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Full Length Article

Comparative Transcriptomic Analysis Reveals Carotenoids Biosynthesis Genes in Peach

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Abstract

Carotenoids are important substances for the yellow color of peach flesh. In this study, the contents of carotenoids in two yellow flesh peach varieties ('Zhongtao Jinmi' and 'Jinxiang') and one white flesh peach variety ('Bairuyu') were determined by high-performance liquid chromatography. The results showed that the carotenoid content in the two yellow flesh peach varieties increased with fruit development and reached the maximum of 25.80 and 27.63 μg.g^{-1} at the mature stage, while that of the white flesh peach variety remained at a very low level during the whole fruit development period. Furthermore, transcriptome sequencing was performed to analyze the differentially expressed genes (DEGs) during fruit development among the three peach varieties. Among these DEGs, four candidate genes were identified to be potentially involved in the biosynthesis of carotenoids, including *LOC18774744* (encoding UDP-glycosyltransferase 87A1 (UGT87A1)), *LOC18792568* (encoding the carotenoid cleavage dioxygenase 4 gene (CCD4)), *LOC109949966* (an unannotated gene), and *LOC18779468* (encoding xyloglucan endoglucosylase/hydrolase protein 8. qRT-PCR confirmed that the expression of *LOC109949966* and *LOC18779468* in two yellow flesh varieties was significantly higher than that in the white flesh variety, while it was the opposite case for *UGT87A1* and *CCD4*. Therefore, we speculate that *CCD4* is directly involved in regulating carotenoid synthesis, while other three genes may play some indirect roles in the process. Our findings are expected to improve the understanding on the mechanism of carotenoid biosynthesis in peach.

Keywords: Peach; Carotenoids; Transcriptome sequencing; Differentially expressed genes

Introduction

Peach *(Prunus persica* L.), a perennial woody plant of Rosaceae, *Prunus* L. and *Amygdalus* L., is the third largest deciduous fruit tree in China (Adami *et al*. 2013). It has a long cultivation history and rich germplasm resources. At present, in the description specifications and data standards of peach germplasm resources, peach fruit can be divided into white flesh, yellow flesh and red flesh varieties (Wen *et al*. 2020). Yellow flesh peach contains a large number of carotenoids and more nutrients than white flesh peach; besides, it has rich and unique flavor and the flesh is characterized by strong anti-browning and anti-oxidation ability (Brandi *et al*. 2011; Falchi *et al*. 2013; Fiedor and Burda 2014). The major carotenoid in yellow flesh peach fruit is β-carotene, followed by xanthophyll purple xanthin, antheraxanthin and zeaxanthin (Kato *et al*. 2004; Centeno *et al*. 2011; Barsan *et al*. 2012; Zhang *et al*. 2013). Modern medical research has shown that these substances are important biologically active ingredients required for human health, particularly β-carotene, which is the most effective precursor substance for human body to synthesize vitamin A (Fraser *et al*. 2007). Due to these advantages, yellow peach is becoming increasing popular among consumers. Therefore, investigation of carotenoid accumulation in the fruit flesh and dissection of the molecular mechanism will help to understand the regulatory mechanism of carotenoid accumulation in yellow peach fruit and provide a scientific basis for the actual production and breeding.

As a kind of terpenoids widely present in nature (Hirschberg 2001), carotenoids are a major type of coloring pigments related to most of the yellow to red colors in plants such as fruits, vegetables and flowers (Khoo *et al*. 2011). In recent years, with the development of molecular biological technologies, the genes related to the carotenoid biosynthetic pathway have been successively isolated and identified from

plants. Geranyl pyrophosphate (GGPP), which is synthesized by geranyl pyrophosphate synthase, is a precursor substance for carotenoid synthesis (Armstrong and Hearst 1996). The number of *PPGG* genes varies among different plants. *Arabidopsis* has the largest number (12 in total) of *PPGG* homologous genes in all plants (Coman *et al.* 2014). Phytoene synthase is the rate-limiting enzyme in carotenoid biosynthesis and metabolism pathway, which is encoded by the *PSY* gene. *PSY* can regulate the flux of precursor metabolites in the biosynthesis pathway, thereby affecting the accumulation of carotenoids in fruits (Zhang *et al.* 2009). Some other genes that are critical to carotenoid biosynthesis have also been cloned in plants, such as *DXS*, *PDS*, *ZDS*, *ZISO*, *CRTISO*, *β-LCY*, *ε-LCY*, *BCH*, *ZEP* and *VDE* (Lois *et al*. 2000; Deng *et al*. 2003; Kato *et al*. 2004; Lou *et al*. 2017). Clarification of the relationship between carotenoid biosynthesis and the formation of fruit flesh color may help to increase the carotenoid content in fruits. However, there have been few reports about the effect of carotenoid accumulation on peach flesh color and the related mechanism so far.

