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Abstract:	Background The Coconut palm (Cocos nucifera, 2n = 32 family Arecaceae (Palmaceae), is an impor tropical tree crop is cultivated in 93 countrie East and West Africa, Southeast Asia and t of more than 11 million hectares. The Coco two main categories: "Tall"(flowering 8-10 y 4-6 years after planting), based on morphol The long generational time of this tropical s breeding. In spite of initial successes, gene Findings A total of 714.67 gigabases (Gb) of raw dat platform, comprising approximately 285.869 (variety "Hainan Tall"). After filtering the low insert size, 419.08 gigabases (Gb) of clean assembled with SOAPdenovo2 [29]. A total with a scaffold N50 of 418 Kb, which repres (2.42Gb). BUSCO evaluation demonstrated reached 90.8%. The coconut genome was genes, which is less than Phoenix dactylifer (34,802). The annotation completeness was 74.1%. Genome annotation results revealed consists of transposable elements, among w make up the largest proportion (92.23%). Conclusions Despite its agronomic importance, Cocos n genome draft of Cocos nucifera. This study information, facilitatingfuture functional genu nucifera.	2), a member of genus Cocos and of the tant tropical fruit and oil crop. Currently, this is, including Central and South America, he Pacific islands, with a total growth area nut palm can generally be classified into ears after planting) and "Dwarf" (flowering ogical characteristics and breeding habits. pecies hinders progress in genetic tic improvement is very slow. a was acquired by the Illumina HiSeq 2000 coverage of the Cocos nucifera genome quality reads, PCR duplication and small data was obtained, these clean reads were scaffold length of 2.20 Gb was generated, the completeness of the coconut genome the completeness of the coconut genome and the completeness of the coconut genome predicted to harbor 28,039 protein-coding ra (DPV01, 4,166) and Elaeis guineensis is also evaluated by BUSCO, reached d that 72.75% of the coconut genome which long-terminal repeat elements (LTRs) ucifera is still under-studied. We report a provides a large amount of genomic omics and molecular breeding in Cocos		
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All datasets and code on which the conclusions of the paper rely must be either included in your submission or deposited in <u>publicly available repositories</u> (where available and ethically appropriate), referencing such data using a unique identifier in the references and in the "Availability of Data and Materials" section of your manuscript.	
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Background

The Coconut palm (*Cocos nucifera*, 2n = 32), a member of genus *Cocos* and of the family Arecaceae (Palmaceae), is an important tropical fruit and oil crop. Currently, this tropical tree crop is cultivated in 93 countries, including Central and South America, East and West Africa, Southeast Asia and the Pacific islands, with a total growth area of more than 11 million hectares. The Coconut palm can generally be classified into two main categories: "Tall"(flowering 8-10 years after planting) and "Dwarf" (flowering 4-6 years after planting), based on morphological characteristics and breeding habits. The long generational time of this tropical species hinders progress in genetic breeding. In spite of initial successes, genetic improvement is very slow.

Findings

A total of 714.67 gigabases (Gb) of raw data was acquired by the Illumina HiSeq 2000 platform, comprising approximately 285.86×coverage of the Cocos nucifera genome (variety "Hainan Tall"). After filtering the low quality reads, PCR duplication and small insert size, 419.08 gigabases (Gb) of clean data was obtained, these clean reads were assembled with SOAPdenovo2 [29]. A total scaffold length of 2.20 Gb was generated, with a scaffold N50 of 418 Kb, which represents 90.91% of the estimated genome (2.42Gb). BUSCO evaluation demonstrated the completeness of the coconut genome reached 90.8%. The coconut genome was predicted to harbor 28,039 protein-coding genes, which is less than Phoenix dactylifera (DPV01, 4,166) and Elaeis guineensis (34,802). The annotation completeness was also evaluated by BUSCO, reached 74.1%. Genome annotation results revealed that 72.75% of the coconut genome consists of transposable elements, among which long-terminal repeat elements (LTRs) make up the largest proportion (92.23%).

Conclusions

Despite its agronomic importance, Cocos nucifera is still under-studied. We report a genome draft of Cocos nucifera. This study provides a large amount of genomic information, facilitatingfuture

61 functional genomics and molecular breeding in *Cocos nucifera*.

