The wax gourd genomes offer insights into the genetic diversity and ancestral cucurbit karyotype

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Supplementary Figure 1. Distribution of 17k-mer depth for the Illumina reads. Source data are provided as a Source Data file.



Supplementary Figure 2. Wax gourd genome. a Genetic markers distribution across 12 chromosomes. b Repeat coverage (50-90%). c GC content (30-40%). d Gene density (0-60). e-i Gene expression profile in root, stem, leaf, flower and fruit respectively (FPKM: 0-100). b-i are drawn in non-overlapping 1 Mb sliding windows.



Supplementary Figure 3. Consistence of the wax gourd genome assembly with the high-density genetic map. The horizontal axes show the assembled scaffolds as boxes with grey white alternating shades on the pseudo-chromosomes. The vertical ones represent the genetic distance in the genetic map.



Supplementary Figure 4. The divergence rates of repeats in the wax gourd genome. Divergence rate: percentage of substitutions in the matching region compared with consensus sequence in the repeat library. Source data are provided as a Source Data file.



Supplementary Figure 5. **Phylogenetic tree of NBS R genes in five** *Cucurbitaceae* **species.** Cucumber, melon, watermelon, bitter gourd, and wax gourd were involved in the analyses. The tree was generated using protein sequences of the conserved NB-ARC domain.



Supplementary Figure 6. Distribution of average synonymous substitution levels between collinear gene pairs in intergenomic blocks and intragenomic blocks. Ks: synonymous substitution. Solid curves: intergenomic blocks. Dashed curves: intragenomic blocks. **a** Before Ks correction. **b** After Eudicot-common hexaploidy (ECH) related evolutionary rate correction. Bhi: *Benincasa.hispida*, Cma: *Cucurbit maxima*, Cme: *Cucumis melo*, Lsi: *Lagenaria siceraria*, Cla: *Citrullus lanatus*, Csa: *Cucumis sativus*, Vvi: *Vitis vinifera*.



Supplementary Figure 7. The syntenic dotplots of wax gourd and melon versus pumpkin genome. The chromosomes marked by asterisk mean the whole chromosome have been well preserved in pumpkin genome. Source data are provided as a Source Data file.



Supplementary Figure 8. The syntenic dotplot of wax gourd and melon versus bottle gourd. Source data are provided as a Source Data file.



Supplementary Figure 9. Syntenic relationships across three genomes.



Supplementary Figure 10. The geographic distribution of the 146 accessions. Each accession is represented by a dot on the map. Magenta: cultivated wax gourds without wax; blue: cultivated ones with wax; green: Xishuangbanna landraces; orange: wild ones. The map was generated using the QGIS software (QGIS3.4; https://www.qgis.org/en/site/about/index.html).



Supplementary Figure 11. Principal component analysis of the 146 wax gourd accessions. Source data are provided as a Source Data file.



Supplementary Figure 12. Delta K analysis for the number of groups for the 146 wax gourd accessions.



Supplementary Figure 13. Histological and morphology of wax gourd fruit. a.

Morphology of cell size during fruit development Bar = 200um. **b.** Images of typical fruits during development, Bar = 10cm. **c.** Increase of cell volume from accession B214 and B227 from the longitudinal section. Error bars represent \pm sd.



Supplementary Figure 14. Differential expression of SAUR gene in two cucumber near isogenic lines.



Supplementary Figure 15. GWAS results of fruit traits in wax gourd genome.

Library	Library	Data	Read	length	GC	(%)	Q	30
type	insert size	(Gb)	Read1	Read2	Read1	Read2	Read1	Read2
Pair-end	180bp	18.7	100	100	33.3	33.3	97	97
	500bp	1.0	100	100	36.3	36.3	98	92
	500bp	9.6	100	100	36.1	36.2	97	93
Mate-pair	2kb	0.9	91.6	91.4	38.5	38.6	95	93
-	2kb	0.4	100	100	38.4	38.5	96	94
	2kb	9.4	100	100	38.6	38.7	95	96
	8kb	0.4	100	100	37.3	37.4	97	96
	8kb	1.0	93.2	92.9	37.0	36.8	88	85
PacBio		15						
	total	55.4						

Supplementary Table 1. Summary of the sequencing data for wax gourd.

