Differentiatiing between Rhizophora mangle, Avicinnea germinans, Laguncularia racemose and Conocarpus erectus populations from Anlo Beach in the Shama District of Ghana using RAPD

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Research Article

Keywords: RAPD, Mangroves, Conocarpus, Genetic variation, Polymorphism

Posted Date: January 18th, 2024

DOI: https://doi.org/10.21203/rs.3.rs-3869008/v1

Additional Declarations: No competing interests reported.

Abstract

Mangroves cover about cover about 3.9% of the entire 550km coastal stretch of land from Eastern to Western region of Ghana. Despite the important ecological services and socio-economic benefits mangroves provide, they are increasingly exploited for domestic activities including fuel wood and therefore require immediate protection to prevent the few remaining populations from extinction. *Rhizophora mangle* (red mangroves), *Laguncularia racemose* (white mangroves) and *Avicennia germinans* (black mangroves) are the three most common mangrove species in Ghana. They mostly grow in association with *Conocarpus erectus* and many people classify *Conocarpus* as mangrove due to some common morphological characters they share. Though it is possible to distinguish between the three common mangrove species and *Conocarpus erectus* using morphological markers, it remains difficult to identify individuals in mixed populations therefore using molecular markers could enhance understanding and identification of the species. Genetic variation in *Rhizophora mangle*, *Laguncularia racemose, Avicennia germinans* and *Conocarpus erectus* was studied using Random Amplified Polymorphic DNA (RAPD). Intra-and interspecific variations were observed using ten out of 15 primers screened. RAPD markers were also used to produce similarity matrix among individuals of a species. A higher degree of polymorphism (94%) was observed within populations of *Rhizophora mangle, Laguncularia racemose, Avicinnea germinans*, and *Conocarpus erectus* while (6%) was observed among the populations. PhiPT-value of 86.3% (0.863) indicates considerable degree of difference between the mangrove populations and *Conocarpus erectus*, however, they shared some few common genes together. Knowledge from this research will help develop conservation strategies for mangroves and *Conocarpus* in Ghana.

INTRODUCTION

Mangroves are woody vegetation types that occur in marine and brackish environments and also are mostly restricted to the tidal zone. Mangroves have both ecological and socio-economic benefits to tropical marine biotypes and they also contribute to world's biological and genetic diversity (Sandilyan and Kathiresan 2012). In Ghana, about 3.9% of the total coastal stretch of about 550km from Eastern to Western regions are covered by mangroves, however, they are limited to areas around lagoons and also around the lower delta of River Volta (Adda 2016; Fianko & Dodd 2019; Nunoo &Agyekumhene 2022). Communities with mangroves include Keta lagoon, Sakumono lagoon, Eture, Korle lagoon, Ada, Winneba, Apam, Shama, Half Assin, Axim, Amanzure lagoon and Takoradi.

Mangroves in Ghana fall into three main genera belonging to three families namely *Rhizophora* (Rhizophoraceae), *Avicennia* (Avicenniaceae) and *Laguncularia* (Combretaceae). The species represented are *Rhizophora mangle* (red mangroves), *Avicennia germinans* (black mangroves), and *Laguncularia racemose* (white mangroves) according to deGraft-Johnson et al. (2010) and Nunoo and Agyekumhene (2022). Many plants that grow in association with mangroves in Ghana include *Thespesia populnea, Acrostichum aureum, Phoenix reclinata, Canavalia rosea, Ipomoea pes-caprae, Dalbergia escastophyllum, Sesuvium portulacastrum, Hibiscus tiliaceus, Conocarpus erectus, Drepanocarpus lunatus,* *Cardiospermum grandiflorum, Sporobolus pyramidalis, Terminalia catappa* and *Paspalum vaginatum* among others, however, *Conocarpus erectus* records the most associated species (Uddin, Huang and Bin 2019). Other lower plants including algae that grow in association with mangroves in Ghana are *Acrostichum aureum*, *Phoenix reclinate* and *Paspalum vaginatum* also grow in association with mangroves (Adekanmbi and Ogundipe 2009; Olowokudejo and Ozioma 2020)

Mangroves provide habitats for many coastal organisms including birds and also help improve fish stocks. Mangroves provide employment and income for coastal communities who engage in mangrove farming (Sackey et al. 1993; Aye et al. 2019; Das et al. 2022). Mangroves serve as wind brakes and protect coastal communities against wind damage (Das and Crépin 2013; Paul et al. 2017; Onyena and Sam 2020). Fishermen in the coastal communities mend their fishing nets and also hold community meetings under shade provided by mangroves (Nunoo and Agyekumhene (2022). In some communities, mangrove shades also serve as meeting place for socializing with other fishermen where they sit to share local alcoholic beverages such as 'pito', play local games such as 'Dame' and 'Ludu', play music and dance together when they close from work and other special occasions (Nunoo and Agyekumhene 2022).

Despite these benefits, mangroves continually face threats to extinction if action is not taken to protect the few remaining populations. Mangroves are removed as fuel wood for domestic consumption and for charcoal production by coastal communities especially the fishing areas (Bandaranayake 1998), mangroves vegetation is cleared for construction of roads and buildings for dwellings, for salt mining, agriculture and aquaculture thereby destroying most of the vegetation (Sackey et al. 1993; Seto and Fragkias 2007; Aldon et al. 2008; Bao et al. 2013; Hamilton 2013; Liingilie et. al. 2015). Domestic animals including goats and sheep feed on seedlings of mangroves during dry season when fresh leaves are not common, some vegetation of mangroves are also lost through wildfires which mostly occur in the dry season (Sackey et al. 1993; Bandaranayake 1998; deGraft-Johnson et al. 2010). Mangroves for servicing of boat and fishing gear, furniture, tannins are used to dye leather for bags, belt and sandals production and also chemical extracts from mangroves are used as insecticides against many insects including mosquitoes (Bandaranayake 1998).

