***New Phytologist* Supporting Information**

Article title: Divergence of duplicated genes by repeated partitioning of splice forms and subcellular localization.

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The following Supporting Information is available for this article:

Fig. S1. Scheme for primer design.

Fig. S2. The translated sequences of the last exon of alternatively spliced and specialized tAPX showing the conserved hydrophobic thylakoid-anchorage chain.

Fig. S3. Case studies of sub-localized duplicated *cpAPX* genes.

Fig. S4. The evolution of *cpAPX* genes in Solanaceae.

Table S1. CpAPX genes examined in this study (submitted separately).

Table S2. Plant materials in this study (submitted separately).

Table S3. Primers designed for this study (submitted separately).

Table S4. The selection analyses of cpAPX genes (submitted separately).

Fig. S1. Scheme for primer design. Alternatively-spliced single copy *cpAPX*, specialized *sAPX* (Type I and II) and specialized *tAPX* are aligned with only the last three exons shown by boxes. Blue and green squares are splicing acceptors corresponding to sAPX and tAPX transcripts, respectively. Red bars are stop codons. Striped bars represent the hydrophobic tail coding sequences. The forward (F) and reverse (R) primers (arrows) are designed outside of the orange bars to distinguish the coding potential for tAPX and sAPX.

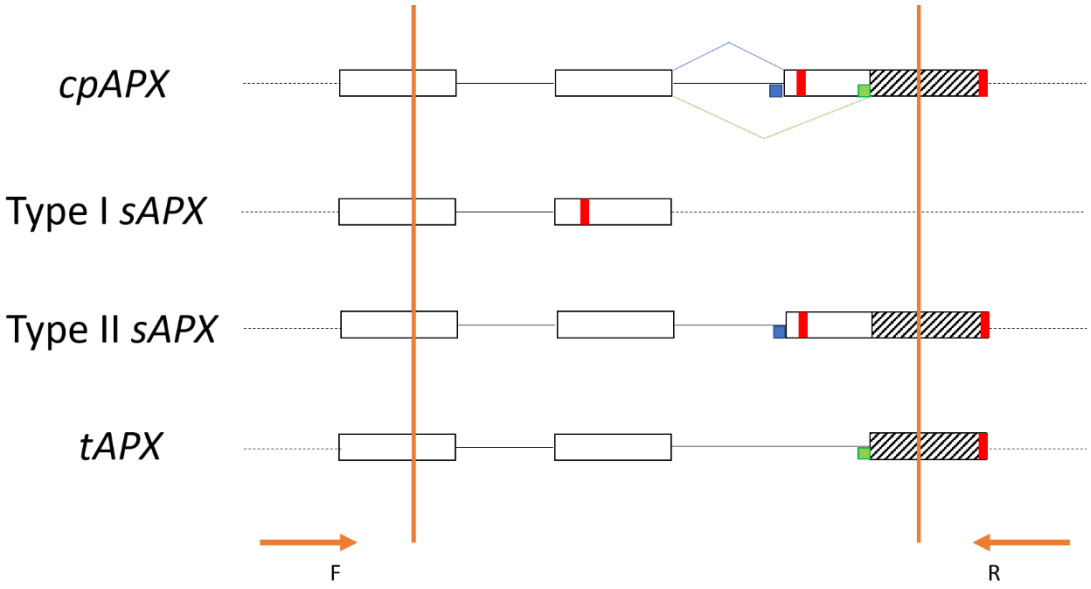


Fig. S2. The translated sequences of the last exon of alternatively spliced (blue) and specialized (red) tAPX showing the conserved hydrophobic thylakoid-anchorage chain. The green triangle points to the spinach tAPX which was purified from the thylakoidal membrane by Miyake and Asada (1992).