64 Data description

65 Background

66 Coconut (*Cocos nucifera*, 2n = 32), the only species of genus *Cocos* of family Arecaceae, is a 67 tropical oil crop and is widely cultivated in tropical regions due to its extensive application in 68 agriculture and industry. The tropical species was thought to have originated from the western pacific 69 region (including Malay Peninsula and archipelago, New Guinea, and the Bismarck Archipelago) and 70 the southwest Pacific. Presently, the tropical tree crop had been distributed across 89 tropical countries, 71 including Central and South American, East and West African, Southeast Asia and the pacific islands, 72 and accounts for over 12 million hectares of land.

In China, the coconut palm grows in the Hainan and Yunnan provinces as an economic and ornamental plant. In the province of Hainan, coconut is cultivated over an area of approximately 43,000 hectares, of which approximately 36,000 hectares is made up by the coconut variety "Hainan Tall" (HAT) [1]. Hainan Tall coconut are slow to mature (flowering 8-10 years after planting), can grow to a height of about 20-30 meters, and have medium to large nut size. Hainan Tall coconuts can adapt to a wide range of environment and have tolerance to biotic and abiotic stress, especially for high tolerant to high salt density. The morphological characteristics of Hainan Tall coconut were showed in Figure 1. Here we present Hainan Tall coconut genome sequence, making it possible to understand its adaption to high salinity. Moreover, the draft genome sequence of its relative species, Elaeis guineensis and Phoenix dactylifera, were also reported. The comparative analysis was performed about genome assembly and annotation between coconut and its relative species in the study.

84 Sample collection and sequencing

Genomic DNA was extracted from the spear leaf of a "Hainan Tall" coconut (*Cocos nucifera* L.
Taxonomy ID: 13894; 19033'3" N, 110047'25" E) individual selected from the coconut garden of the
Coconut Research Institute (Wenchang, Hainan province, China) using the CTAB extraction method
[2]. Subsequently, four pair-end (PE) libraries with insert size 170 bp, 500 bp, 450 bp and 800 bp and

five Mate-pair (MP) libraries with insert size 2 Kb, 5Kb, 10Kb, 20Kb and 40 Kb were constructed using the standard procedure provided by Illumina (San Diego, USA). After library preparation and quality control of DNA samples, template DNA fragments were hybridized to the surface of flow cells on an IlluminaHiSeq2000sequencer, amplified to form clusters, and sequenced following the standard Illumina manual. Finally, we generated 714.67 Gb of raw reads from all constructed libraries, raw sequenced outputs for each library are summarized in Table 1. Before assembly, reads with low quality (base quality less than 7 with percent higher than 25% or N percent higher than 1%), PCR duplication or adapter contamination were removed by using SOAPfilter, a software application in the SOAPdenovo package [3]. After filtering, 419.08 Gb (173.17×) high-quality sequences were obtained for genome assembly.

99 De novo assembly of short reads of Cocos nucifera

We used clean reads of the short-insert libraries (170bp) to estimate the coconut genome size by k-mer
frequency distribution analysis [3]. The genome size (G) of *Cocos nucifera* could be estimated by the
following formula:

$$G = N \times (L - 17 + 1)/K_{depth}$$

where N represents the total of number of reads, L represents the read length and K_depth refers to the
main peak in the k-mer distribution curve. In our calculations, N was 2,049,520,223, L was 100 and
K_depth was 71, therefore *Cocos nucifera* genome was estimated to be 2.42 gigabases (Gb). K-mer
size distribution analysis (Figure 2) indicated that *Cocos nucifera* was a diploid species with low
heterozygous and high repetitive sequence.

We then assembled the Cocos nucifera genome using SOAPdenovo2 in three steps: contig construction, scaffold construction and gap filling. In the contig construction step: the SOAPdenovo2 with parameters "pregraph-K 63 -R -d 1" was employed to construct de Bruijn graphs from pair-end libraries with an insert size from 170 to 800 bp. Then the kmers from the de Bruijn graphs were used to form contiguous sequences (contigs) with the parameters "contig -R" by clipping tips, merging bubbles and removing the low coverage links. In the scaffold construction step: the orders of the contigs were