In order to explore the law of carotenoid synthesis during peach fruit development, transcriptome sequencing was carried out to analyze the differentially expressed genes (DEGs) during the development of yellow and white flesh peach fruit, and the regulatory genes of the carotenoid biosynthesis pathway were identified, including four candidate genes that are directly (*CCD4*) or indirectly (*LOC18774744*, *LOC109949966* and *LOC18779468*) related to yellow peach carotenoid synthesis.

Materials and Methods

Plant Materials

The fruit samples were collected from three peach varieties, including 'Jinxiang' (yellow flesh), 'Zhongtao Jinmi' (yellow flesh) and 'Bai Ruyu' (white flesh) provided by the Horticultural Research Institute of Sichuan Academy of Agricultural Sciences. All peach trees were 5 years old. The samples were collected at three stages: young fruit stage (S1, 60 d after flowering), color transformation stage (S2, 85 d after flowering), and mature period (S3; 'Jinxiang': 95 d after flowering; 'Bai Ruyu' and 'Zhongtao Jinmi':102 d after flowering). Three fruit trees were collected from each variety in each period, and each tree in each period collected 2 fruits in four directions: east, south, west and north and each varieties had a total of 24 fruits in each period. The peach flesh was collected, frozen immediately in liquid nitrogen and then stored at −80°C until RNA preparation. Three biological replicates were prepared for each period.

Determination of Carotenoids Content

High-performance liquid chromatography (HPLC) was used to analyze the carotenoids in peach flesh according to the method previously described by Brandi *et al*. (2011).

RNA Extraction, cDNA Synthesis and Transcriptome Sequencing

RNA extraction was conducted with the TaKaRa MiniBEST Plant RNA Extraction Kit (TaKaRa, https://www.takarabiomed.com.cn/). By using the TaKaRa's PrimeScript RT Agent kit with gDNA Eraser (TaKaRa, https://www.takarabiomed.com.cn/), cDNA was synthesized from total RNA. Transcriptome sequencing was performed by the Illumina2 HiSeq^{TM} 4000 sequencing platform and quality control on the raw reads was carried out after sequencing. After removal of low-quality data, clean reads were obtained and then aligned to the peach reference genome using the Bowtie2 software.

Transcriptome Data Analysis

The gene expression was expressed as RPKM (reads per kilobase of exon model per million mapped reads). DEseq2 software was used to identify the differentially expressed genes (DEGs) at different stages with the screening criteria of $|\log2\ Ratio| \geq 1$ and $FDR < 0.05$.

qRT-PCR Analysis

According to the instructions of SYBR® Premix Ex Taq (Takara, https://www.takarabiomed.com.cn/), qRT-PCR was performed with the CFX96 real-time quantitative PCR system (Bio-Rad, https://www.bio-rad.com/). *PpMUB6* was adopted as the housekeeping gene. The relative expression levels of the genes were calculated using the $2^{-\triangle\triangle Ct}$ method (Schmittgen and Livak 2008). The oligonucleotide primers used in this study are shown in Table 1.

Results

Accumulation of Carotenoids in Yellow Flesh Peach Fruit

In this study, 'Zhongtao Jinmi' and 'Jinxiang' were representative varieties of yellow flesh peach and 'Bai Ruyu' was the representative of white flesh peach. Fig. 1 shows the changes of carotenoid content in the flesh of the three varieties at different stages of fruit development. The two yellow flesh peach varieties showed a similar gradual upward trend in carotenoid content at the three stages. The young fruit had the lowest carotenoid content ('Zhongtao Jinmi': 5.06 $\mu g.g^{-1}$; 'Jinxiang': 8.24 $\mu g.g^{-1}$), while the mature fruit had the highest carotenoid content ('Zhongtao Jinmi': 25.80 $\mu g.g^{-1}$; 'Jinxiang': 27.63 $\mu g.g^{-1}$). Nevertheless, the white flesh peach variety 'Bai Ruyu' consistently showed extremely low carotenoid content in the flesh at the three stages. These results suggested that compared with white flesh peach, yellow flesh peach has a higher accumulation of carotenoids.

