COMPARATIVE TRANSCRIPTOMICS ANALYSIS OF HIGH AND LOW YIELD *CAMELLIA OLEIFERA* (OIL-CAMELLIA) CLONES

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Abstract. *Camellia oleifera* is an important woody oil plant. In this study, the transcriptomes and leaf nutrient contents of *C. oleifera* that displayed high-yield and low-yield group were studied. A *C. oleifera* clone with a stable yield was used for the control group. The genes that were differentially expressed in the groups with different yield were also screened. It was found that the contents of N, P, K, Mg, Zn, and Fe were lower in high-yield group than in low-yield and control group. High expression of aspartate aminotransferase in high-yield group may have promoted N conversion and absorption. Further, the high expression of malate dehydrogenase in high-yield group clones may have been directly related to yield. The yield difference and its cause were thus preliminarily expounded.
Keywords: *Camellia oleifera, comparative transcriptomics, yield, nutrient element, differentially expressed genes*

Introduction

Camellia oleifera (Fam.: *Theaceae*; Gen.: *Camellia*) is one of the main woody oil species in China. *Camellia* plants have high oil content, and the composition of the oil is very similar to that of olive oil (Ma et al., 2010). Camellia oil is unique in its chemical composition and medical and healthcare functions. It is rich in unsaturated fatty acids (which account for more than 90% of the total oil), of which the monounsaturated fatty acid, oleic acid, contributes to about 80% of the total oil content (Lin et al., 2019). Besides, *C. oleifera* oil also contains a multitude of bioactive components, such as sterol, squalene, polyphenols, sasanquasaponin, tocopherols, and other functional substances (Zhang et al., 2022). The appearance of camellia flowers in the process of fruit cultivation, fruit rate, yield per unit area of heterogeneity, the occurrence of a bumper year or shortfall year, and the yield and quality of *C. oleifera* are controlled by genetic and environmental factors, such as flower bud differentiation. The most suitable temperature range for growing *C. oleifera* is 27–33°C, and an average daily sunshine time of more than 10 h is optimal. Low temperature inhibits the normal opening of camellia flowers (Jiang et al., 2018; Wu et al., 2020). Zeng et al. (2023) conducted a study on the impact of altitude on the growth and yield of *C. oleifera*. The results showed that the photosynthetic parameters, tree height, stem diameter, crown width, and yield of *C. oleifera* gradually decreased with the increase of Planting altitude. The growth and yield were relatively higher at altitudes of 310 m and 450 m (Zeng et al., 2023). Regression analysis showed that soil pH, organic matter, Ca content, and leaf Ca and Zn content were the main controlling factors of fruit yield, among which pH had the highest influence. Fruit quality was mainly affected by

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soil organic matter, N, K, Fe, Mn content, leaf Mg content, and fruit P, K, Fe, Zn content (Deng et al., 2024). Yield and quality are key factors influencing the development of the *C. oleifera* industry. During the growing period, spraying nutritional elements (ABT vitality, GGR6, Komix, and fruit retention agent) or plant growth regulators (Brassinolide and Atonik) has a significant impact on the fruit set rate and yield of *C. oleifera*. Spraying the fruit retention agent can significantly increase the weight of individual fruits (Zhang et al., 2024).

RNA sequencing (RNA-seq) is a new generation sequencing technology developed in recent years. RNA-seq is widely used to explore the function of genes, construct expression maps, screen for molecular markers, compare varieties (Tai et al., 2015), and to analyze evolutionary relationships. For example, in a previous study, by analyzing different stages of oil accumulation, the differentially expressed genes (DEGs) related to oil accumulation were found (Lin et al., 2018). Wild *Camellia* populations at different latitudes display genetic differences and may undergo cold domestication at temperatures below 10°C (Chen et al., 2017). Potential associations have previously been found between the expression of genes and catechins, theanine, and caffeine contents in *C. sinensis* and *C. oleifera* (Tai et al., 2015). Transcriptome sequencing was performed to identify genes related to *Camellia oleifera* tea saponin metabolism. A total of 8107 differentially expressed genes were identified, and these genes were classified into 49 Gene Ontology (GO) functional categories and involved 129 Kyoto Encyclopedia of Genes and Genomes (KEGG) metabolic pathways. Genes highly correlated with tea saponin metabolism showed significant variation (Liu et al., 2022).

Currently, research on *C. oleifera* mainly focuses on the primary economic traits of its fruit, oil quality and Composition, the expression of genes related to oil transformation and accumulation during different periods, the bioactive ingredients, biological functions and extraction technologies (Qin et al., 2024). In this study, the nutrient content and transcriptomic analysis were carried out on the leaves of *C. oleifera* clones with high and low yields to analyze the DEGs related to the yield. The aim was to provide a theoretical basis for the screening of improved varieties with high yields, and to provide insight into the mechanisms underlying stable yields.