determined using paired-end and mate-pair information with parameters "map -k 43" and "scaff -F -u". Initially, SOAPdenovo2 map the reads from pair-end and mate pair libraries to contigs based on a hash table (keys are unique k-mers on contigs; values are positions). In this case, two contigs are considered to be linked if the number of read pairs bridging the contigs exceeds the threshold three. Gaps within scaffolds were filled by utilizing KGF [4] (V1.06) and GapCloser software (v1.12-r6) [5] with pair-end libraries with an insert size from 170 to 800 bp in cases where one end could be mapped to one contig and the other end extended into a gap. To achieve optimal assembly result, Rabbit (a Poisson-based K-mer model, ftp://ftp.genomics.org.cn/pub/Plutellaxylostella/) was used to determine repeat sequences, segmental duplications or divergent haplotypes on the assembly. After removal of redundant sequences, a total scaffold length of 2.20 Gb was generated, comprising 90.91% of the predicted genome size (Table 2), which was larger than the other species in palmae. Meanwhile, the obtained contig N50 was 72.64 Kb and the scaffold N50 was 418.06 Kb while the length of scaffolds less than 100 bp were excluded. Comparison of coconut assembly N50s with four previously published palm genomes Phoenix dactylifera (PDK30) [6], Phoenix dactylifera (DPV01) [7], Elaeis guineensis [8] and Elaeis *oleifera* [8] confirmed that our results were better quality (Table 3).

130 Genome evaluation

The 57,304 unigenes (transcript obtained from three different issues, spear leaves, young leaves and fruit flesh) reported in previous Fan's research [9] were aligned to the assembled genome of *Cocos nucifera* using BLAT [10] with threshold "E-value = 10e-6, identity = 90%, coverage >90%". The alignment results predicted that the assembled genome of *Cocos nucifera* covered 96.78% of all expressed unigenes, suggesting a high level of coverage (Table 4).

We also evaluated the completeness of the assembly using BUSCO [11], which quantitatively assess genome completeness using evolutionarily-informed expectations of gene content from near-universal single-copy orthologs selected from OrthoDB v9 (http://busco.ezlab.org/, plant set). BUSCO analysis showed that 90.8 and 3.4% of the 1,440 expected plant genes were identified as complete and fragmented, respectively, while 5.8 % were considered missing in the assembly. The BUSCO results showed that our assembly was more complete than the previously reported data palm and oil palm genome assembly (Table 5).

Repeat annotation

144 We combined a homology and *de novo* method to identify transposable elements (TEs) and the tandem

repeats in the Cocos nucifera genome. In homology step: TEs at DNA and protein levels were identified by searching against Repbase library (version 20.04) [12] with RepeatMasker (version 4.0.5) [13] and RepeatProteinMasker (v4.0.5) [13]. In de novo step: de novo libraries were constructed based the genome sequences using the *de novo* prediction program RepeatModeler on (http://www.repeatmasker.org/RepeatModeler.html,version1.0.5) and LTR FINDER [14] by removing contamination and multi-copy genes. Then the novel transposable elements were identified and classified using RepeatMasker. The tandem repeat sequences were identified by TRF (Tandem Repeat Finder) software [15] with the following parameters "Match=2, Mismatch=7, Delta=7, PM=80, PI=10, Minscore=50 and MaxPerid =2000". The total length of the tandem repeat sequences predicted by the software is 151,229,585 bp, comprising 6.86% of the coconut genome. Finally, a total of 1.6 Gb of non-redundant repetitive elements were identified, accounting for 74.48% of the coconut genome, while transposable elements took up 72.75%. The most predominant transposons were long-terminal repeat (LTR), which account for 92.23% of all TEs and 67.1% of the coconut genome (Table 6).

158 Gene prediction

We combined homology, *de novo* and transcript alignment to predict genes in *Cocos nucifera* genome. For homology prediction: The gene sets of Arabidopsis thaliana [16], Oryza sativa [17], Sorghum bicolor [18] and Zea [19] were downloaded from the phytozomev9.1 mays (https://phytozome.jgi.doe.gov/pz/portal.html). The gene sets of *Elaeis guineensis* and *Phoenix* dactylifera (DPV01) were download from the NCBI ftp site. The longest transcript was selected to represent the genes with alternative splicing variants. We aligned these homologous proteins to the coconut genome using TBLASTN [20] with parameter "-e 1e-5 -F -m 8", and connected the BLSAT hit results to candidate gene loci by SOLAR with parameter "-a prot2genome2 -z" (https://sourceforge.net/p/treesoft/code/HEAD/tree/branches/lh3/solar/). Next, we extracted the genomic sequences of candidate gene loci with up and down stream 1k flanking sequences, applying Genewise 2.2.0 [21] to define the intron-exon boundary. The genes with pre-stop codon or frame-shifted were excluded for further analysis.