In some few cases, some coastal communities use mangroves vegetation as site for illegally disposing all types of household waste household (Numbere 2019). In Ghana, illegal mining for mineral resources especially gold which is locally called "galamsey" has also removed mangrove vegetation in some of the coastal communities. However, some few populations of mangroves along the coast of are naturally removed through disease and pathogenic attack and/or global warming (Bhowmik 2022; Nunoo and Agyekumhene 2022) Lack of enforcement of the existing environmental laws in the coastal areas has been one of the main drawbacks of protecting the few remaining mangrove populations from extinction in Ghana (Fianko & Dodd 2019; Nunoo and Agyekumhene 2022). In some few coastal communities, traditional leaders have introduced "traditional taboos" to deter encroachers from removing mangrove vegetation where it is against the gods to remove mangrove vegetation without any permit from the fetish priests, Chiefs and elders (Abeku Essel 2020; Abayie-Boaten 1998). Several attempts to restore and conserve mangrove vegetation in Ghana by government and non-governmental organizations in collaboration with coastal communities such as Songor and Keta Lagoon Complex Ramsar sites failed to due to land related issues including ownership, access and land tenure system. (Asante et al. 2017).

Many researchers have reported that Ghana has lost greater percentage of its original mangrove cover between 2002 and 2022 (Dankwa 2002; Mensah 2013; Aheto 2016; Essel et al. 2019; Dali 2020; Nunoo and Agyekumhene 2022; Dali et al. 2023). Ghana's mangrove cover therefore needs immediate and appropriate interventions to protect the few remaining vegetation from degradation that could lead to extinction. Losing mangroves and *Conocarpus* vegetation may financially affect coastal communities that depend on these species for livelihood.

Mangroves and *Conocarpus* have many local uses. In Nigeria the leaves of these species are either eaten raw or leave extract is drunk for treatment of fever or malaria, however, root extract of the same species is boiled, cooled down and drunk for treatment of gonorrhoea, coughing and other respiratory diseases(Adda 2016). Again, latex extracted from the bark of mangroves is also used stop bleeding from wounds when there is cut on the skin (Adda 2016).

Mangroves and *Conocarpus* are usually shrubs of about 1.5m to 4m in height but can also grow as trees of about 20m tall (Ghate and Sridhar 2016). Other morphological features common to both species include possession of weak and brittle laterals and fine roots with dark brown colour, possession of erect or multiple trunks (Farooq et al. 2018). Both mangroves and *Conocarpus* possess thin corky bark of about 8mm thick which is grey or brown in colour from outside but the inner part is dark cream in colour (Farooq et al. 2018). In both species the stem in both mangroves and *Conocarpus* is hard, heavy and strong, however, the branches are brittle with slender yellow-green twigs (Farooq et al. 2018). The leaves of mangroves and *Conocarpus* are spirally arranged, elliptic, lanceolate and fleshy with measurement of about 2-10cm long which includes leaf petiole of about 3-9mm long (Farooq et al. 2018). Uddin, Huang and Bin (2019) documented that mangroves grow in association with other plant species including *Conocarpus erectus* in brackish water environment but there is a clear morphological indication that *Conocarpus* may not be the same as mangrove species. C*onocarpus erectus* is not much studied in the mangrove forest areas in Ghana and there have been different views, arguments and opinions in Ghana on classification of *Conocarpus erectus* as mangrove species. Many Ghanaians including some researchers classify *Conocarpus erectus* as mangroves due to some similar morphological features they share in common (Nunoo and Agyekumhene 2022). Nortey et al. (2016) used observable characters and classified *Conocarpus erectus* as one of the mangrove species in Ghana. Others including Boateng (2018) also used morphological feature to classify *Conocarpus erectus* as one of the six main mangrove species found in Ghana. Several other studies have also used similar morphological approaches including reliance on morphological characteristics of the species to classify *Conocarpus erectus* as mangrove species (Asuk et al. 2018; Yaney-Keller et al. 2019). However, Nascimento et al. (2016) and Aheto et al. (2016) argued that using morphological features alone might not be sufficient enough to establish a strong basis for *Conocarpus erectus* classification as a mangrove without any DNA analysis therefore may be scientifically wrong. The researchers further argued that morphological analysis technique alone is not sufficient enough for such classification. Distinguishing between mangroves and

Cornocarpus erectus using morphological features is possible, however, it is very difficult to identify individuals in mixed populations. An estimation and description of genetic relationships such as possible hybridisation between the species and introgression of the populations in the field is difficult using morphological and phenological characters. The use of molecular markers might enhance understanding and might help distinguishing between mangroves and *Cornocarpus erectus*. Aheto (2016) therefore recommended the need to distinguish between the two based on DNA analysis.

This study therefore aims at using Random Amplified Polymorphic DNA (RAPD) to investigate genetic relationship between *Rhizophora mangle*, *Avicinnea germinans, Laguncularia racemosa and Conocarpus erectus.* RAPD is a PCR based technique that detects nucleotide sequence (8–12 nucleotides) polymorphisms in a DNA amplification-based assay using only a single primer of arbitrary nucleotide sequence. The RAPD technology has provided a quick and efficient screen for DNA-sequence polymorphisms at a very large number of loci. RAPD is good as it requires no prior knowledge of sequence and also a small amount of DNA is needed for amplification (Williams et al. 1990). RAPD is quick and economical for population genetics studies as compared with other molecular markers. The molecular basis for RAPD has been discussed (Hadrys et al. 1992) and may provide either single base pair changes in the primer binding sites or deletion in the region between the primer binding sites.

