

Identification of Genes Regulating Leaf Expansion and Traits Development of *Spathiphyllum* Plant Based on Full-length Transcriptome Sequencing

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Spathiphyllum is a perennial herb plant belonging to the family Araceae and popular ornamental tropical plant around the world. As a foliage plant the leaves are the main parts above-ground of this herb plant and also are the organs for ornamental. However, the molecular mechanism of leaf development and expanding regulation of *Spathiphyllum* is still limited. In this study *Spathiphyllum* ‘Mojo’ and one of its somatic mutant lines (Ssm-1) with highly significant differences of leaf morphological traits were used as experimental materials. The full-length transcriptome sequence of leaves in the Pre-growth stage (S1), Fast-growth stage (S2), and Post-growth stage (S3) were performed on the PacBio platform, and the obtained transcripts were annotated with Nr, Swissprot, GO, COG, KOG, Pfam, and KEGG databases. A combination of bioinformatics analysis and RT-qPCR techniques was used to mine key genes and related metabolic pathways regulating leaf development and expansion of *Spathiphyllum*. Expression of genes was determined by RT-qPCR using a combination of *TUB* 和 *GAPDH* as reference genes. Total RNA was

extracted using TIANGEN RNAPrep Pure Plant Plus Kit. The Evo M-MLV RT Mix Kit with gDNA Clean for qPCR kit was used to reverse transcribed RNA into cDNA. RT-qPCR amplification was performed using Bio-Rad CFX Connect fluorescence quantitative PCR. SYBR Green Premix Pro Taq HS qPCR Kit was used. Weighted correlation network analysis was used to evaluate the relation between gene module and leaf traits. The experiments were repeated three times.

Compared with parental *S. 'Mojo'* somatic mutant line Ssm-1 showed great changes in leaf morphology with highly significant differences in leaf length, leaf width, leaf shape index, leaf thickness, leaf area, and SPAD.

Transcriptional sequencing yielded 44,976 unigene sequences, and 95.38% of which could be annotated in Nr, Swissprot, GO, COG, KOG, Pfam, and KEGG databases. Differential expression genes of 3902 were analyzed in three growth logistics stages of leaf between somatic mutant and parental *S. 'Mojo'*.

Bioinformatic analysis of leaf transcriptional sequences uncovered 45 differential expression genes related to plant hormone signal transduction and 179 differential expression transcription factors which distributed in 38 families. *SpTCP2*, *SpTCP3*, *SpTCP4*, *SpTCP10*, *SpTCP14*, *SpTCP24*, *SpGRF1*, *SpGRF2*, *SpGRF5*, *SpGRF7*, and *SpGAI*,

were identified to regulate leaf development and expansion of *Spathiphyllum* (Figure 1) .

Total 34 gene family members *SpTCP1-SpTCP34* encoding TCP were found in the transcriptome of *Spathiphyllum* leaves, and could be classified into two subfamilies: Class I and Class II combined with the phylogenetic relationship results. *SpTCP2*, *SpTCP4*, *SpTCP18*, *SpTCP26*, and *SpTCP33* were highly expressed in the three stages of the leaf development but were found to have different expression patterns between the somatic mutation line and parental ‘Mojo’ of *Spathiphyllum*, which means these genes could regulate the leaf development and expansion of *Spathiphyllum* with different expression(Figure2).

Weighted correlation network analysis released that the module brown was highly correlated with the leaf length of *Spathiphyllum*, while the genes *KNOX5*, *bHLH49*, *bHLH78*, *AXF4*, thioredoxin M-type, and *bHLH129* were the hub genes in the module brown.

Keywords: *Spathiphyllum*; Foliage plant; Leaf development;
Full-length Transcriptome sequence;

Acknowledgement: This study was Fund by the Guangzhou municipal Science and Technology Project (202002020025)