Table 1: Oligonucleotide primers used in this study

Primer name	Sequence (5'-3')
PpMUB6-F	AAGATACTGGAAAACAACAGGACC
PpMUB6-R	CAATAGGAGGACGCACAACC
LOC18774744-F	AAACCCAAGTCCTTGACGTCT
LOC18774744-R	CACAGAGCTACAAGGTTGAGAATC
LOC18792568-F	CCTACCACCTGTTTGACGGA
LOC18792568-R	AGCCAGCATCACGCTCAAT
LOC109949966-F	AACTAAGTCCTCCACGAACGC
LOC109949966-R	ATAAGGGCCATGAGAAATCTGA
LOC18779468-F	TGCGTCTCCACCACAACAA
LOC18779468-R	TGGCAAAGTTGGGTAGCGT

Table 2: Statistical results of basic transcriptome data of three peach varieties

Samples	Raw reads	Clean reads	Clean reads mapped to genome	Detected gene number
BY-S1-1	48532186	21495922	19868638	25959
BY-S1-2	43520810	21697164	20510935	25959
BY-S1-3	55762432	48621496	47115336	25959
BY-S2-1	47543072	42433038	40968044	25959
BY-S2-2	51250778	47457578	46056774	25959
BY-S2-3	51017008	46479246	44968358	25959
BY-S3-1	41313428	38849542	37621685	25959
BY-S3-2	38967020	31843250	30583374	25959
BY-S3-3	39145522	34559506	33413177	25959
JM-S1-1	52256932	14834108	12781307	25959
JM-S1-2	54352736	32240642	29924509	25959
JM-S1-3	44341136	37426040	35642226	25959
JM-S2-1	46445350	43142004	41864947	25959
JM-S2-2	50245976	34518958	32521905	25959
JM-S2-3	62995308	54663536	52594296	25959
JM-S3-1	49335438	41279942	39672680	25959
JM-S3-2	42687652	35477218	34118028	25959
JM-S3-3	38509870	31857778	30564397	25959
JX-S1-1	58848898	48825278	47072529	25959
JX-S1-2	47927838	33762206	32407289	25959
JX-S1-3	57330042	40688982	36685142	25959
JX-S2-1	53127454	46518446	45291355	25959
JX-S2-2	52324882	36323708	34724307	25959
JX-S2-3	48431850	42442702	41425852	25959
JX-S3-1	41796406	29594416	28207471	25959
JX-S3-2	44725372	29878622	27875826	25959
JX-S3-3	48048502	24870372	23028998	25959

JM, Zhongtao Jinmi (yellow flesh); JX, Jinxiang (yellow flesh); BY, Bai Ruyu (white flesh). S1, young fruit stage; S2, color transformation stage; S3, mature stage. Three biological replicates were prepared for each stage.

Table 3: Differentially expressed genes in 'Zhongtao Jinmi' and 'Bai Ruyu' at different stages

Comparison group	Up-regulation	Down-regulation	Total
JM-S1_vs_BY-S1	200	399	599
JM-S2 vs_BY-S2	431	738	1169
JM-S3_vs_BY-S3	504	647	1151

JM, Zhongtao Jinmi (yellow flesh); BY, Bai Ruyu (white flesh). S1, young fruit stage; S2, color transformation stage; S3, mature stage

Illumina Sequencing and Transcriptome Data

The Illumina platform was used to identify the DEGs between yellow flesh ('Zhongtao Jinmi' and 'Jinxiang') and white flesh peach ('Bai Ruyu') at the three fruit development stages with three biological replicates for each stage. As a result, a total of 3.851×10^7 ~ 6.300×10^7 raw reads were generated from 27 libraries and after the removal of low-quality reads, a total of 1.483×10^7 ~ 5.466×10^7 clean reads were obtained. Then, the Bowtie 2 software was used to compare the clean reads to the peach reference genome and finally 25959 genes were detected (Table 2).

Identification of Differentially Expressed Genes

In this study, (Log2 ratio) ≥1 and FDR < 0.05 were used as thresholds to identify DEGs and the genes were considered as DEGs only when they were detected in all three biological replicates. Table 3 shows that between 'Zhongtao Jinmi' and 'Bai Ruyu', 599 DEGs were identified at the young fruit stage (200 up-regulated and 399 down-regulated); 1169 DEGs were found at the color transformation stage (431 up-regulated and 738 down-regulated) and a total of 1151 DEGs were detected at the fruit mature stage (504 up-regulated and 647 down-regulated). Table 4 shows that between 'Jinxiang'

Table 4: Differentially expressed genes in 'Jinxiang' and 'Bai Ruyu' at different stages

Comparison group	Up-regulation	Down-regulation	Total
BY-S1_vs_JX-S1	322	217	539
BY-S2_vs_JX-S2	1064	1576	2640
BY-S3_vs_JX-S3	982	2455	3437