Though RAPD is simple, the major drawback is that, their results lack reproducibility of some amplification products. Again, RAPD is criticised in population genetics because heterozygosity cannot be determined (Williams and St. Clair 1993). Despite these criticisms, RAPD is used in population genetics studies. The objective of this paper is to use clearly amplified RAPD fragments to examine the genetic variation within and between populations of *Rhizophora racemosa, Avicenna germinans, Laguncularia racemosa and Carnocarpus erectus* from Shama in the Western region of Ghana. Inadequate information on mangroves in Ghana is affecting its management and protection in order to prevent the species from extinction. This study will provide information on genetic diversity of the three most common mangroves and *Cornocarpus erectus* in Ghana so that effective conservation strategies could be put in place to prevent the few remaining populations from extinction.

MATERIALS AND METHODS

Sample collection

Healthy, young and fresh leaves of *Rhizophora mangle*, *Avicinnea germinans, Laguncularia racemosa and Conocarpus erectus* were collected from Anlo Beach along the coastal belt in the Shama District in the Western Region of Ghana (Fig. 1). The area lies within latitudes 5°1'30" N and 5°3'5" N, and longitudes 1°34'30" W and 1°37'30" W and also covers about 50.42 km^2. Samples were collected in September, 2021. and stored with silica gel to effect rapid drying of samples in different labelled zip-lock plastic bags until extracted in the laboratory. From each of the sites, three individuals were randomly selected and small quantities of leaf samples were harvested.

2.2. DNA extraction and amplification

Random Amplified Polymorphic DNA (RAPD) method was used to characterize mangroves and *Conocarpus erectus*. The silica gel-dried leaf samples (1 g) were ground into fine powder in liquid Nitrogen using a pre-chilled mortar and pestle. The ground powder was quickly transferred to a centrifuge tube. A total of 20mg of the powdered leaf samples was used to extract genomic DNA using DNeasy plant mini kit (QIAGEN, UK.LTD.) at the Molecular Laboratory of Forestry Research Institute of Ghana (FORIG), following the manufacturer's procedure as follows:

Addition of lyses buffer and RNase to the leaf sample and incubated for 10 mins at 65ºC in a water bath. RNase digests the RNA in the sample. Lysate was spun at 13,000 rpm for 5 min to remove salt precipitate, detergents, proteins and polysaccharides. This step was repeated twice due to wide spectrum of polysaccharides and polyphenols including flavonoids and other secondary metabolites of mangroves and *Conocarpus.* Which could interfere with the extraction of pure genomic DNA. The non-pelleted precipitate and cell debris were removed by transferring the supernatant to a QIAshredder spin column and spun for 2 min at 13000 rpm. Binding buffer to which a specified amount of ethanol had been added (prior to use) was added to the collected lysate in a new tube to promote binding of DNA to the DNeasy membrane. The mixture was transferred to a DNeasy mini spin column and spun for 1 min at 8000 rpm to enable the DNA bind to the membrane. This is promoted by addition of ethanol (prior to use) to buffer AP3 while remaining contaminants such as polysaccharide and proteins were removed by a two-wash step with buffer AW. The first step involved spinning for 1 min at 8000 rpm and second step involved spinning at the same speed for 2 min to dry the membrane and get rid of residual ethanol that might be carried over to the next step to interfere with the remaining reactions. Pure DNA was eluted in a 100 µl volume of low salt suspension buffer after transferring the DNeasy mini column to another collection and storage tube and spun for 1 min at 8000 rpm after salt suspension buffer had been added. A second elution with another 50µl was repeated.

Quantification of the DNA extracts was performed by using spectrophotometer (UV/Vis Spectrophotometer U-2001, Hitachi Instruments Inc.) to measure absorbance at 260nm (A260) before use. Estimating the ratio of absorbance at 260 nm to that of 280 nm and 230 nm was used to check the purity of the extracted DNA. The ratio (A260/A280) was used to check any contamination by RNA or protein and the ratio (A260/A230) was also used to check any contamination by polyphenol or polysaccharide or both. The quality of the extracted DNA was verified by running the extracted DNA samples on 0.8% agarose gel stained with ethidium bromide in 1×TBE buffer solution at 80V for 90 minutes. The agarose gel was visualized and photograph was taken under UV trans-illuminator.

Primer screening

Random testing of primers for their polymorphism in mangroves and *Conocarpus* were initially used to select best primers that could be used to differentiate the species. A total of 15 different primers were screened for their quality. Reproducibility of the amplified products was tested and the same reaction was repeated at least twice. Out of these, 10 primers which produced strong and reproducible amplification profiles were selected for the study.

PCR amplification and running of agarose DNA gels

DNA amplification reactions were performed in volumes of 25µl per run per individual sample containing a reaction mixture of DNA suspended in buffer solution, primer, Taq DNA polymerase enzyme, nucleotides (DNTPs), 1 x enzyme buffer and MgCl2. Each amplification test included a negative control omitting template DNA (targeted DNA replaced by deionized water). Thermocycling was conducted in a Hybaid PCR Express Thermocycler (PCR Express, Hybaid, UK., Ltd.) with 36 cycles of 94^{O}C for 2 min where double stranded DNA is denatured; 45^{O}C for 1min for primer annealing; the last extension cycle was 72^{O}C for 2min. A final extension cycle of 72^{O}C for 7min was done soon after completion of all the cycles. According to Doulis et al. (2000), this cycle ensures a complete extension of the DNA. PCR products were stored at 4^{O}C before electrophoresis. PCR samples were run on a 1% ethidium bromide stained agarose gel in 1×TBE buffer solution at 100V for 80 minutes. DNA amplification fragments were visualized under ultraviolet trans-illuminator and bands were observed in gel documentation system (Alpha Innotech) and photographed using Gel Doc (Bio Rad).