For *De novo* prediction: We randomly selected 1000 full length genes (GeneWise score equal 100,
intact structure: start codon, stop codon, perfect intron-exon boundary) from gene sets predicted by
homology method to train the model parameters for AUGUSTUS2.5 [22]. Two software programs,
AUGUSTU2.5 and GENSCAN 1.0 [23], were used to do *de novo* prediction on repeat-masked genome

of *Cocos nucifera*. Genes with incomplete structure or protein coding length less than 150bp werefiltered out.

Then Genes from homology and *de novo* method were combined to get non-redundant gene sets by using GLEAN [24] with the following parameters: minimum coding sequence length 150 bp and maximum intron length 50 kb. Genes were filtered with the same thresholds used for homology annotation.

For transcriptome-based prediction: RNA-seq data (SRR606452) from previous Fan's study were mapped onto the coconut genome to identify the splice junction using the software Tophat [25]. And then cufflinks [26] was used to assemble transcripts by the aligned reads. The coding potential of these transcripts was identified using fifth-order Hidden Markov Model, which was estimated with the same gene sets used in AUGUSTUS training by trainGlimmerHMM, a application in the GlimmerHMM [27] package. The transcripts with intact open reading frames (ORFs) were exacted and the longest ORF was retrieved while multiple isoforms located in the same locus.

At last, we merged the GLEAN and the transcriptome result to form a comprehensive gene set using an in-house annotation pipeline. If a transcript gene model had not overlapped with the GLEAN result, the transcriptome result would be added to the GLEAN result. If a transcript gene model had overlapped (overlap length >100bp), the transcriptome result would be used to perfect the structure of the GLEAN result (such as adding the un-translated regions (UTRs), completing the exon boundary). A diagrammatic pipeline is shown in Figure 3.

194 Gene evaluation

After all these steps, we obtained a final gene set contained 28,039 genes (Table 5), which is less than the gene numbers of *Phoenix dactylifera* (DPV01, 41,660) and *Elaeis guineensis* (34,802). Meanwhile the BUSCO evaluation demonstrated 74.1 and 11.2% of 1,440 expected plant genes were identified as complete and fragmented, 14.7% genes were considered missing in the gene sets. The BUSCO results showed that our gene prediction was more completely than *Phoenix dactylifera* (PDK30) and *Elaeis guineensis*, less completely than *Phoenix dactylifera* (DPV01) (Table 7), maybe the higher repetitive elements hinder the gene prediction of coconut genome. Gene function annotation was identified bysequence similarity and domains conservation. In sequence similarity step: we searched the coconut protein coding against KEGG protein [28], SwissProt and TrEMBL [29] using BLASTP at a cut-off E-value threshold of 10⁻⁵. Then we use the best match of alignment to represent the gene function. We obtained 18,445 KEGG, 18,867 Swissprot and 24,882 Tremble annotated genes. In domains conservation step: InterProScan5.11-51.0 software [30] was employed to identify the motif and domain against the public databases Pfam [31], PRINTS [32], ProDom [33], SMART [34], PANTHER [35], TIGRFAM [36] and SUPERFAMILY [37]. This revealed that 21,087 of the coconut proteins had conserved motifs, 1,622 Gene Ontology (GO) terms were assigned to 15,705 coconut proteins from the corresponding InterPro entry [38].. In total, approximately 89.41% of these genes were functionally annotated using above methods,

213 Conclusion

214 Cocos nucifera (2n = 32) is an important tropical crop, and is also used as an ornamental plant in 215 the tropics. In the present study, we sequenced and *de novo* assembled the coconut genome. A total 216 scaffold length of 2.2 Gb was generated, with a scaffold N50 of 418 Kb. The data output of the coconut 217 genome will provide a valuable resource and reference information for the development of high density 218 molecular makers, construction of high density linkage maps, detection of QTL (quantitative trait loci), 219 genome-wide association mapping , and molecular breeding.