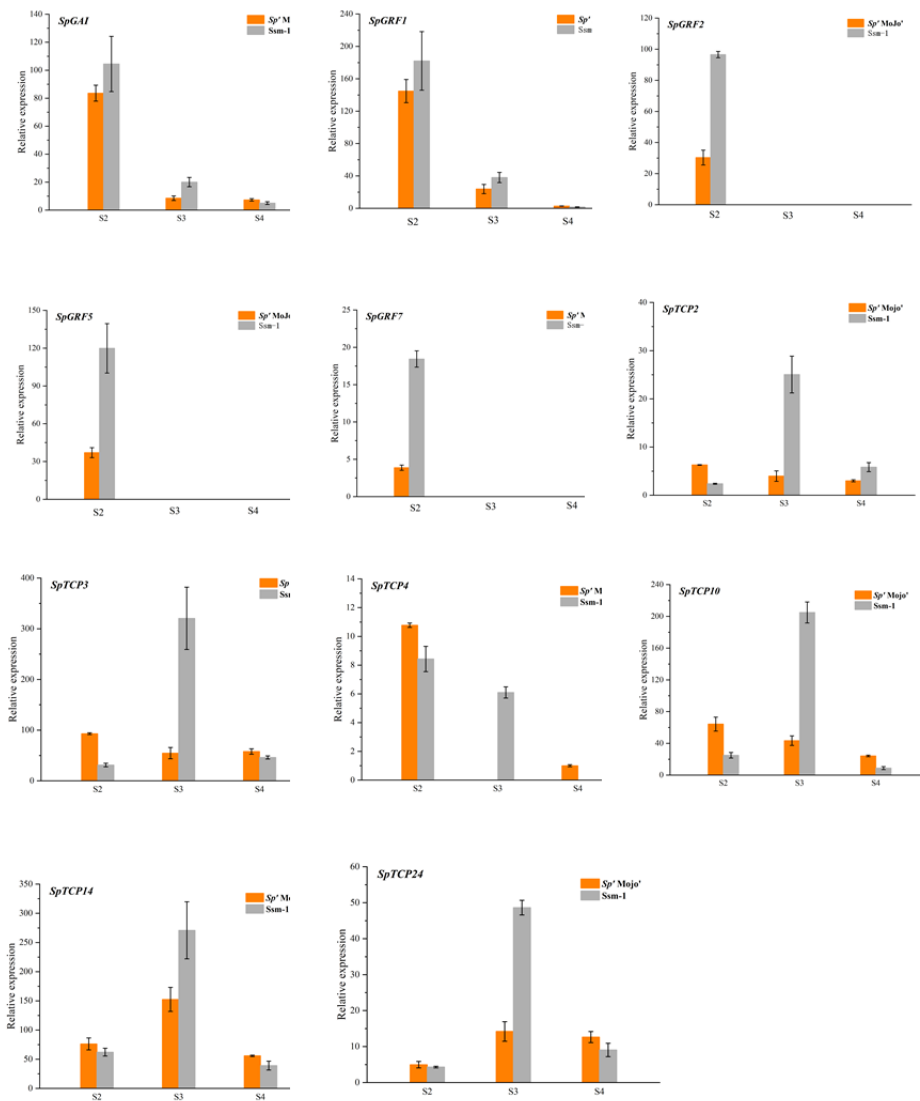


Figure1 The expression of transcript factors related the leaf development screened in parental 'Mojo' and mutant line Ssm-1 at different development stage

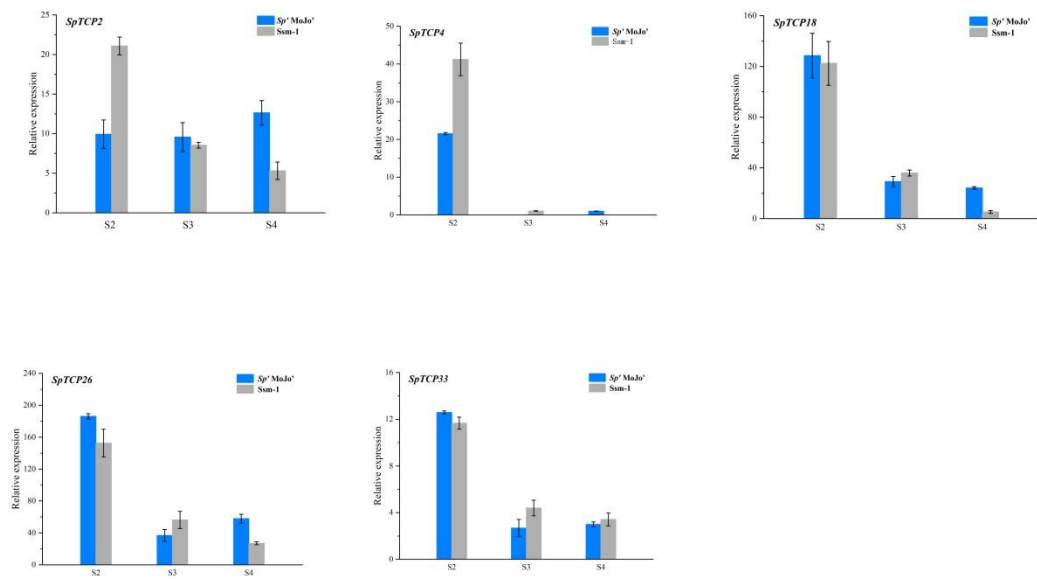


Figure 2 SpTCP family expressed in the three stages of the leaf development with different expression patterns between the somatic mutation line and parental ‘Mojo’ of *Spathiphyllum*.