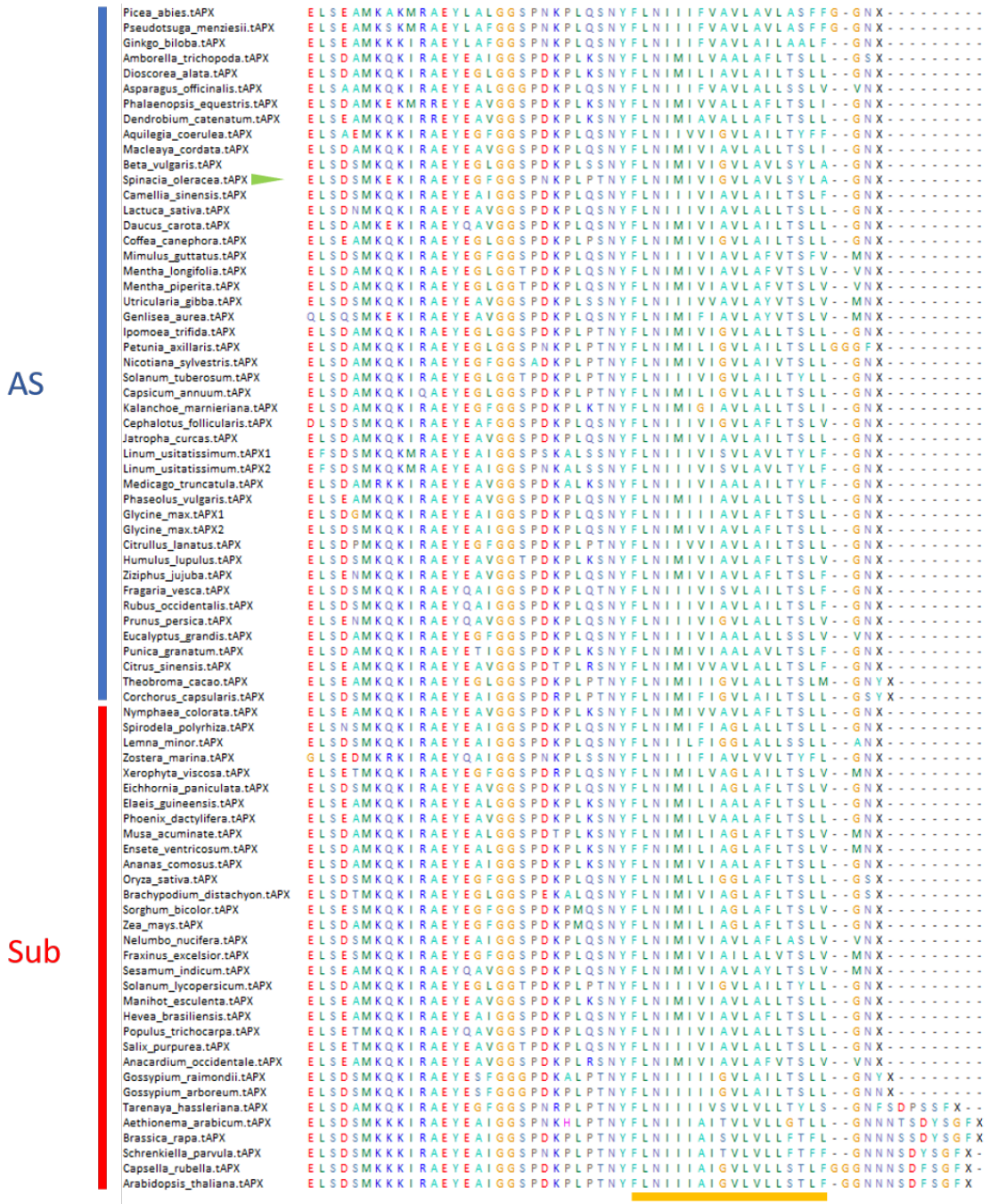
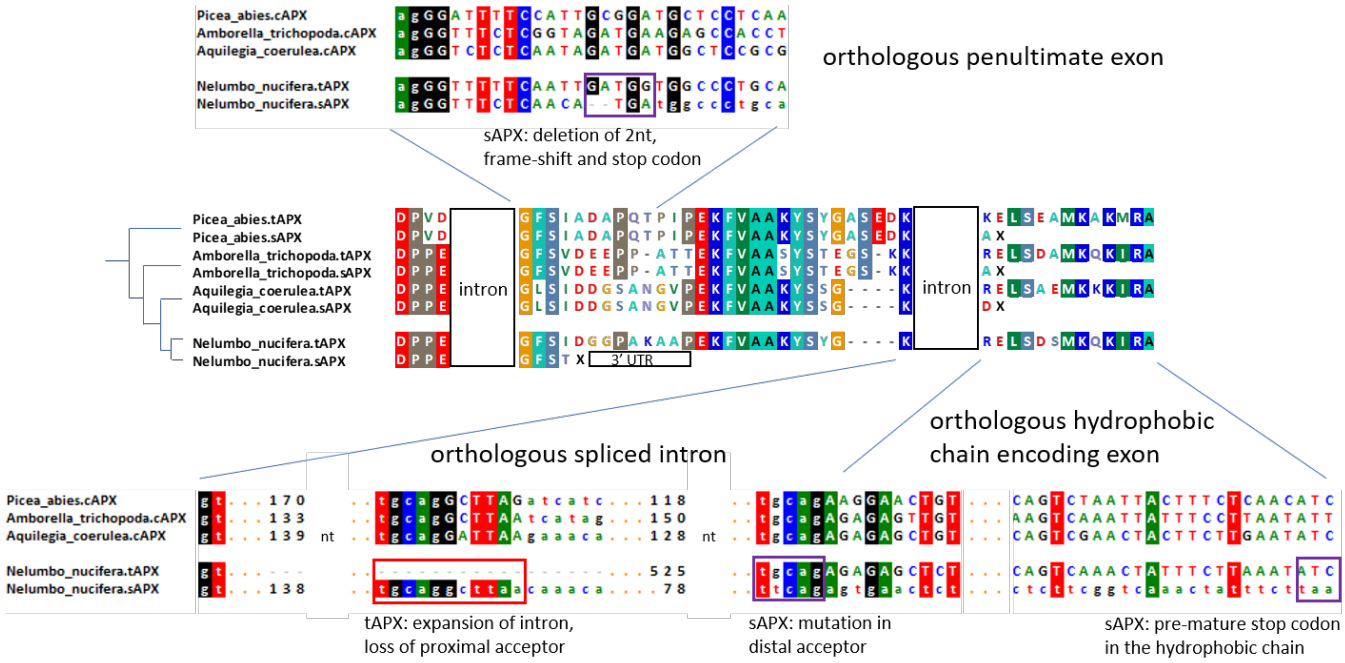
