriver orange mangrove *Bruguiera sexangula* is one of the important species of mangrove forests in Thailand. This species belongs to Rhizophoraceae, a true mangrove family, and is mostly distributed in the Indo-West Pacific region (Duke & Ge 2011). Upriver orange mangrove is an economically important mangrove species. The wood of this species is utilized for fuelwood, charcoal production, and house construction, and the bark is used as a source of tannin. Moreover, the fruit and the roots of *B. sexangula* can be used for medicinal purposes (Hanum & Van der Maesen 1997). However, the genetic diversity and population structure of upriver orange mangroves in Thailand have not previously been examined. Understanding genetic variations within and between populations is crucial to conservation management (Toro & Caballero 2005), as these parameters significantly influence fitness and population viability (Frankham 2010). The evaluation of genetic parameters in mangrove species helps to identify populations that are at risk of extinction, prioritize them for conservation efforts, and guide management interventions (Wee et al. 2019). For example, populations exhibiting high levels of genetic variation are of particular conservation importance due to their enhanced ability to adapt to alterations in the environment. Conversely, populations that exhibit low diversity or show signs of inbreeding may require genetic management interventions, such as the promotion of enhanced gene flow (Frankham et al. 2019). Overall, genetic analyses can provide information that helps ensure that conservation efforts are targeted and effective, thereby preventing the extinction of mangrove species.

Molecular markers provide an accurate and effective tool for estimating the genetic diversity and population structure of a plant species (Porth & El-Kassaby 2014). Several studies have been conducted to analyze the genetic diversity and structure of man-

grove species using different types of molecular markers: Simple Sequence Repeat (SSR) for *Rhizophora apiculata* (Azman et al. 2020), *Sonneratia alba* (Wee et al. 2017) and *Rhizophora stylosa* (Islam et al. 2014); Inter-Simple Sequence Repeat (ISSR) for *Nypa fruticans* (Jian et al. 2010) and *Rhizophora mangle* (Chablé Iuit et al. 2020); Random Amplified Polymorphic DNA (RAPD) for *Bruguiera gymnorhiza* and *Heritiera fomes* (Dasgupta et al. 2015); nuclear gene for *B. gymnorhiza* (Minobe et al. 2010) and *Rhizophora* species (Chen et al. 2015); and chloroplast DNA for *Ceriops* species (Huang et al. 2008), *Excoecaria agallocha* (Guo et al. 2018), and *R. stylosa* (Islam et al. 2014). Among different types of markers, single nucleotide polymorphisms (SNPs) have proved to be the most abundant type of molecular marker, and their high density provides a better insight into the genetic basis of a population (Howe et al. 2013). The restriction site-associated DNA sequencing (RAD-seq) is one of the reduced-representation library sequencing techniques, facilitating the rapid discovery of a large set of genome-wide SNP markers across many individuals (Davey & Blaxter 2010, Davey et al. 2011). This approach combines restriction enzyme digestion of the genome with high-throughput sequencing and represents a cost-effective and powerful genotyping method that is applicable to both model organisms and non-model species with no existing genomic resources (Miller et al. 2007, Davey et al. 2011). Consequently, RAD-seq has increasingly been applied to identify and genotype genome-wide SNP markers in several plant species, including mangrove species, to study genetic diversity (Gao et al. 2017, Tsujimoto et al. 2019, Cai et al. 2020, Hsu et al. 2022, Khanbo et al. 2022, Nagano et al. 2022, Ruangareerate et al. 2022, Naktang et al. 2023).

In this study, we characterized 101 *B. sexangula* accessions from a number of mangrove forests in Thailand using SNP markers obtained by the RAD-seq approach. We aimed to reveal the genetic diversity and structure of *B. sexangula* populations, understand the level of genetic variation, and provide useful genetic information to support mangrove forest conservation.

