1 Supplementary Figures



3 Supplementary Figure S1. Comparison of contig N10-N90 between the old and current



4 version of the *Nelumbo nucifera* var. 'China Antique' assembly.



6 Supplementary Figure S2. Chromatin interaction matrix for eight chromosomes from
7 HI-C experiment. Darker color indicates stronger chromatin interaction.



9 Supplementary Figure S3. Distribution of SNP markers on the eight linkage groups (A),

```
10 on the 'China Antique' assembly v2013 (B) and our current version (C).
```



Eudicot genomes in Plant Genome Duplication Database

Supplementary Figure S4. Comparison of the numbers of syntenic ortholog genes in *Brachyopodium distachyon* and *Oryza sativa* in relationship to eudicot assemblies from Plant Genome Duplication Database including two lotus assemblies (v2013 and the current version).





Supplementary Figure S5. Detection of lotus whole genome duplication and its relation to species divergence. A. 4dTv distributions of intra-specific (*Nelumbo*) syntenies and predicted orthologous gene pairs between lotus and *Macadamia*. B. Chronogram showing divergence time and WGDs in lotus and other key taxa.





24



26 groups based on origins and duplication status. A. All lotus genes. B. Lotus orphan genes.



Supplementary Figure S7. 4dTv divergences between the closest paralogs and between
orthologs (*Nelumbo-Amborella*) for dispersed duplicates as 'angiopserm conserved single
copy genes' (Angio-single) and the other dispersed duplicates. A. Violin plot of 4dTv
divergence for orthologs and paralogs. B. Presumed duplication event before split of *Nelumbo*and *Amborella*. C. Presumed duplication event after split of *Nelumbo* and *Amborella*.



Supplementary Figure S8. Differences between expressed dispersed duplicates and silent
dispersed duplicates. A. Mean copy number of orthologs in three related taxa. B. C.V. of
copy number of orthologs in three related taxa. C. Gene length. D. Protein-protein interactions
inferred from Arabidopsis. E. The number of exons. F. CDS length. G. 4dTv between the
closest paralogs in lotus. H. Ratio of deleted sequence in CDS. I. Nucleotide diversity in
CDS.



Supplementary Figure S9. Differences in average CG, CHG and CHH methylation level
(ML) along the gene (on genic and flanking regions) among different gene groups based
on duplication status in lotus leaf, petal, stamen petaloid, and stamen.



Supplementary Figure S10. Differences in average CG, CHG and CHH methylation
level (ML) along the representative transcript (on transcript and flanking regions)
among different gene groups based on duplication status in lotus leaf, petal, stamen
petaloid, and stamen.





Supplementary Figure S11. Differences in TE distribution of genic and flanking regions
among different gene groups (based on duplication status) in lotus. A. sRNA- TEs. B.
sRNA+ TEs.





Supplementary Figure S12. Top 30 enriched Gene Ontology terms of genes from
different gene groups (based on duplication status) in lotus. A. Genes retained from WGD.
B. Dispersed duplicate genes. C. Proximal duplicate genes. D. Genes underwent both WGD
and tandem duplication. E. Tandemly duplicated genes. F. Single copy genes.



Supplementary Figure S13. Average CG, CHG and CHH methylation level (ML) in lotus
leaf, petal, stamen petaloid, and stamen along the gene among duplicate genes that
display different levels of expression divergence (Group A, Group B, Group C and
Group D).



84

Supplementary Figure S14. Average CG, CHG and CHH methylation level (ML) in lotus
leaf, petal, stamen petaloid, and stamen along the representative transcript among
duplicate genes that display different levels of expression divergence (Group A, Group B,
Group C and Group D).



Supplementary Figure S15. Differences in TE distribution of genic and flanking regions
among WGD duplicates displaying different levels of expression divergence (Group A,
Group B, Group C and Group D). A. sRNA- TEs. B. sRNA+ TEs.



Supplementary Figure S16. Violin plots of *r* (correlation coefficient) for CHG and CHH
methylation on gene body, gene upstream (-2kb) and downstream (+2kb) in different
tissues between WGD-derived duplicate genes with different level of expression
divergence (Group A, Group B, Group C and Group D). A-C: CHG. D-F: CHH. Black
line: median; grey line: quantile.