Data analysis

The banding patterns obtained from the RAPD gel photograph were scored for the presence or absence of specific PCR bands (DNA marker) generated by a primer. Presence and absence of bands were represented by 1 and 0 respectively. Bands of low visual intensity were considered ambiguous and were not scored. Data obtained from the bands scoring were pooled for different analyses including Jaccard's coefficient of similarity (Jaccard, 1908) which was measured to generate a dendrogram based on similarity coefficients generated by the un-weighted pair group method using arithmetic averages (UPGMA) and hierarchical cluster analysis (Sneath and Sokal, 1973) was performed using DARwin version 6.0.21. Confirmation of the grouping of the accessions was done by performing Principal co-ordinate analysis (PCA).

RESULTS

Although all fifteen RADP primers used in the experiment produced clear bands for the determination of variability among the three mangroves and *Conocarpus erectus* accessions ten out of the fifteen primers produced more clearly polymorphic bands and were considered for further analysis. Purity of extracted DNA was good. The A260/A280 ratio ranged from 1.68 to 1.74 and A260/A230 ratio was > 2. This showed that the preparations were adequately free of proteins, polyphenolics and polysaccharide compounds. DNA concentration was also between 6.8 and 7.6µg/mL. Average number of 12 and10.2 bands were obtained per primer for the mangrove species and *Conocarpus erectus* respectively and amplification products ranged in size from 50 bp to 2000 bp. Average number of 85.3 and 85.1 polymorphic bands were obtained per primer for mangroves and *Conocarpus erectus* respectively with the highest number of polymorphisms obtained with primers GEN I-60B and GEN I-60J and the lowest with primers GEN I-60C and GEN I-60E (Table 1). A considerable amount of genetic variation within and between populations of mangroves and *Conocarpus erectus* was detected by the 10 RAPD primers used in this stud (Table 2).

Table 1 RAPD Profiling of *mangroves and Conocarpus* showing levels of polymorphism from 10 primers.

Primer code	Nucleotide sequence (5'-3')	Total no. of bands	Polymorphic bands Number	%	Molecular size range (bp)
GEN I-60A	CGCAGTACTC	15, *11*	12,*10*	80, *90.9*	200-2000, *200-2000*
GEN I-60B	GTCCTACTCG	11, *10*	11,*9*	100, *90*	200-2000, *200-2000*
GEN I-60C	CTACACAGGC	9, *7*	8, *6*	88.9, *85.7*	200-1500, *200-1300*
GEN I-60D	GTCCTTAGCG	13, *10*	11, *8*	84.6, *80*	200-2000, *200-2000*
GEN I-60E	GTCCTCAACG	8. *9*	7, *7*	87.5, *77.8*	200-1000, *200-1200*
GEN I-60F	GAGTCACTCG	9, *11*	8, *9*	88.9, *81.8*	200-2000, *200-2200*
GEN I-60F	GTCCTCAGTG	15,*10*	11,*8*	73.3 , *80*	200-2200, *200-2000*
GEN I-60H	CGTCGTTACC	14,*11*	11,*9*	78.6 , *81.8*	200-2000, *200-2000*
GEN I-60I	GCAGACTGAG	14,*12*	10,*11*	71.4, *91.7*	200-2000, *200-1500*
GEN I-60J	GCAGACTGAG	12,*11*	12,*10*	100, *90.9*	200-2000, *200-2000*
Average		**12, *10.2***	**10.1, *8.7***	**85.3, *85.1***	

*Numbers in italics are RAPD profiling of *Conocarpus erectus*

Table 2
Analysis of molecular variance (AMOVA) for 19 mangroves and 10 *Conocarpus erectus* individuals from Anlo Beach

Source of variation	df	SS	MS	Est. Var.	%	PhiPT
Among population	1	17.841	17.641	0.660	6.0	0.863
Within population	26	254.409	0.785	9.785	94.0	
Total	27	272.25		10.445	100.0	
*p < 0.01						

Analysis of molecular variance (AMOVA) identified 6% variance among and 94% variance within subpopulations (Table 2), indicating a high gene exchange or low genetic differentiation between the four populations. The PhiPT-value was 86.3% (0.863). indicates considerable degree of difference between the populations. There is therefore genetic variation between the mangroves *and Conocarpus erectus* populations.

The dendrogram obtained was generated based on dissimilarity values from DARwin analysis (Fig. 3) The figure shows that M10 and M9 have the lowest dissimilarity. This is supported by the index that recorded 40% (0.4) dissimilarity between them (Fig. 4). Most of the *Conocarpus erectus* clustering together with some mangroves indicates that they share some few similar genes together in contrary to many common morphological features they share together.

The principal co-ordinate analysis clearly separated mangroves and *Conocarpus* into four distinct groups along the first coordinate axis (Fig. 5), however, *Conocarpus erectus* accessions clustering together in fourth quadrant and also separating themselves from mangroves gives an indication of genetic differences between mangroves and *Conocarpus erectus*.

Discussion

In our study, nineteen accessions of mangroves and nine accessions of *Conocarpus erectus* were fingerprinted using ten RAPD marker. Partitioning of variation showed that most of the variation was detected within the populations of mangroves and *Conocarpus.* The successful identification of these two species using species-specific nuclear markers and only a limited set of primers proved to be promising for species detection. The DNA amplification fingerprinting technique allowed us to unambiguously differentiate *Rhizophora mangle*, *Avicinnea germinans, Laguncularia racemosa and Conocarpus erectus* populations. Although the three mangrove species have many similar morphological structures with *Conocarpus erectus* including leaf morphology, leaf length, colour of petiole, colour of the midrib, they shared just a few similar genes together and their RAPD fingerprinting differed markedly to show distinction between mangroves and *Conocarpus erectus*. This was supported by the high PhiPT-value of 0.863 which showed high degree of differentiation between the mangroves and *Conocarpus erectus* populations

The nine elements of *Conocarpus ecrectus* got separated from the other nineteen mangroves constituting red, white and black mangroves. However, few accessions of *Conocarpus* clustered with the mangroves indicating that they share some few common genes which could account for the similar morphological features they share in common and also justifying their taxonomic alienation. The result was also confirmed using principal coordinate analysis (Fig. 5). showing grouping of the three species of mangroves together and segregating that of *Conocarpus ecrectus* were segregated in the groupings. However, a few accessions shared some commonalities with mangroves.