2. MATERIALS AND METHODS

2.1. Plant materials and DNA extraction

Leaf samples were collected from *Bruguiera sexangula* individuals in 8 provinces of Thailand along the coasts of the Gulf of Thailand (Chumphon: CMP;

Nakhon Si Thammarat: NST; Surat Thani: SNI; Trat: TRT; Chanthaburi: CTI) and the Andaman Sea (Satun: STN; Trang: TRG; Ranong: RNG) between 2020 and 2021 (Fig. 1). We collected a total of 101 individuals, comprising 7 individuals from CMP, 11 individuals from CTI, 9 individuals from NST, 19 individuals from RNG, 17 individuals from SNI, 5 individuals from TRG, and 18 individuals from TRT (Table 1). Different numbers of samples were collected at each of the 8 sites since the population sizes and distribution characteristics varied among the 8 sites. The individuals were selected at a distance of at least 20 m from each other to avoid collecting closely related individuals and to maximize the likelihood of collecting diverse genotypes (Ngeve et al. 2017, Triest et al. 2020, Canty et al. 2022). Total genomic DNA was isolated from fresh young leaves using the cetyltrimethyl ammonium bromide (CTAB) method followed by a cleanup using a DNeasy Plant Mini Kit (Qiagen). The concentration of the isolated genomic DNA was quantified using the Qubit fluorometer (Thermo Fisher Scientific) and a Qubit dsDNA BR Assay kit (Invitrogen).

2.2. RAD library construction and SNP calling

The libraries for RAD-seq were constructed using the MGIEasy RAD library preparation kit (MGI Tech) following the manufacturer's protocols. Briefly, genomic DNA was digested with the *TaqI* restriction enzyme and DNA fragments were ligated with uniquely bar-coded adapter pairs. Following polymerase chain reaction (PCR) and quantification, the samples were pooled in an equimolar manner. Pair-end sequencing with a read length of 150 bp was conducted on the MGISEQ-2000RS according to the manufacturer's instructions (MGI Tech).

The RAD-seq data were processed using the Genome Analysis Toolkit (GATK) (McKenna et al. 2010)