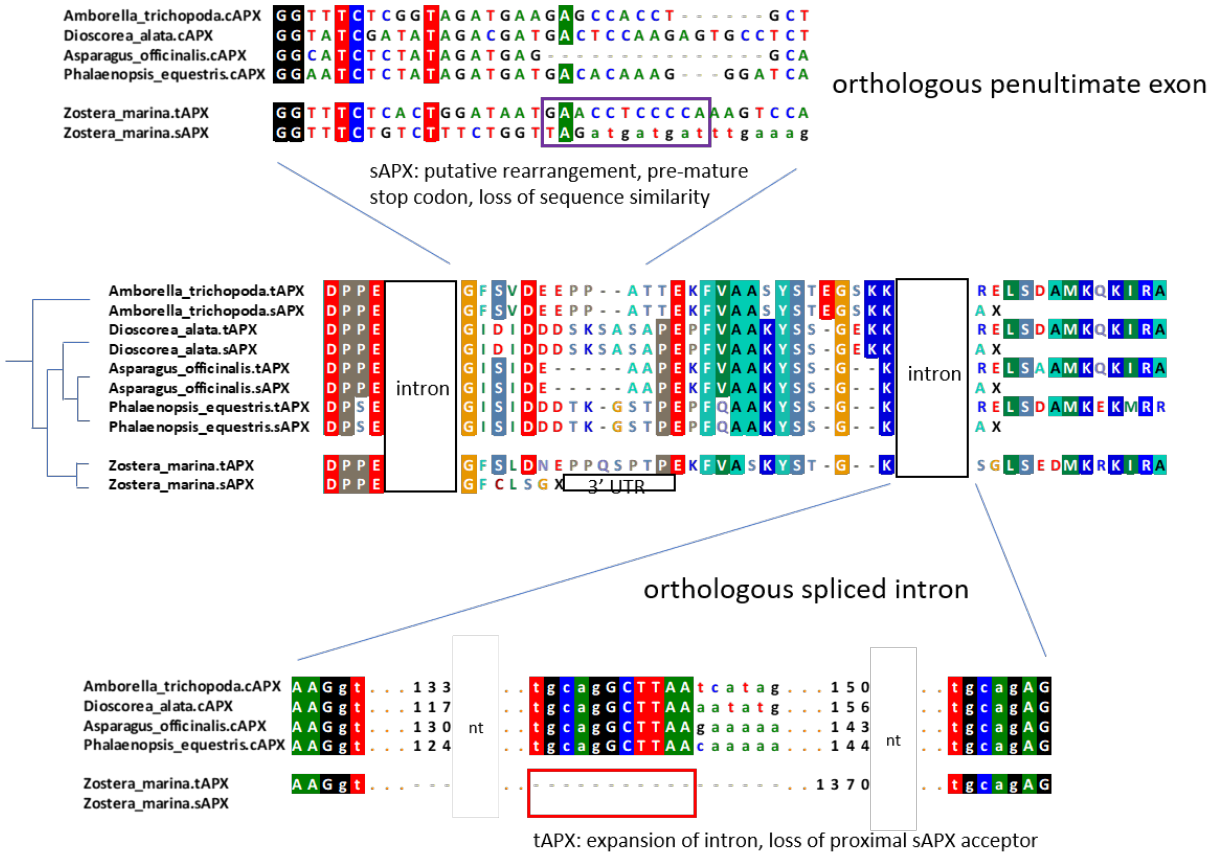