Analysis of molecular variance (AMOVA) identified 96% variance within populations, indicating a high gene exchange (or low genetic differentiation) within populations mostly of *Rhizophora mangle*, *Avicinnea germinans and Laguncularia racemosa* but AMOVA identified very low gene flow of 4% variance between populations which is mostly between mangroves and *Conocarpus erectus* populations. There is therefore genetic barrier preventing free flow of gene between mangroves and *Conocarpus erectus* making them two distinct populations. This findings is in line with other researchers including Allendorf, Luikart and Aitken (2013) and Frankham, Ballou and Briscoe (2002) that gene flow is critical for population resilience and persistence as environmental conditions change because it improves genetic diversity within and between populations especially following major disturbances. This study also strengthens other studies including Carugati et al. (2018) that the alarming rate at which mangroves forests are deteriorating due to anthropogenic impacts and global change calls for the need to preserve the few extant populations from extinction. Restoring degraded mangrove forests will help provide the needed goods and services to support biodiversity and proper functioning of the ecosystem.

Conclusion

This study demonstrated that RAPD marker can be an effective and useful method to show differences among individual genotypes, assessed and delineated the pattern of genetic variation within and among populations of *Rhizophora mangle*, *Avicinnea germinans, Laguncularia racemosa and Conocarpus erectus*. Despite the reproducibility problem, the RAPD method is probably important due to its straightforwardness, less time involved, low labour the primers and other chemicals used in the whole process are relatively cheap as compared to other markers. Though mangroves and *Conocarpus erectus* populations shared many morphological features, some genes are shared genetically. Exchange of genes between the two populations is very low indicating genetic hinderance preventing successful fertilization or germination of pollen on stigmatic surfaces of mangroves and *Conocarpus erectus.* The two major species are distinct even though they shared some similar genes as well as many morphological characters. Though they can be classified as mangroves, however, they are not true mangroves. This study has provided useful baseline information on genetic diversity of mangrove and *Conocarpus erectus* that could be used on conservation and management of the species in Ghana. However, due to limitations of RAPD data, the present study recommends the need to use other DNA markers in studying genetic variation between mangrove and *Conocarpus erectus* in order to compare results from RAPD. Again, more numbers of mangrove species and *Conocarpus erectus* should be used. Again, there should be continuous education and law enforcement as well as active community involvement in protecting the few remaining mangrove vegetation in Ghana.

Declarations

Author Contribution

Author D.D. and RAA wrote the main manuscript text and prepared all figures and tables. Authors E.A. and D.S. helped with data collection and laboratory work and data analysis.