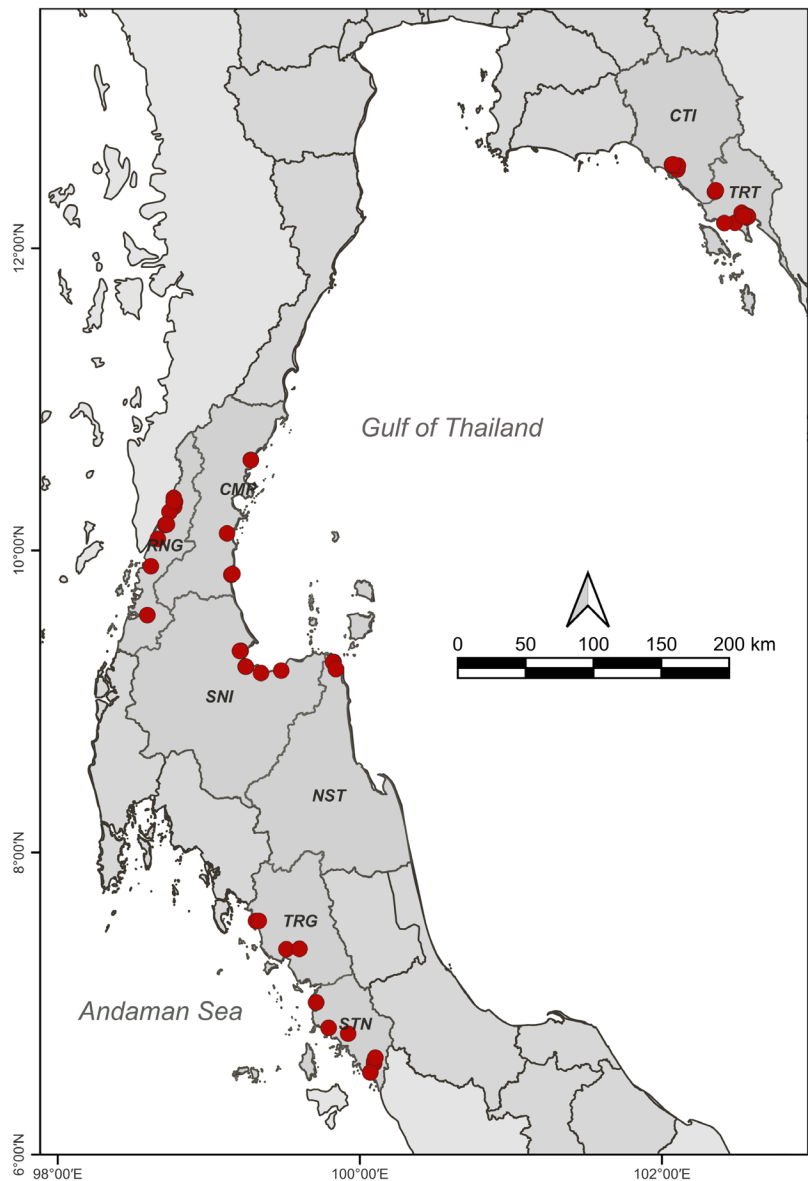


Fig. 1. The geographical location of 101 *Bruguiera sexangula* accessions in Thailand. Accessions were collected from mangrove forests on the Gulf of Thailand coast (Chumphon: CMP; Chanthaburi: CTI; Nakhon Si Thammarat: NST; Surat Thani: SNI; Trat: TRT) and the Andaman Sea coast (Trang: TRG; Ranong: RNG; Satun: STN), respectively. Red dots represent sites at which the accessions were collected

pipeline. The sequences of each sample were sorted depending on the barcodes. Sequenced reads were aligned with the reference genome of *B. sexangula* (GenBank accession number JAHG000000000) using Burrows-Wheeler Alignment mapping algorithm (Li & Durbin 2009). For SNPs calling, GATK HaplotypeCaller (McKenna et al. 2010) was utilized. The SNP calls from the GATK were filtered using the following criteria: (1) depth coverage between 10X–

Table 1. Information of population samples used in this study

Coast region	Province (abbreviation)	Date of collection	Sample size
Gulf of Thailand	Chumphon (CMP)	3 August 2021–4 October 2021	7
Gulf of Thailand	Chanthaburi (CTI)	1 March 2021–17 May 2021	11
Gulf of Thailand	Nakhon Si Thammarat (NST)	17 May 2021–11 October 2021	9
Andaman Sea	Ranong (RNG)	26 October 2020–14 February 2021	19
Gulf of Thailand	Surat Thani (SNI)	26 October 2020–28 February 2021	17
Andaman Sea	Satun (STN)	12 October 2020–25 January 2021	15
Andaman Sea	Trang (TRG)	17 July 2021–31 July 2021	5
Gulf of Thailand	Trat (TRT)	24 November 2020–1 February 2021	18

200X; (2) fewer than 5% missing data; and (3) a minor allele frequency > 0.05.

We obtained a total of 1 518 523 970 raw reads, ranging from 1 062 916 (TRT_16) to 7 241 794 (SNI_09), with an average of 15 034 890 reads per sample (see Table S1 in the Supplement at www.int-res.com/articles/suppl/b032p031_supp.xlsx). An average of 13 205 970 reads (86.36%) were successfully mapped onto a reference genome (Pootakham et al. 2022b), with the highest (94.59%) mapping rate for SNI_04. A total of 2 823 114 SNP loci were initially identified by the GATK pipeline. After filtering, 3482 high-quality SNP loci were used for downstream analyses and we found that the polymorphism information content (PIC) value of SNP markers ranged from 0.09 to 0.50, with an average PIC value of 0.241. The minor allele frequency distribution ranged from 0.05 to 0.50 with an average of 0.218. The PIC values are a good indication of informative markers which can be utilized for studying genetic diversity (Soumya et al. 2021). Similar results were also found in other studies (Pootakham et al. 2022c, Ruang-areerate et al. 2022), supporting the fact that our PIC values are acceptable in mangrove species and suitable for further analysis.