JX, Jinxiang (yellow flesh); BY, Bai Ruyu (white flesh). S1, young fruit stage; S2, color transformation stage; S3, mature stage

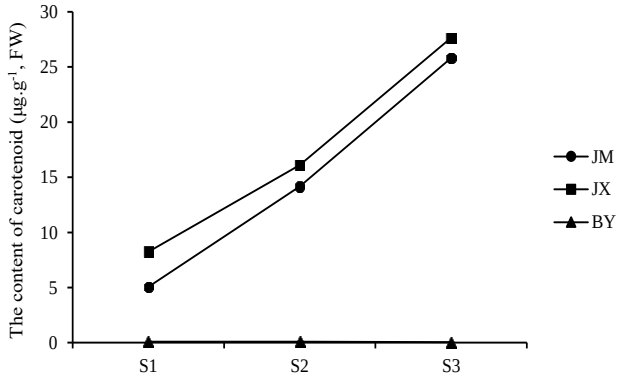

Fig. 1: Changes of carotenoid content in the flesh of three peach varieties at three stages

JM, Zhongtao Jinmi (yellow flesh); JX, Jinxiang (yellow flesh); BY, Bai Ruyu (white flesh). S1, young fruit stage; S2, color transformation stage; S3, mature stage

and 'Bai Ruyu', 539 DEGs were identified at the young fruit stage (322 up-regulated and 217 down-regulated); 2640 DEGs were found at the color transformation stage (1064 up-regulated and 1576 down-regulated); and 3437 DEGs were found at the fruit mature stage (982 up-regulated and 2455 down-regulated).

Gene Annotation

GO provides three ontologies (cellular components, biological processes, and molecular functions) to analyze genes (Conesa *et al*. 2005). At the young fruit stage of 'Zhongtao Jinmi' and 'Bai Ruyu', the DEGs were annotated into 94 GO functional categories. Among them, 39 categories belonged to 'biological processes', with 'metabolic process' showing the highest degree of enrichment; 26 categories were classified into 'cellular components', with 'cytoplasm' exhibiting the highest degree of enrichment and 29 categories belonged to 'molecular functions' and 'catalytic activity' was the most enriched (Fig. 2). At the stage of color transformation of 'Zhongtao Jinmi' and 'Bai Ruyu', the DEGs were annotated into 179 GO functional categories. Among them, 107 categories belonged to 'biological process', with 'stimulus response' exhibiting the highest degree of enrichment; 34 categories were classified into 'cellular components' and 'cytoplasm' was the most enriched; 37 categories belonged to 'molecular functions', with 'catalytic activity' being the most enriched (Fig. 3). At the mature stage of 'Zhongtao Jinmi' and 'Bai Ruyu', the DEGs were annotated into 119 GO categories. Among them, 53 categories belonged to 'biological process' and 'stimulus response' was the most enriched; 32 categories were classified into 'cellular components', with 'membrane' showing the highest degree of enrichment and 34 categories belonged to 'molecular functions', and 'catalytic activity' was the most enriched (Fig. 4).

At the young fruit stage of 'Jinxiang' and 'Bai Ruyu', the DEGs were annotated into 104 GO categories. Among them, 55 categories belonged to 'biological process', in which the 'membrane' was the most enriched; 24 categories belonged to 'cellular components', and the highest enrichment was found for 'organelle subcompartment'; 25 categories fell into 'molecular functions', and the 'intrinsic component of membrane' was the most enriched (Fig. 5). At the stage of color transformation of 'Jinxiang' and 'Bai Ruyu', the DEGs were annotated into 215 GO categories. Among them, 124 categories belonged to 'biological process' and the highest degree of enrichment was found for 'stimulus response'; 42 categories belonged to 'cellular components', with cytoplasm showing the highest degree of enrichment; 49 categories were found for 'molecular functions' and 'catalytic activity' was the most enriched (Fig. 6). At the fruit mature stage of 'Jinxiang' and 'Bai Ruyu', the DEGs were annotated into 256 GO categories. Among them, 166 categories belonged to 'biological process' and 'chemical' showed the highest degree of enrichment; 55 categories were classified into 'cellular components', of which 'cytoplasm' had the highest enrichment; 35 categories belonged to 'molecular functions' and the highest degree of enrichment was found for 'catalytic activity' (Fig. 7).

Identification of Genes Involved in Carotenoid Biosynthesis

By comparing the DEGs during fruit development and color transition of the three varieties, it was found that 42 common genes were differentially expressed in each comparison, among which 16 were up-regulated and 13 were down-regulated (Fig. 8). According to the fold changes of the DEGs at different stages, P values and the expression results at the fruit mature stage, four candidate genes were identified, including *LOC18774744* (encoding UDP-glycosyltransferase 87A1 (UGT87A1)), *LOC18792568* (encoding the carotenoid cleavage dioxygenase 4 gene (CCD4)), *LOC109949966* (an unannotated gene), and *LOC18779468* (encoding xyloglucan endoglucosylase/hydrolase protein 8.