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Figures

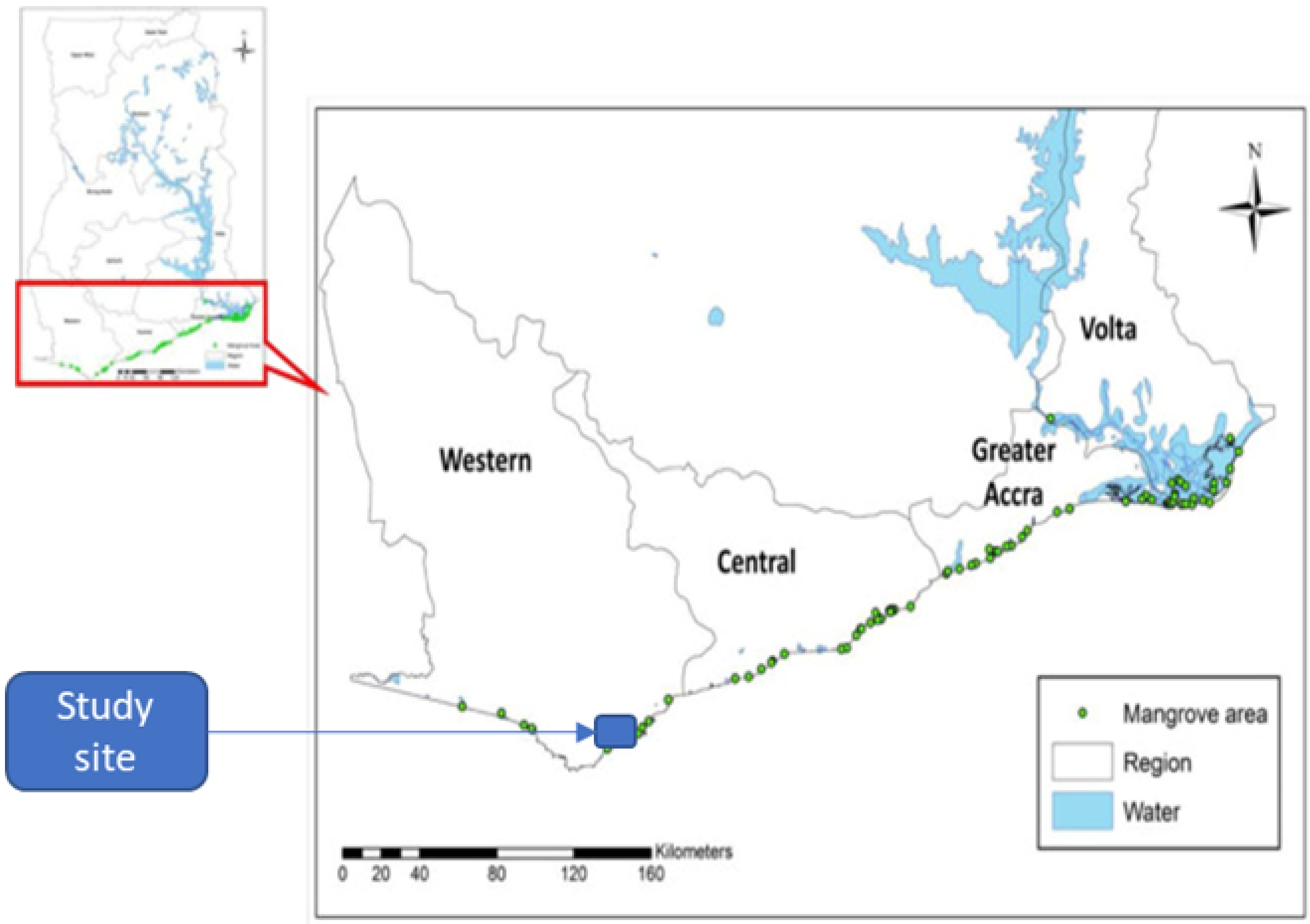

Figure 1

Map of Ghana showing Anlo Beach in the Shama District of the Western region of Ghana

Source: Nunoo and Agyekumhene, 2022

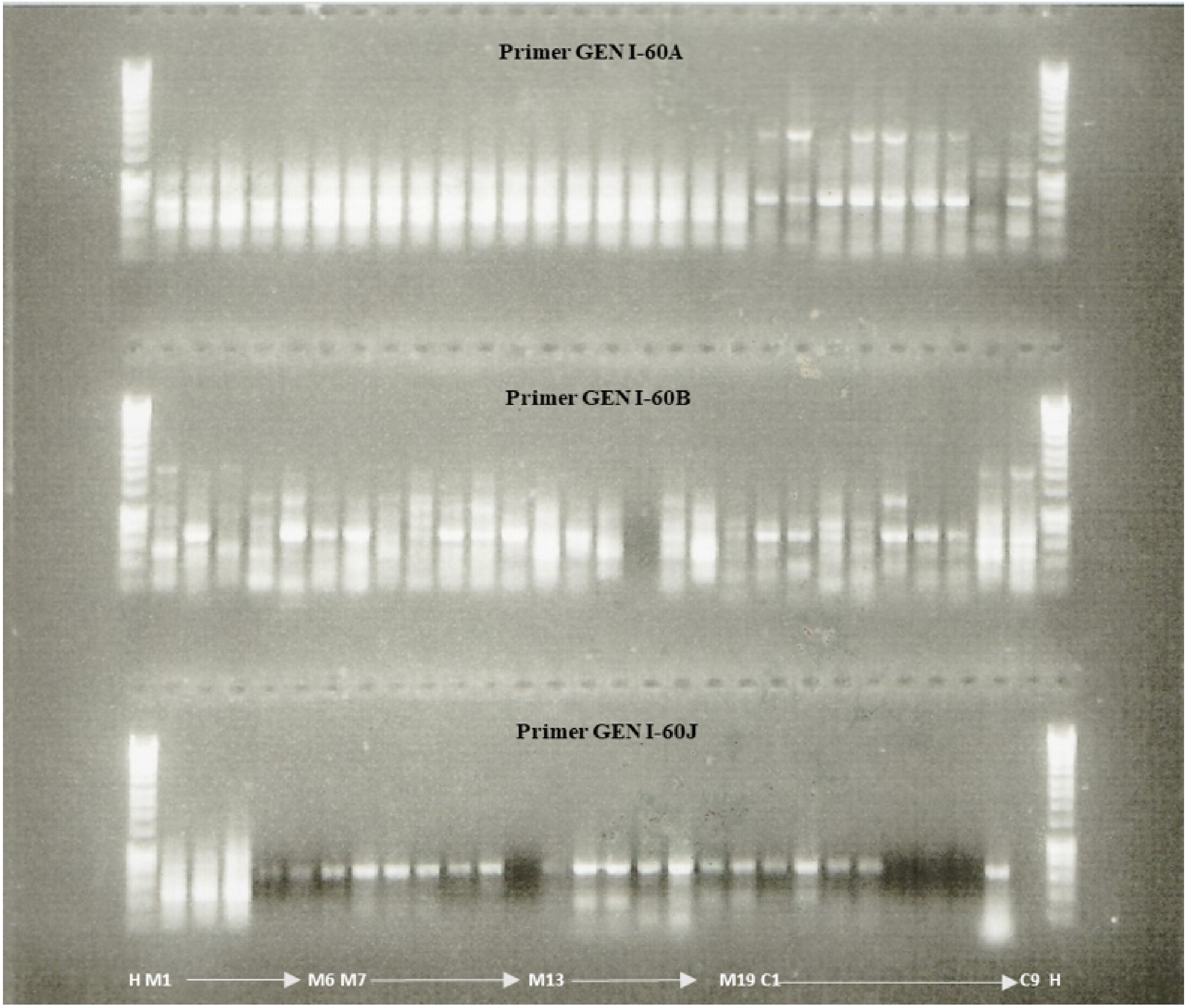

Figure 2

Gel Image showing the PCR product amplifications using three different primers.