2.3. Population structure and genetic diversity assessment

To infer the genetic structure of *B. sexangula* populations, we applied 3 methods. First, a Bayesian approach implemented in the program STRUCTURE v.2.3.4 (Pritchard et al. 2000) was used to determine population structure. We performed 20 replicates for each K value ($K = 1–10$), with a burn-in period of 10 000 and a run length of 10 000 iterations. The optimal K value was calculated using the ΔK method (Evanno et al. 2005) in the web-based STRUCTURE HARVESTER software (Earl & vonHoldt 2012). The

average cluster membership proportions for the 10 000 replicates of a given K value were estimated using CLUMPP v.1.1.2 (Jakobsson & Rosenberg 2007). The analysis of molecular variance (AMOVA) was performed on the groups obtained by STRUCTURE, using ARLEQUIN v.3.5 (Excoffier et al. 2005) with 100 000 permutations. Population differentiation (F_{ST}) was also estimated using Arlequin. Second, a principal components analysis (PCA) was performed to explore group conformation within the population using TASSEL v.5.2 (Bradbury et al. 2007), and data were plotted based on the first 3 principal components. Third, the phylogenetic tree was constructed based on a maximum likelihood (ML) method under the 1000 bootstrap replicates with MEGA X (Tamura et al. 2021) to further assess the relationship between the accessions.

Genetic diversity was assessed across groups defined by STRUCTURE using GenAlEx v.6.502 (Peakall & Smouse 2012) to estimate the number of effective alleles (N_e), Shannon's information index (I), observed heterozygosity (H_o), expected heterozygosity (H_e), and the inbreeding coefficient (F_{IS}). Gene flow (Nm) across 2 populations was estimated using the following formula: $Nm = [(1/F_{ST}) - 1]/4$. The PIC values for SNP markers were calculated using PowerMarker v.3.25 (Liu & Muse 2005).

3. RESULTS

3.1. Genetic structure of *Bruguiera sexangula*

SNPs generated from RAD-seq were utilized to infer the genetic structure of the *B. sexangula* population. Genetic structure was evaluated using STRUCTURE, principal components analysis (PCA), and ML tree. For the STRUCTURE analysis results based on 3482 SNPs, the distribution of ΔK revealed that the optimal K value was $K = 2$ (Fig. 2a).