Verification of Carotenoid Synthesis Related DEGs by qRT-PCR

qRT-PCR was performed to verify the expression levels of the four candidate DEGs at the color transformation stage of fruit (Fig. 9). As a result, the two yellow flesh peach varieties showed significantly higher expression of *LOC109949966* and *LOC18779468*, but significantly lower expression of *UGT87A1* and *CCD4* relative to the white flesh peach variety.

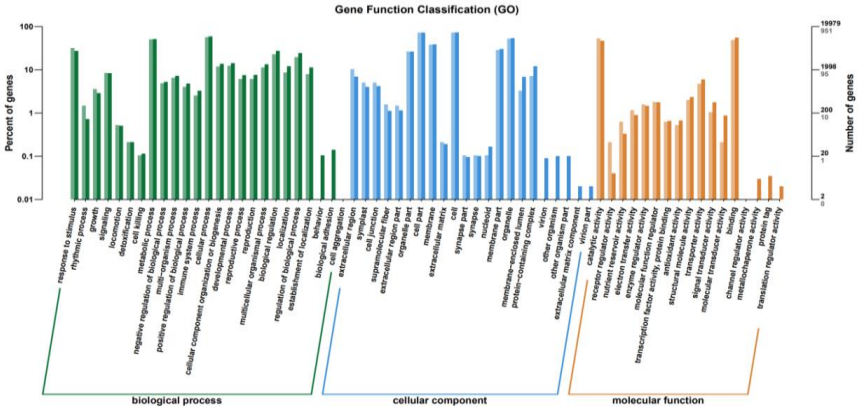

Fig. 2: Annotation of GO functions of differentially expressed genes at the S1 stage of 'Zhongtaojinmi' and 'Bairuyu'

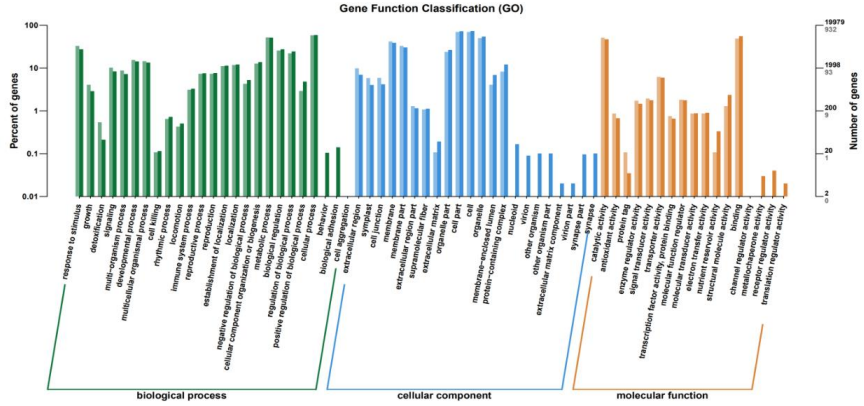

Fig. 3: Annotation of GO functions of differentially expressed genes at the S2 stage of 'Zhongtaojinmi' and 'Bairuyu'

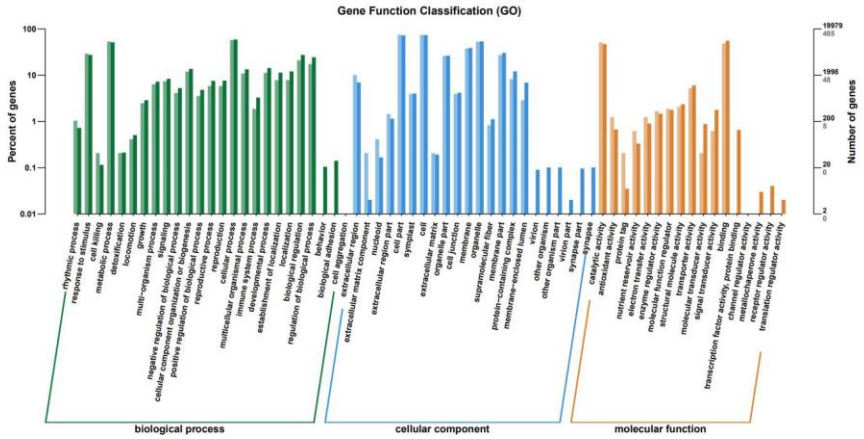

Fig. 4: Annotation of GO functions of differentially expressed genes at the S3 stage of 'Zhongtaojinmi' and 'Bairuyu'