H=hyperladder, M1-M6=white mangroves, M7-M14=red mangroves, M15-M19=black mangroves C1-C9= *Conocarpus erectus*

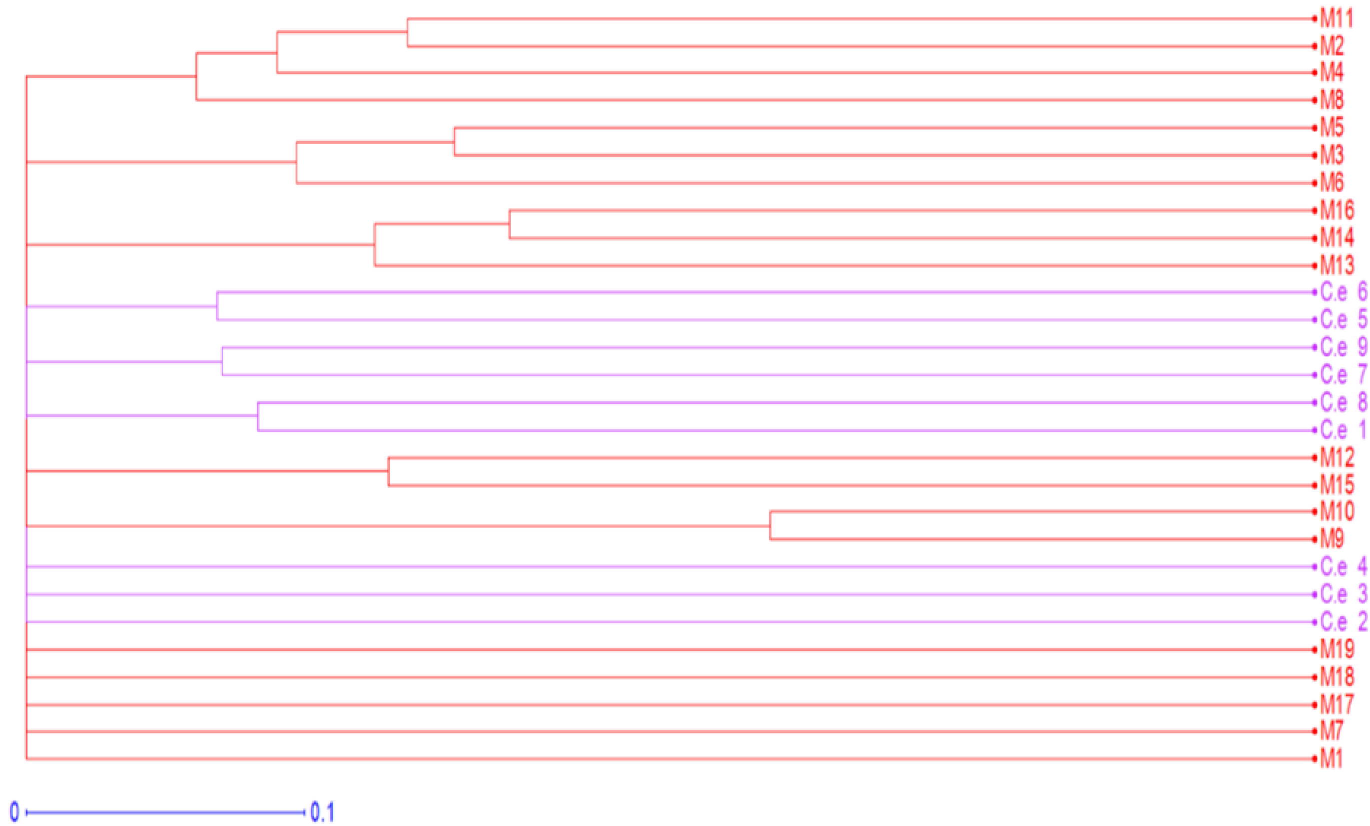

Figure 3

Dendrogram of the genomic relationship among 19 mangroves and 9 *Conocarpus erectus* accessions using RAPD markers. M1-M6= *Rhizophora racemosa*, M8-M14 =*Lagunculalia racemose*, M15-M20 =*Avicennia germinalis*, C1-C9= *Conocarpus erectus*

	M1	M2	M3	M4	M5	M6	M7	M8	M8	M10	M11	M12	M13	M14	M15	M16	M17	M18	M19	Ce1	Ce2	Ce3	Ce4	Ce5	Ce6	Ce7	Ce8	Ce9
M1	0.9																											
M2	0.8	1.0																										
M3	0.9	0.7	0.9																									
M4	0.7	1.0	0.8	1.0																								
M5	0.8	1.0	0.7	1.0	0.7																							
M6	0.8	1.0	1.0	1.0	0.8	1.0																						
M7	1.0	0.7	0.8	0.7	1.0	1.0	1.0																					
M8	0.8	0.7	0.8	0.6	0.7	0.6	1.0	0.7																				
M9	1.0	0.8	1.0	0.7	0.9	1.0	1.0	0.6	0.4																			
M10	0.9	0.5	0.9	0.6	1.0	1.0	1.0	0.7	0.7	0.8																		
M11	1.0	0.7	1.0	1.0	1.0	1.0	1.0	0.9	1.0	0.8	0.6																	
M12	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	0.8	0.6	1.0	0.8																
M13	0.9	0.9	0.7	0.9	1.0	1.0	1.0	0.8	1.0	1.0	0.8	0.8	0.7															
M14	1.0	1.0	1.0	1.0	1.0	0.9	1.0	1.0	1.0	0.8	0.9	0.6	0.5	0.8														
M15	1.0	1.0	1.0	1.0	0.9	1.0	1.0	1.0	1.0	0.9	1.0	0.9	0.5	0.6	0.7													
M16	1.0	1.0	1.0	1.0	0.8	0.9	1.0	1.0	1.0	0.9	1.0	0.9	0.9	0.7	0.7	0.8												
M17	1.0	0.7	1.0	0.7	1.0	1.0	1.0	0.8	0.7	0.7	0.7	0.9	1.0	0.9	1.0	0.9	0.8											
M18	0.9	1.0	1.0	1.0	0.9	1.0	1.0	1.0	1.0	0.7	0.9	0.8	0.7	0.8	0.8	0.9	0.7	1.0										
M19	0.8	1.0	1.0	1.0	0.8	1.0	1.0	1.0	1.0	0.7	0.9	0.9	0.8	0.7	0.8	0.9	0.8	0.9	1.0									
Ce1	1.0	1.0	1.0	1.0	1.0	1.0	1.0	0.9	1.0	1.0	1.0	1.0	1.0	0.7	0.7	0.7	1.0	0.8	1.0	0.8								
Ce2	0.9	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	0.9	1.0	1.0	0.8	1.0	1.0	1.0	0.1	0.9	0.9	0.8							
Ce3	1.0	0.9	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	0.9	1.0	1.0	0.9	1.0	1.0	1.0	1.0	0.9	1.0						
Ce4	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	0.9	0.9	1.0	1.0	1.0	0.9	1.0	1.0	0.8	1.0	0.5	0.8	1.0	1.0					
Ce5	1.0	1.0	1.0	1.0	1.0	1.0	1.0	0.8	1.0	0.8	1.0	1.0	1.0	1.0	0.8	0.8	1.0	1.0	1.0	1.0	0.7	1.0	0.9	1.0				
Ce6	1.0	0.9	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	0.9	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	0.9	0.9	0.7			
Ce7	0.9	1.0	0.9	0.9	1.0	1.0	1.0	1.0	1.0	1.0	0.9	1.0	1.0	0.7	1.0	1.0	1.0	0.8	1.0	0.8	0.8	1.0	0.9	0.8	1.0	1.0		
Ce8	1.0	0.9	1.0	1.0	1.0	1.0	0.9	0.9	1.0	1.0	0.9	0.9	1.0	0.8	0.8	1.0	0.9	0.8	1.0	1.0	0.6	0.9	0.8	1.0	0.8	1.0	1.0	
Ce9	1.0	1.0	1.0	0.9	0.8	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	0.9	1.0	1.0	0.8	1.0	0.8	0.8	0.8	0.9	1.0	0.5	1.0	0.9	0.8	0.9