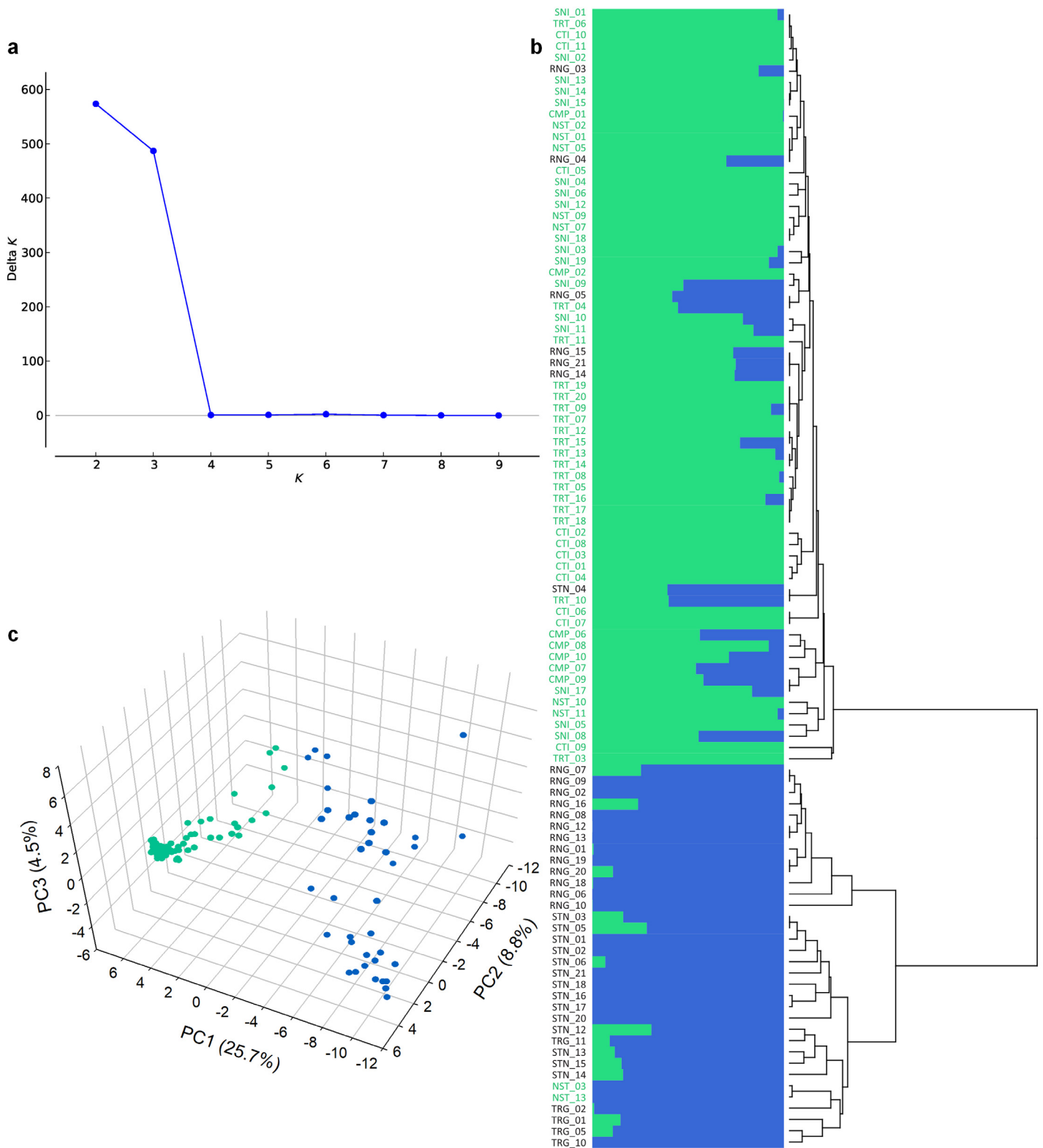


Fig. 2. Population structure and phylogeny of *B. sexangula*. (a) Number of subpopulations indicated by the highest ΔK ; (b) population structure of *B. sexangula* accessions estimated by STRUCTURE and the maximum likelihood (ML) phylogenetic tree. (c) principal components analysis (PCA) plots of the first 3 components of *B. sexangula* accessions. Accessions in green and blue were collected from mangrove forests on the Gulf of Thailand and the Andaman coasts, respectively

When $K = 2$, most of the accessions from the Gulf of Thailand coast were grouped together into a first cluster, and a second cluster comprising mainly of accessions from the Andaman Sea coast (Fig. 2b). The first cluster, the largest group with 58 accessions, was collected from Chumphon (CMP), Chanthaburi (CTI), Nakhon Si Thammarat (NST), Surat Thani (SNI), Trat (TRT), and Ranong (RNG); the second cluster consisted of 36 accessions collected from RNG, Satun (STN), Trang (TRG), NST, and TRT. Most of the accessions from the same geographic region fell under the same cluster classification. However, some accessions were genetically mixed with a membership probability (q-values) of less than 0.6 in 2 clusters. Seven of the accessions were considered to be admixtures, and they originated from CMP, TRT, SNI, and RNG. The classification of $K = 2$ reflected the separation of accessions corresponding to their geographical regions and was supported by the results of a PCA. Based on 3482 SNP markers, the PCA also revealed 2 distinct groups of *B. sexangula* accessions and 3 principal components, accounting for 39% of the total variation observed (Fig. 2c). Additionally, the ML tree showed that the 101 *B. sexangula* accessions were clustered into 2 clades (Fig. 2b). Clade 1 included 67 accessions, mainly from the Gulf of Thailand, of which 7 were from the Andaman Sea coast. Clade 2 contained 34 accessions, including 32 from the Andaman Sea coast and 2 from the Gulf of Thailand coast. These results were consistent with the STRUCTURE analysis.