Discussion

Previous studies have shown that α-carotene, β-carotene, lutein, zeaxanthin and β-cryptoxanthin, particularly β-carotene, are the main components of carotenoids in peach fruit, and the composition and content of carotenoids are closely related to the color of peach fruit (Kato *et al*. 2004; Centeno *et al*. 2011; Barsan *et al*. 2012; Zhang *et al*. 2013). However, there were still many unknowns in the regulation of yellow peach carotenoid synthesis. In this study, the carotenoid content in the mature fruit of two yellow flesh peach varieties was significantly higher than that in the white flesh variety, suggesting a high accumulation of carotenoids in mature yellow flesh peach fruit.

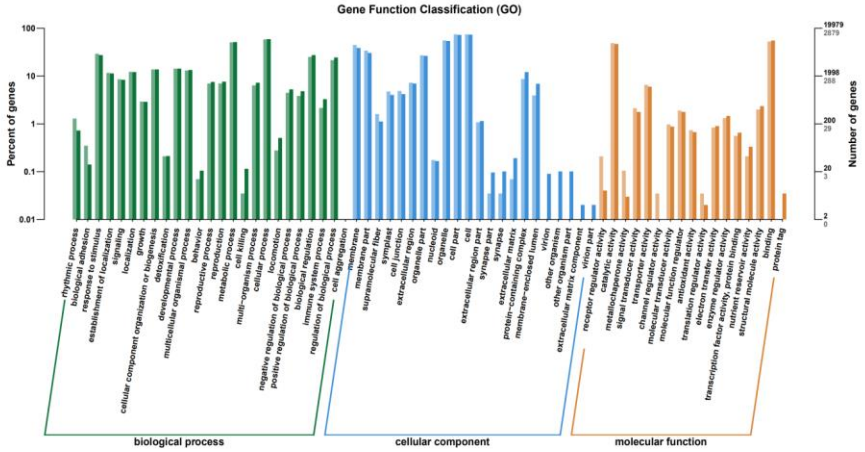

Fig. 5: Annotation of GO functions of differentially expressed genes at the S1 stage of 'Jinxiang' and 'Bairuyu'

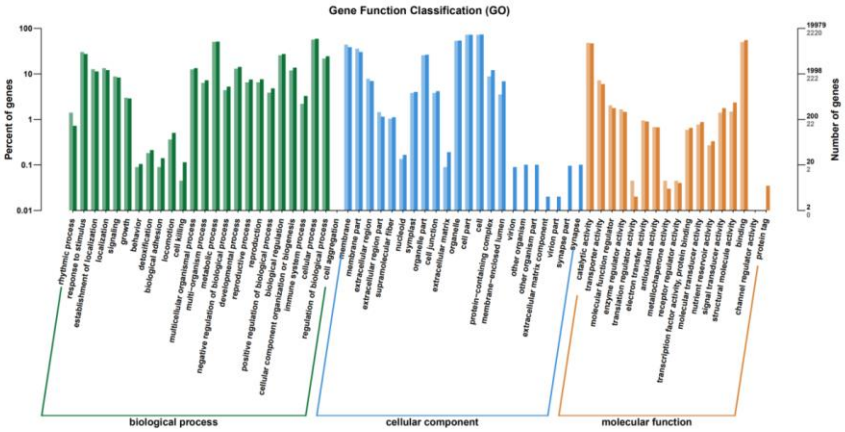

Fig. 6: Annotation of GO functions of differentially expressed genes at the S2 stage of 'Jinxiang' and 'Bairuyu'

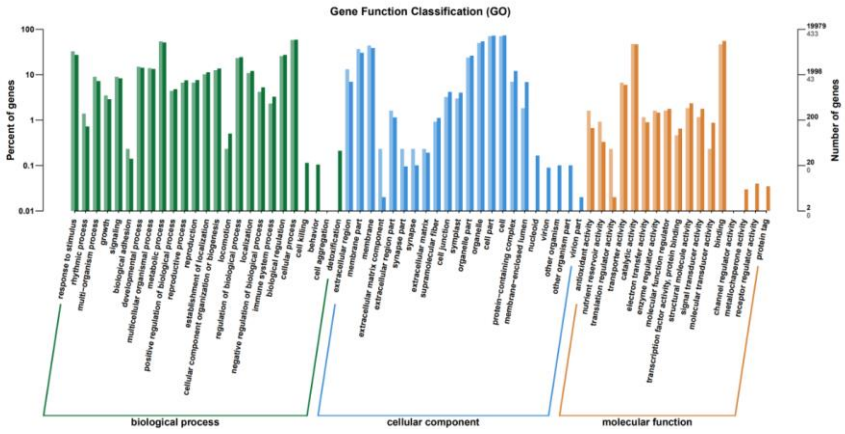

Fig. 7: Annotation of GO functions of differentially expressed genes at the S3 stage of 'Jinxiang' and 'Bairuyu'