Chromosome	Size of N base (bp)
chr10	947,933
chr11	1,254,718
chr12	1,099,274
chr1	1,359,133
chr2	1,029,499
chr3	1,068,153
chr4	1,109,909
chr5	955,781
chr6	914,502
chr7	907,125
chr8	1,033,663
chr9	1,498,830
contigs	1,600,148
Total	14,778,668

Supplementary Table 2. Summary of gapped regions.

Contig	Size(bp)
Total length	898,172,958
Count	26,315
Average	34,132
Median	19,501
N00	479,328
N10	164,196
N20	126,004
N30	101,266
N40	83,251
N50	68,515
N60	55,048
N70	42,867
N80	30,679
N90	18,560
N100	349

Supplementary Table 3. Summary of contigs.

Repeat sequence type		Copy number	Sequence length (Mb)	Percentage (%)
Dispersed	DNA transposable	310,469	94.06	10.30
ronast	elements			
Tepeat	CMC	18,688	8.56	0.94
	hAT	42,093	13.78	1.51
	Helitron	1,132	0.32	0.04
	MuLE	46,675	19.19	2.10
	PIF	10,903	3.25	0.36
	TcMar	87	0.008	0.001
	other	190,891	48.96	5.36
	RNA transposable	769,754	545.39	59.73
	elements			
	non-LTR RT			
	LINE	57,565	16.20	1.77
	SINE	11,350	1.10	0.12
	LTR RT			
	Copia	376,614	329.41	36.08
	Gypsy	201,768	142.45	15.50
	other	122,457	56.22	6.12
	Unknown transposable	44,510	12.95	1.42
Tandem	elements			
reneat	Microsatellite	34,509	1.48	0.16
repear	Minisatellite	301,262	23.91	2.62
	Satellite	21,343	11.72	1.28

Supplementary Table 4. Summary of predicted repeat sequences in the wax gourd genome.

Туре	Number	Percentage (%)
Total	27,467	100
NR	21,002	76.46
InterPro	17,767	64.68
Swiss-port	20,770	75.62
Arabidopsis	20,480	74.56
GO	17,767	64.68
Annotated	21,227	77.28

Supplementary Table 5. Functional annotation of the wax gourd protein-coding genes.

Species	Number	Full NB - ARC domain
Cucumber	74	43
Melon	84	56
Wax gourd	82	59
Water melon	49	34
Bitter gourd	78	48
Soybean	164	NA
Arabidopsis	251	NA
Grape	330	NA
Tomato	265	NA
Rice	535	NA

Supplementary Table 6. NBS-LRR genes in different genomes.

NA: Not analyzed

	Wax gourd	Cucumber	Melon	Watermelon
Genome size(Mb)	913	196	407	355
Gene number	27,467	24,274	27,427	23,440
Mean gene size(bp)	3,962	3,674	2,776	2,770
Mean CDs size(bp)	1,098	1,149	950	1,109
Mean exon size(bp)	328	276	271	242
Mean exon number/gene	6.6	7.1	4.6	4.6
Mean intron size(bp)	1,044	814	506	654

Supplementary Table 7. Comparisons of gene models between the wax gourd and the other three *Cucurbitaceae* species.

RT_type	Watermelon	Cucumber	Melon	Wax gourd
DNA_RT	6.35	22.07	27.46	94.06
LINE	10.43	10.26	4.52	16.20
SINE	0.01	0.49	0.29	1.10
LTR_Copia	35.06	15.99	38.39	329.41
LTR_Gypsy	20.48	12.53	25.16	142.45
LTR_other	28.97	8.74	3.92	56.22
Unknown	32.74	1.02	26.60	12.95

Supplementary Table 8. Comparisons of the repeats content.

Note: repeat content: Mb.