Based on the 2 genetic clusters from the STRUCTURE and AMOVA outputs, variation among clusters accounted for 12.23% of the total genetic variation while a larger amount of variation (87.77%) was found within clusters (Table 2). The measure of population differentiation (F_{ST}) among the clusters was 0.122, at $p < 0.001$, and the estimated gene flow among populations (Nm) was 1.799.

3.2. Genetic diversity of *B. sexangula*

Genetic diversity parameters were calculated for the entire population and separately for each cluster (Table 3). Mean values were $N_e = 1.491$, $I = 0.458$, $H_o = 0.416$, and $H_e = 0.295$. All diversity parameters were similar in the 2 cluster. Negative values of the inbreeding coefficient (F_{IS}) were found in both clusters (-0.295 in cluster 1 and -0.291 in cluster 2), indicating an excess of heterozygosity.

4. DISCUSSION

Mangrove areas in Thailand have declined dramatically as a result of anthropogenic disturbance, primarily through conversion to shrimp aquaculture (Pumijumnong 2014). Therefore, the evaluation of genetic diversity and population structure of mangroves represents the first step to understanding the current status of mangrove species biodiversity,

Table 2. Analysis of molecular variance (AMOVA) among and within 2 clusters of *Bruguiera sexangula* according to STRUCTURE analysis using 3482 SNPs. df: degrees of freedom; F_{ST} : genetic differentiation; Nm: gene flow

Source of variation	df	Sum of squares	Variance component	Percentage of variation	p-value
Among clusters	1	6571.36	63.84	12.23	<0.001
Within clusters	200	91616.25	458.08	87.77	
Total	201	98187.61	521.92		
F_{ST}	0.122				
Nm	1.799				

Table 3. Genetic diversity indices for the 2 clusters of *B. sexangula* based on 3482 SNPs. N : number of samples; N_e : number of effective alleles; I : Shannon's information index; H_o : observed heterozygosity; H_e : expected heterozygosity; F_{IS} : inbreeding coefficient

Population	N	N_e	I	H_o	H_e	F_{IS}
Cluster 1	62	1.485 ± 0.006	0.447 ± 0.003	0.417 ± 0.005	0.288 ± 0.003	-0.295 ± 0.006
Cluster 2	39	1.495 ± 0.006	0.456 ± 0.003	0.416 ± 0.005	0.295 ± 0.003	-0.291 ± 0.006
Total	101	1.491 ± 0.006	0.458 ± 0.003	0.416 ± 0.005	0.295 ± 0.003	-0.284 ± 0.006

which is of importance to the protection of mangrove genetic resources in Thailand. The present study sheds light on the genetic diversity of *Bruguiera sexangula*, which is an important mangrove species with ecological and economic significance. Our study is the first research on the genetic diversity of *B. sexangula*, and it contributes to the existing body of knowledge by using a novel set of SNP markers and analyzing populations from a wide geographic range.