The carotenoid content in the fruit is directly or indirectly regulated by the enzymes related to carotenoid biosynthesis in cells, and some genes further regulate the carotenoid biosynthesis by mediating the biosynthesis of relevant enzymes (Lu and Li 2008; Yuan *et al*. 2015). The multiple steps in the carotenoid biosynthesis pathway in fruit have been clarified, and many genes that directly regulate carotenoid synthesis have been cloned. The carotenoid biosynthesis process is regulated not only directly by related synthase genes, but also indirectly by some other genes.

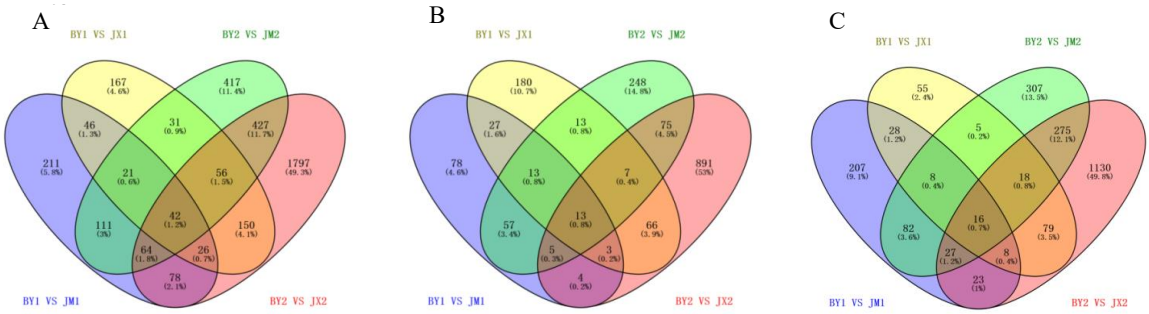

Fig. 8: Venn diagrams of differentially expressed genes (DEGs) showing changes of three peach cultivars at the S1 and S2 periods. (A) DEGs compare results at different stages. (B) downregulated DEGs. (C) upregulated DEGs. BY1, Bai Ruyu in young fruit stage (S1 period); BY2, Bai Ruyu in color transformation stage (S2 period); JM1, Zhongtao Jinmi in young fruit stage (S1 period); JM2, Zhongtao Jinmi in color transformation stage (S2 period); JX1, Jinxiang in young fruit stage (S1 period); JX2, Jinxiang in color transformation stage (S2 period)

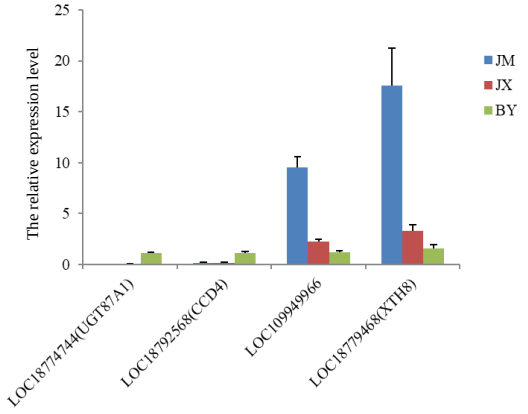

Fig. 9: qRT-PCR analysis of four candidate genes possibly involved in carotenoid biosynthesis at the fruit color transformation stage. JM, Zhongtao Jinmi (yellow flesh); JX, Jinxiang (yellow flesh); BY, Bai Ruyu (white flesh). Data represent the meaan ± standard error (SE)