220 Availability of supporting data

Supporting data are available in the GigaDB database [39], and the raw data were deposited in the
SRA539146 with the project accession PRJNA374600 for *Cocosnucifera* genome. Previously
published RNA-seq data used for transcriptome-based prediction is available from the accession
number SRR606452.

225 Competing interests

226 The authors declare that they have no competing interests.

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3	234	Author's contribution
) L	235	YX, HF, YY, MP, QL, AG designed the study and contribute to the project coordination; XY, PX, WZ
2 3	236	wrote the paper; LZ, JL, YW collected the samples and extracted the genomic DNA; YX, BL, BS, JX
1 5	237	AA, EI, NL conducted the genome analyses.
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329 Tables

39.

Table 1 Sequencing libraries and data yields for whole genome sequencing

Library tpe	Reads Length(bp)	Insert Size(bp)	Raw data (Gb)	Clean data(Gb)
PE101	100	170	128.75(53.20)	111.32(46)
PE251	250	450	73.86(30.52)	56.42(23.31)
PE101	100	500	64(26.45)	65.11(26.90)
PE101	100	800	78.16(32.30)	64.90(26.82)
PE50	49	2000	128.6(53.14)	60.70(25.08)
PE50	49	5000	71.75(29.65)	18.62(7.69)
PE50	49	10000	74.65(30.85)	18.53(7.66)
PE50	49	20000	70.7(29.21)	19.35(7.99)
PE50	49	40000	24.2(10.08)	4.13(1.71)

331 Note: The sequencing depth was shown in parentheses, calculated based on a genome size of 2.42G. Clean data

were obtained by filtering raw data with low-quality and duplicate reads.

Table 2 Comparison of genome features of four palmae species

Genome features	PDK30	DPV01	EG	EO	Coconut
Assembly size (G)	0.38	0.56	1.54	1.40	2.20
Scaffold N50 (kb)	30.48	334.08	1045.41	333.11	418.07
Contig N50 (kb)	6.44	10.81	9.37	8.45	72.64
Gene Number	2,949	41,660	34,802	-	28,039
TEs percent (%)	23.6	38.87	43.24	-	72.75

Note: Coconut: *Cocos nucifer* (Hai nan Tall); PDK30: *Phoenix dactylifera* (PDK30); DPV01: *Phoenix dactylifera* (DPV01); EG: *Elaeis guineensis* (American oil palm E5 build); EO *Elaeis oleifera* (American oil palm, O8-build); The TEs result was obtained using the same pipeline with the Coconut

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Table 3 Summary statis	tics of fiv	ve palmae	species						
Species	Sequen	cing	Sequence	Estim	ated	Assembly	Con	tig	Scaffold
	technol	logy	coverage	size(Gb)	size(Gb)	N50	(Kb)	N50(Kb
Phoenix dactylifera (PDK30)	Illumin GAIIx	a	53.4x	0.66		0.38	6.44		30.48
Phoenix dactylifera	454,SO	DLiD,	139x	0.67		0.56	10.8	1	334.08
(DPV01)	ABI373	30							
<i>Elaeis guineensis</i> (African oil palm)	454		16X	1.8		1.54	9.37	,	1045.41
Elaeis oleifera	454		16x	1.8		1.40	8.45	i	333.11
(American oil palm)									
Cocos nucifera	Illumin	a	173X	2.42		2.20	72.6	4	418.07
(Hai nan Tall)	HiSeq								
	Tabl	e 4 The g	enome BUS	CO asses	ssment	of palmae s	pecies		
	Co	oconut	PDI	K30	D	PV01]	EG	
BUSCOs	Ν	P (%)	Ν	P (%)	Ν	P (%)	Ν	P (%)	N
Total	1440		1440		1440		1440		1440
Complete single-copy	1192	82.8	1042	72.4	1160	80.6	1100	76.4	1004
Complete duplicated	115	8.0	81	5.6	134	9.3	116	8.1	63
Fragment	49	3.4	98	6.8	42	2.9	60	4.2	84
	0.4	50	210	15.2	104	7.2	164	11.3	280

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Table 5 The gene coverage of Cocos nucifera by transcriptome data