Figure 4

A dissimilarity index of 19 mangroves and 9 *Conocarpus erectus accessions* obtained with 10 primers using Darwin version 6.0.21.

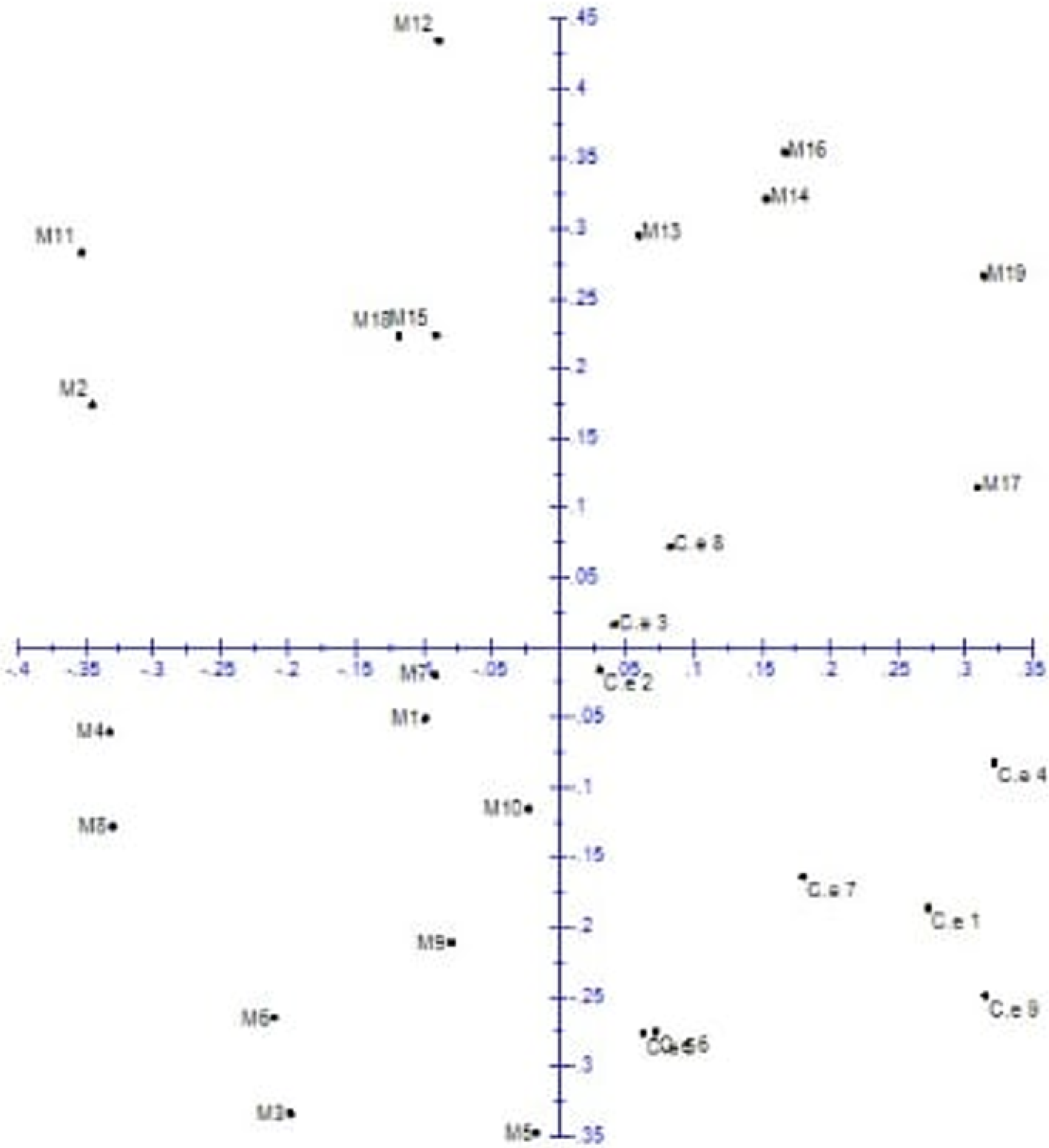

Figure 5

Principal co-ordinate analysis (PCoA) showing grouping of mangroves and *Conocarpus erectus* (the abbreviations are as used in Fig. 3&4).