The assessment of genetic structure revealed that the *B. sexangula* population was composed of 2 genetic populations. One population consists mainly of accessions located on the Gulf of Thailand coast and the other population includes mainly accessions on the Andaman Sea coast. The PCA results coincided with the STRUCTURE results. Moreover, the ML tree gave similar results that were clustered into 2 clades. These clustering patterns corresponded with their geographic regions, the Gulf of Thailand and Andaman Sea coasts. Our findings are consistent with the previously reported population structures of mangrove species in Thailand, including *Bruguiera parviflora* (Pootakham et al. 2022c), *Bruguiera cylindrica* (Khanbo et al. 2022), *Ceriops tagal* (Pootakham et al. 2022a), *Rhizophora apiculata* (Inomata et al. 2009, Ruang-areerate et al. 2022), *Bruguiera gymnorrhiza* (Ruang-areerate et al. 2023), and *Rhizophora mucronata* (Inomata et al. 2009). These species appear to exhibit geographical separation, specifically along the Gulf of Thailand and Andaman Sea coasts. Additionally, population structures of several species within the same regions have been extensively studied. For instance, genetic differentiation was observed between populations from the western and eastern coasts of the Malay Peninsula for *B. gymnorrhiza* (Minobe et al. 2010, Urashi et al. 2013), *C. tagal* (Ge & Sun 2001, Liao et al. 2007), *R. mucronata*, *Rhizophora stylosa* (Wee et al. 2015), *Sonneratia alba* (Yang et al. 2017), and *Avicennia marina* (Triest et al. 2021). According to Wright (1965), populations are considered to have low genetic differentiation when $F_{ST} \leq 0.05$, moderate differentiation when $0.05 < F_{ST} \leq 0.15$, and high differentiation when $F_{ST} > 0.15$. The F_{ST} value obtained for differentiation between the 2 populations of *B. sexangula* was moderate ($F_{ST} = 0.122$, $p < 0.001$), indicating that these 2 populations were genetically differentiated. In addition, our results revealed a high gene flow ($Nm = 1.799$) between the 2 populations. Consequently, the high genetic flux among populations led to their low or moderate genetic differentiation. Other mangrove studies also showed moderate to high population dif-

ferentiations, such as *R. apiculata* in Malaysia ($F_{ST} = 0.315$) (Azman et al. 2020), *C. tagal* in the Indo-Western Pacific ($F_{ST} = 0.267$) (He et al. 2019), *A. marina* along the coastline of Western Australia ($F_{ST} = 0.174$) (Binks et al. 2019), *R. mangle* along West and East Florida ($F_{ST} = 0.19$) (Kennedy et al. 2017), and *B. gymnorrhiza* in Japan ($F_{ST} = 0.089$) (Islam et al. 2012). One possible explanation for the differences in the genetic structure of *B. sexangula* is the presence of a land barrier that prevents gene flow between mangrove species occurring along the coasts of the Andaman Sea and Gulf of Thailand, leading to population differentiation between the coasts. This is consistent with the land barrier hypothesis of the Malay Peninsula that prevented gene flow between the East and West coasts (Duke et al. 2002). The Malay Peninsula has been reported as a land barrier for several mangrove species, such as *R. apiculata* (Ng et al. 2015), *B. gymnorrhiza* (Minobe et al. 2010, Urashi et al. 2013, Wee et al. 2020), *Xylocarpus granatum* (Tomizawa et al. 2017), *C. tagal* (Liao et al. 2007, Huang et al. 2008), *Avicennia alba* (Wee et al. 2020), and *S. alba* (Wee et al. 2017, Yang et al. 2017). This barrier prevented interregional seawater exchange, which blocked the movement of sea-drifted gene flow between the 2 regions. Ocean currents have also been reported to act as a barrier to propagule dispersal and play an important role in preventing gene flow (Wee et al. 2014), although the constraints may vary among species depending on the mobility and survivability of the propagules (Duke et al. 2002). Additionally, the adaptation to different environments, such as sea level and climatic changes, may also result in a different spatial genetic structure in the *B. sexangula* population.

In the STRUCTURE analysis, admixtures were found among 2 clusters in population structure. Genetic admixture of the 2 clusters in the *B. sexangula* population might occur from the genetic exchange between isolated populations. As has been observed for other mangroves (Li et al. 2016, Yang et al. 2017, Banerjee et al. 2020), the oceanic circulation pattern may allow for infrequent long-distance dispersal detouring around the Malacca Strait, which could explain genetic admixture across the land barrier (Rizal et al. 2012). In addition, anthropogenic factors, such as human-mediated movement of propagules or seedlings for mangrove reforestation, could be a possible factor that contributed towards the genetic admixture between the 2 regions.