Deteret	Number	Total	Base coverage	Sequence coverage
Dataset	Number	length (bp)	by assembly	by assembly (%)
All	57,304	43,090,665	96.78	99.57
>200bp	57,304	43,090,665	96.78	99.57
>500bp	25,713	33,470,388	96.36	99.85
>1000bp	13,796	25,004,919	95.99	99.94

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Table 6 Transposable elements in the coconut genome

	Repabse TEs	Protein TEs	De novo TEs	Combin	ed TEs
	length	length	length	length	percentage
DNA	20,936,158	24,655,089	35,131,002	58,119,982	2.64
LINE	4,251,185	9,631,472	7,610,172	19,197,064	0.87
SINE	85,717	0.00	186,364	270,055	0.012
LTR	361,968,154	512,700,933	1,419,281,798	1,478,182,089	67.10
Other	8,145	0.00	0.00	8,145	0.0004
Unknown	0.00	12,360	139,084,335	139,096,695	6.31
Total	385,037,442	546,965,774	1,552,582,881	1,602,630,396	72.75

361 Note: Repabse TEs means RepeatMask against Repbase; Protein TEs means RepeatProteinMask result against
362 Repbase protein; *De novo* TEs means RepeatMask against the *de novo* library; Combined TEs means the combine
363 results of three steps.

Table 7 The gene BUSCO assessment of palmae species										
	Coc	Coconut		PDK30		DPV01		EG		
BUSCOs	N	P (%)	Ν	P (%)	N	P (%)	Ν	P (%)		
Total	1440		1440		1440		1440			
Complete single-copy	965	74.1	748	51.9	1195	83.0	555	38.5		
Complete duplicated	102	7.1	81	5.6	159	11.0	53	3.7		
Fragment	162	11.2	255	17.7	44	3.1	270	18.8		

		Missing	211	14.7	356	24.8	42	2.9	562	39.0	
367 368 369	Note: C <i>dactylife</i> oil palm	Coconut: <i>Cocos r</i> era (DPV01); EG: , O8-build) was m	<i>uucifer</i> (Hai na <i>Elaeis guineen</i> iissing, not attai	n Tall); P <i>usis</i> (Ameri ined from t	DK30: <i>I</i> can oil p he public	Phoenix d alm E5 bu database;	<i>actylife</i> iild); Th	ra (PDK e gene of	30); DP f <i>Elaeis c</i>	V01: oleifera (<i>Phoenix</i> American
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371	Figure	legends									
372	Figure	1 Morphologica	l characteristic	es of coco	nut tree	(A) and c	coconut	tree bea	aring nut	ts (B).	
373	Figure 2 Kmer analysis of the coconut genome.										
374	Figure 3 The pipeline for integrating GLEAN and Transcriptome data.										

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Cover Letter

Dear editor

We would like to submit our manuscript entitle "The genome draft of coconut (*Cocos nucifera*)" for consideration of publication in Gigascience as a data note.

Coconut palm (*Cocos nucifera*, 2n = 32), belonging to the genus *Cocos* and the family Arecaceae (Palmaceae), is an important tropical fruit and woody oil crop. Meanwhile, the tropical crop is often used as an ornamental tree, which is a symbol for a tropical region. In order to accelerate molecular biology research and genetic breeding in *Cocos nucifera*, the whole genome of the tropical crop was sequenced and a total of 419.08 gigabases (Gb) of clean data was obtained, these clean reads were assembled with SOAPdenovo2. A total scaffold length of 2.2 Gb was generated, with a scaffold N50 of 418 Kb, which represents 91.67% of the estimated genome (2.4G). The coconut genome was predicted to harbor 28,039 protein-coding genes, which is slightly greater than *Phoenix dactylifera*(24,908). Genome annotation resultsrevealed that 72.75% of the coconut genome consists of transposable elements, among which long-terminal repeat elements (LTRs) make up the highest proportion (92.23%). The study provides a large amount of genomic information, facilitating future functional genomics and molecular breeding in *Cocos nucifera*. We believe that our study will be beneficial for the community, particularly who work on the study of genomic and molecular biology research in *Cocos nucifera*.

The authors also declare that the present work hasnot been submitted elsewhere for publication, in whole or in parts.Besides, all the listed authors have agreed and approved the contents of the manuscript.

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