Fig. S3. Case studies of sub-localized duplicated *cpAPX* genes. Amino acid and nucleotide sequences of sub-localized paralogous are aligned with several single-copy orthologs. Red boxes and purple boxes indicate the nucleotide sequence changes contributing to the tAPX specialization and sAPX, respectively, and a short description is indicated beside the boxes. (a) *Nelumbo nucifera*; (b) *Zostera marina*; (c) Musaceae.

a



b


Amborella_trichopoda.cAPX
Dioscorea_alata.cAPX
Asparagus_officinalis.cAPX
Phalaenopsis_equestris.cAPX
Zostera_marina.tAPX
Zostera_marina.sAPX
orthologous penultimate exon
sAPX: putative rearrangement, pre-mature stop codon, loss of sequence similarity
Amborella_trichopoda.tAPX
Amborella_trichopoda.sAPX
Dioscorea_alata.tAPX
Dioscorea_alata.sAPX
Asparagus_officinalis.tAPX
Asparagus_officinalis.sAPX
Phalaenopsis_equestris.tAPX
Phalaenopsis_equestris.sAPX
intron
intron
3' UTR
orthologous spliced intron
nt
nt
tAPX: expansion of intron, loss of proximal sAPX acceptor

c

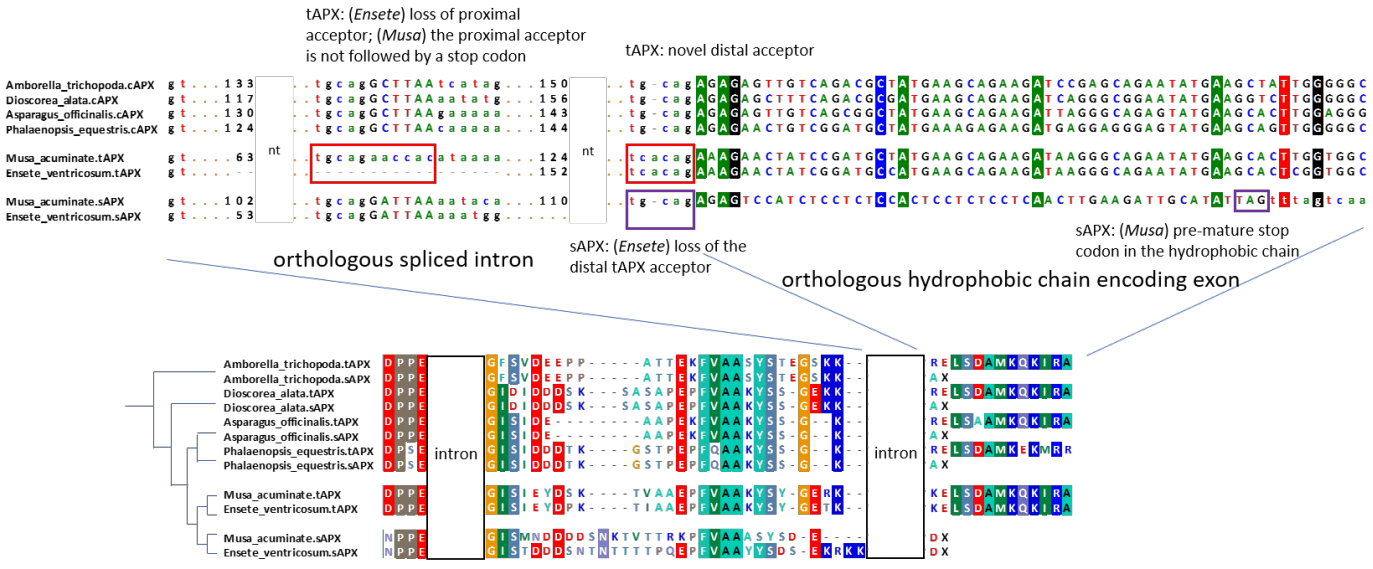
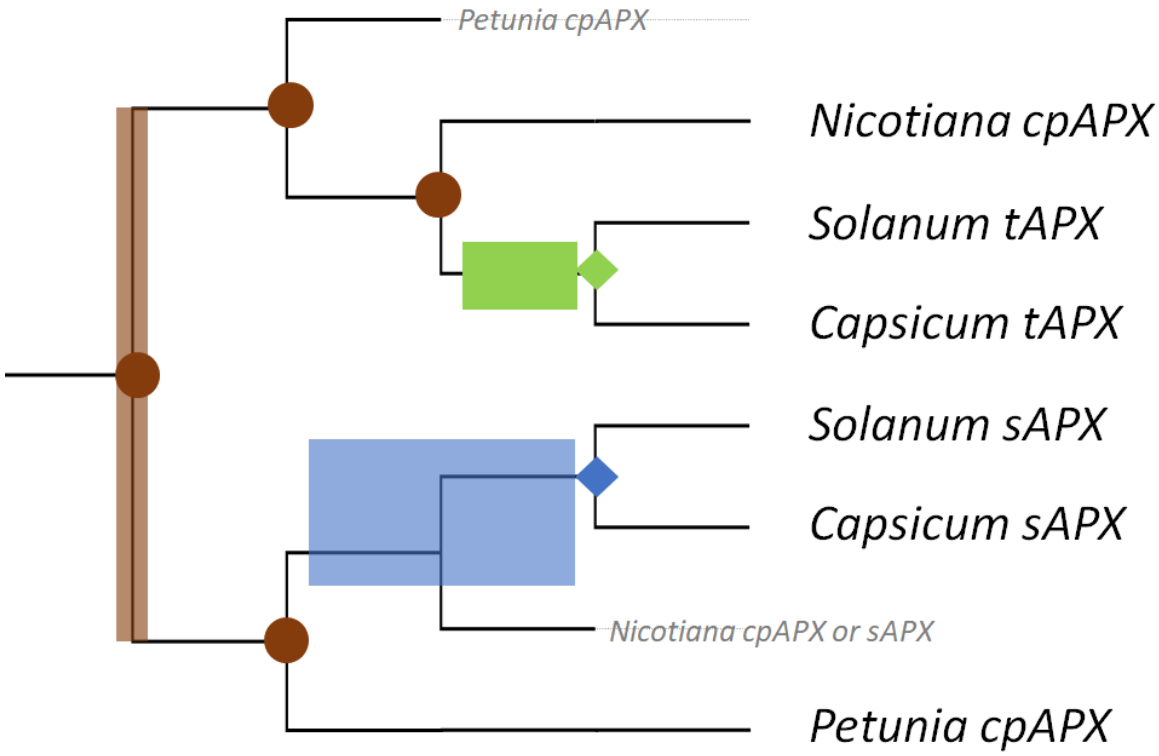