The genetic diversity of the *B. sexangula* population from mangrove forests along coastlines in Thailand was assessed. Moderate levels of genetic diver-

sity (mean $H_o = 0.416$ and $H_e = 0.295$, Table 3) were observed in the present study. This result is comparable to other mangrove species such as *R. apiculata* in Thailand ($H_o = 0.48$, $H_e = 0.36$) (Ruang-areerate et al. 2022), *Kandelia obovata* in China ($H_e = 0.363$) (Chen et al. 2010), and *A. marina* worldwide ($H_o = 0.407$, $H_e = 0.494$) (Maguire et al. 2000). However, a low level of genetic diversity was also reported in several studies on mangrove species, such as *R. apiculata* in Malaysia ($H_o = 0.299$, $H_e = 0.352$) (Azman et al. 2020), *S. alba* in the Indo-West Pacific ($H_o = 0.271$, $H_e = 0.327$) (Wee et al. 2017), *R. mucronata* ($H_o = 0.306$, $H_e = 0.354$) and *R. stylosa* in the Indo-West Pacific ($H_o = 0.327$, $H_e = 0.321$) (Yan et al. 2016), and *Nypa fruticans* in Southeast Asia ($H_e = 0.0279$) (Jian et al. 2010). In *Bruguiera* species populations, the average H_o value was greater in the *B. sexangula* population than in *B. cylindrica* and *B. parviflora* populations (Khanbo et al. 2022, Pootakham et al. 2022c), suggesting that the *B. sexangula* population studied here may have experienced less inbreeding than the *B. cylindrica* and *B. parviflora* populations investigated. It is generally established that genetic diversity plays a pivotal role in natural populations, imparting significant ecological consequences such as the maintenance of evolutionary potential and an individual's capacity to adapt and endure environmental changes (Hughes et al. 2008). Increased genetic drift, inbreeding, and limited gene flow can greatly diminish the genetic variation within populations (Schlaepfer et al. 2018).

Information on genetic diversity and differentiation within and among populations has the potential to impact biodiversity conservation. This study provides crucial information that can enhance existing conservation strategies aimed at protecting dwindling populations and rehabilitating degraded habitats. Our analysis revealed a moderate level of genetic variation and identified 2 distinct genetic structures within *B. sexangula* populations. According to the delineated genetic structure, we recommend treating each population as an independent conservation unit. Since the majority of genetic variation occurs within populations, it is crucial to implement preferential *in situ* conservation for populations. Conservation efforts should focus on maintaining habitat integrity and promoting gene flow and preserve overall genetic diversity. Furthermore, reforestation plans should carefully consider the geographic origins of the propagules and seedlings to ensure appropriate genetic representation and minimize the potential risks associated with mixing genetically distinct populations. Incorporating these genetic findings into conservation and management strategies will aid in the preservation of mangrove ecosystems and ensure the survival of this species.

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5. CONCLUSIONS

This study is the first report on the level of genetic diversity and population structure of *Bruguiera sexangula*. A moderate level of genetic diversity of *B. sexangula* was observed. The genetic structure of *B. sexangula* population can be clustered into 2 possible genetic clusters corresponding to the geographical regions of the Andaman Sea and Gulf of Thailand, and the level of genetic differentiation between the groups was moderate. The genetic structure was explained mainly by the presence of a land barrier. Genetic variation within the population was greater than between population. The establishment of on-site protection zones for this species to reduce the impact of human activities would enable natural regeneration of its habitats. The information obtained in this study revealed the genetic status of *B. sexangula* in Thailand, which should be useful in developing management and conservation guidelines for the species in the long term.

Data availability. Raw sequence data from this study have been deposited in the NCBI Sequence Read Archive under BioProject accession PRJNA909866.

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