The indirect regulatory mechanisms include mediation of the carotenoid synthase activities and the substrates required for carotenoid biosynthesis (Ma *et al.* 2014). In this study, the transcriptome data at the young fruit stage and color transition stage were analyzed to screen the DEGs, because in the yellow peach varieties, the carotenoid content was low at the young fruit stage and increased significantly at the color transition stage, while the white flesh variety showed a consistently low carotenoid content without significant changes at these two stages. Therefore, the gene expression in the fruit at these two stages may be directly related to carotenoid biosynthesis. As a result, four candidate DEGS were identified to be involved in carotenoid biosynthesis, including *LOC18774744*, *LOC18792568*, *LOC109949966* and *LOC18779468*. *LOC18774744* encodes UDP-glycosyltransferase 87A1 (UGT87A1) and the gene product is responsible for the first glycosylation step in the sophorolipid biosynthetic pathway in *Candida bombicola* ATCC 22214 (Saerens *et al.* 2011). In this study, the expression of *UGT87A1* in two yellow flesh peach varieties was significantly lower than that in the white flesh variety. We speculated that it may catalyze the transfer of sugar to receptor molecules, reducing the substrate sugar required in the process of carotenoid biosynthesis. *LOC18792568* encodes the carotenoid cleavage dioxygenase 4 gene (*CCD4*), whose main function is to cleave β-carotenoids (Falchi *et al.* 2013). Brandi *et al.* (2011) reported that there was no significant difference in the expression levels of carotenoid biosynthesis genes in the flesh between white flesh peach and yellow flesh peach. The reason for the white flesh color of peach was ascribed to the degradation of carotenoids in the flesh under the action of CCD4. In this study, we detected that the expression level of *CCD4* in two yellow flesh varieties was significantly lower than that in the white flesh variety, suggesting that *CCD4* plays a key role in the formation of yellow flesh in peach. *LOC109949966* is an unannotated gene. We found that its expression level in two yellow flesh varieties was significantly higher than that in the white flesh variety, indicating that it may be involved in the carotenoid biosynthesis metabolic pathway. *LOC18779468* encodes xyloglucan endoglucosylase/hydrolase (XTH) protein 8, which is widely present in various tissues and cells of plants (Cosgrove 2005). *XTHs* catalyze the breakage and reconnection of xyloglucan molecules to modify the cellulose-xyloglucan composite structure of plant cell walls and achieve cell wall reconstruction (Rose *et al.* 2002; Cosgrove 2005). The *XTHs* family comprises many members, whose expression characteristics vary greatly among different tissues and developmental stages of plants. They play important roles in the growth and development of plants and participate in multiple metabolic pathways (Potter and Fry 1994; Nishitani 1995; Catalá *et al.* 2001; Rose *et al.* 2002; Cosgrove 2005). For example, *XTH8* is highly expressed in the early developmental stage of leaves in *Arabidopsis*, which may be necessary for the growth and development at this stage (Becnel *et al.* 2006). *XTH8* also plays a very important role in fruit ripening and softening process (Goulao *et al.* 2007; Atkinson *et al.* 2009; Harb *et al.* 2012). In this

study, the expression level of *XTH8* in two yellow flesh peach varieties was significantly higher than that in the white flesh variety, implying that *XTH8* may be involved in some unknown pathways to indirectly regulate carotenoid biosynthesis. In summary, four candidate genes that may be involved in carotenoid biosynthesis were found through transcriptome sequencing analysis in this study. Some of these genes may participate in the indirect regulation pathways of carotenoid biosynthesis, such as the regulation of carotenoid synthase activities and the substrates required for carotenoid biosynthesis. Further research is needed to clarify the mechanisms.

Conclusion

Carotenoid content is closely associated with the quality of peach fruit. In this study, HPLC analysis demonstrated a high accumulation of carotenoids in the flesh of two yellow peach varieties ('Zhongtao Jinmi' and 'Jinxiang'), while the carotenoid content in the white peach variety ('Bairuyu') was extremely low. Transcriptome sequencing was performed to analyze the DEGs at three fruit development stages. Four genes, including *LOC18774744* (encoding UDP-glycosyltransferase 87A1 (UGT87A1), *LOC18792568* (encoding the carotenoid cleavage dioxygenase 4 gene (CCD4), LOC109949966 (an unannotated gene) and *LOC18779468* (encoding xyloglucan endoglucosylase/hydrolase protein 8), were identified as the candidate DEGs involved in carotenoid synthesis. Further qRT-PCR analysis demonstrated that the expression of *LOC109949966* and *LOC18779468* in yellow flesh peach was significantly higher than that in white flesh peach, while it was the opposite case for *UGT87A1* and *CCD4*. Further research is needed to explore how these genes are involved in carotenoid synthesis.

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Author Contributions

Haiyan Song and Ke Yang carried out the experiments and wrote the manuscript. Shuxia Sun, Ronggao Gong, Dong Chen and Jing Li participated and analyzed data. Meiyan Tu, Zihong Xu and Piao Liu participated in the collection of plant materials. Guoliang Jiang designed the experiment of this study.

Conflict of Interest

The all authors declare that they have no conflict of interest.

Data Availability

The author confirms that the data will be provided with a fair request to the corresponding author.

Ethics Approval

Not applicable to this paper.

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