Supplementary Figure S17. Top 30 Gene Ontology terms enriched in lotus WGD
duplicates that display different levels of expression divergence (Group A, Group B,

104 **Group C and Group D).** A. Group A. B. Group B. C. Group C. D. Group D.



Supplementary Figure S18. Grouping of *Arabidopsis* syntenies into three presumed
WGDs, At-α, At-β and At-γ by pairwise *t*-test based on *Ks* values.



108

109 Supplementary Figure S19. Top 30 Gene Ontology terms enriched in At-a WGD

110 duplicates that display different levels of expression divergence (Group A, Group B,

111 Group C and Group D). Note that GO terms with more than 10,000 annotated genes, such

as cellular process, are not shown. A. Group A. B. Group B. C. Group C. D. Group D.





114 Supplementary Figure S20. Top 30 Gene Ontology terms enriched in At- β WGD

115 duplicates that display different levels of expression divergence (Group A, Group B,

116 Group C and Group D). Note that GO terms with more than 10,000 annotated genes, such

117 as cellular process, are not shown. A. Group A. B. Group B. C. Group C. D. Group D.

118



120 Supplementary Figure S21. Distinct expression patterns of lotus *CRY1a,b* and *CRY2a,b*.



Supplementary Figure S22. Differences in average CG, CHG and CHH methylation
level (ML) in lotus leaf, petal, stamen petaloid, and stamen along gene (on genic and
flanking regions) between duplicates that belong to less fractionated blocks (LFs) and
more fractionated blocks (MFs).



Supplementary Figure S23. Differences in average CG, CHG and CHH methylation
level (ML) in lotus leaf, petal, stamen petaloid, and stamen along the representative

- 131 transcript (on transcript and flanking regions) between duplicates that belong to less
 132 fractionated blocks (LFs) and more fractionated blocks (MFs).
- 133



Supplementary Figure S24. Differences in average TE distribution along gene (on genic
and flanking regions) between duplicates that belong to less fractionated blocks (LFs)
and more fractionated blocks (MFs). A. sRNA- TEs. B. sRNA+ TEs.



140 Supplementary Figure S25. Differences in average CG methylation level (ML) in lotus

leaf, petal, stamen petaloid, and stamen along gene (on genic and flanking regions)
between duplicates that belong to less fractionated blocks (LFs) and more fractionated
blocks (MFs) in Group A, Group B, Group C and Group D.



Supplementary Figure S26. Differences in average CHG methylation level (ML) in lotus
leaf, petal, stamen petaloid, and stamen along gene (on genic and flanking regions)
between duplicates that belong to less fractionated blocks (LFs) and more fractionated
blocks (MFs) in Group A, Group B, Group C and Group D.



Supplementary Figure S27. Differences in average CHH methylation level (ML) in lotus
leaf, petal, stamen petaloid, and stamen along gene (on genic and flanking regions)
between duplicates that belong to less fractionated blocks (LFs) and more fractionated
blocks (MFs) in Group A, Group B, Group C and Group D.



159

Supplementary Figure S28. Differences in average CG methylation level (ML) in lotus leaf, petal, stamen petaloid, and stamen along the representative transcript (on transcript and flanking regions) between duplicates that belong to less fractionated blocks (LFs) and more fractionated blocks (MFs) in Group A, Group B, Group C and Group D.





Supplementary Figure S29. Differences in average CHG methylation level (ML) in lotus leaf, petal, stamen petaloid, and stamen along the representative transcript (on transcript and flanking regions) between duplicates that belong to less fractionated blocks (LFs) and more fractionated blocks (MFs) in Group A, Group B, Group C and Group D.



Supplementary Figure S30. Differences in average CHH methylation level (ML) in lotus
leaf, petal, stamen petaloid, and stamen along the representative transcript (on
transcript and flanking regions) between duplicates that belong to less fractionated
blocks (LFs) and more fractionated blocks (MFs) in Group A, Group B, Group C and
Group D.