tAPX: (Ensete) loss of proximal acceptor; (Musa) the proximal acceptor is not followed by a stop codon
tAPX: novel distal acceptor
Amborella_trichopoda.cAPX
Dioscorea_alata.cAPX
Asparagus_officinalis.cAPX
Phalaenopsis_equestris.cAPX
Musa_acuminate.tAPX
Ensete_ventricosum.tAPX
Musa_acuminate.sAPX
Ensete_ventricosum.sAPX
nt
nt
sAPX: (Musa) pre-mature stop codon in the hydrophobic chain
orthologous spliced intron
sAPX: (Ensete) loss of the distal tAPX acceptor
orthologous hydrophobic chain encoding exon
Amborella_trichopoda.tAPX
Amborella_trichopoda.sAPX
Dioscorea_alata.tAPX
Dioscorea_alata.sAPX
Asparagus_officinalis.tAPX
Asparagus_officinalis.sAPX
Phalaenopsis_equestris.tAPX
Phalaenopsis_equestris.sAPX
Musa_acuminate.tAPX
Ensete_ventricosum.tAPX
Musa_acuminate.sAPX
Ensete_ventricosum.sAPX
intron
intron

Fig. S4. The evolution of *cpAPX* genes in Solanaceae. The genes in black are in the extant genomes. The genes in grey were presumably present in the ancestral genome and were lost. Brown bar indicates gene duplication. Brown circles indicate ancestral alternatively spliced genes. Green and blue boxes indicate possible timing when the inferred gene function specialization to *tAPX* and *sAPX* took place. Green and blue diamonds indicate the ancestral *tAPX* and *sAPX* gene before the divergence of *Solanum* and *Capsicum* from a common ancestor.



References:

Miyake C, Asada K. 1992. Thylakoid-bound ascorbate peroxidase in spinach chloroplasts and photoreduction of its primary product monodehydroascorbate radicals in thylakoids. Plant Cell Physiology **33**(5):541–553.