Materials and methods

Materials

The experimental materials were taken from Yuping County, Tongren City, Guizhou Province, China (27.362°N, 108.938°E), which is one of the main production areas of *C. oleifera*. In the same year, a single *C. oleifera* clone with plants that displayed significant yield differences was selected. In the high-yield group (Group A), fruit yield per plant was 11.76 kg (*Table 1*), but the leaves were yellow-green. The fruit yield of the low-yield group (Group B) was only 0.78 kg, but the flower quantity was much more than the high-group. The clone, with stable annual yields, was selected as the control group (Group C). The leaves in Group C were bright green. A total of 12 plants were selected from each group, and 20 leaves were collected from each plant from different parts of the tree. The materials for each group were mixed, wrapped in tin foil, and stored in liquid N_2 for RNA extraction. In the same way, leaves from the three test groups were collected, brought back to the laboratory, dried to constant weight at 80°C in an air-blast drying oven, ground into powder, and packed into a zipline bag for element content analysis.

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Table 1. *Growth traits and yield of C. oleifera*

Group	Age (years)	Height (m)	Ground diameter (cm)	Crown width (m×m)	Fruit weight (g)	Yield per plant (Kg)	Leaf color
High-yield	7	2.24	8.9	2.53×2.60	11.36	11.76	yellow-green
Low-yield	7	2.60	12.0	2.50×2.46	11.25	0.78	green
Control	7	2.52	9.0	2.58×2.20	14.01	8.21	green

Determination of nutrient elements in leaves

The N, P, and K contents in the leaves were determined using the H_2SO_4 -H_2O_2 digestion method (Fang et al., 2011). After digestion, the total N content was measured using the Kjeldahl method; the total P content was determined using the vanadium molybdenum yellow colorimetric method; and the K content was measured using a flame photometer. The Ca, Mn, Fe, Zn, Mg and Cu contents in the leaves were determined using the H_2SO_4-$HClO_4$ digestion method (Zhang et al., 1999), the elements were measured using an atomic absorption spectrometer (AAS). Each experiment was repeated three times.

RNA extraction and library preparation

A Trizol Kit (Invitrogen, USA) was used to isolate total RNA. Gel electrophoresis was used to detect RNA integrity. A Nanodrop ND200 (Thermo Fisher, Waltham, MA, USA) was used to measure the RNA purity at $OD_{260/280}$. An Agilent 2100 bioanalyzer (Agilent, California, USA) was used to measure the RNA concentration, RIN value, and 28S/18S value. The RNA samples were required to have values as follows: RIN ≥ 8, 28S/18S ≥ 1.5, and $OD_{260/280} \geq 1.8$. The eukaryotic mRNAs were enriched by magnetic beads with oligo(dT) after the RNA concentration and integrity had been tested. The mRNA was then fragmented by adding fragmentation buffer. Using mRNA as the template, one strand of cDNA was synthesized with 6-base random primers, and then two strands of cDNA were synthesized. After purification, terminal repair was performed, poly(A) tails were added, and sequencing adapters were connected. Segments were selected according to size using AMPure XP Beads. Illumina Hiseq™ sequencing was performed after using the USER enzyme to degrade the second strand of cDNA containing U, which allowed for the retention of the strand with the directivity of the mRNA.

Evaluation, assembly, and annotation of raw data

The quality of the original data was evaluated using FastQC (https://www.bioinformatics.babraham.ac.uk/projects/fastqc/). Trimmomatic (Bolger et al., 2014) was used for quality trimming and to remove sequences with N bases, joint sequences (forward, AGATCGGAAGAGCACACGTCTGAAC and reverse, AGATCGGAAGAGCGTCGTGTAGGGA), low-quality bases (Q < 20), reads shorter than 35 nt, and paired reads. Through these processes, clean reads were obtained. Trinity (Haas et al., 2013) was used to combine the valid data from the samples using the default parameters and min_kmer_cov 2. The National Center for Biotechnology Information (NCBI) Basic Alignment Search Tool (BLAST+) was used to compare the transcriptomes with the Conserved Domain Database (CDD), Eukaryotic Orthologous Groups of

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proteins (KOG), Clusters of Orthologous Groups of proteins (COG), NR, NT, PFAM, Swiss-Prot, and TrEMBL databases, amongst others, so as to obtain functional annotation information. Gene Ontology (GO) functional annotation information was obtained based on the transcripts and the Swiss-Prot and TrEMBL annotation results. Functional Kyoto Encyclopedia of Genes and Genomes (KEGG) annotation of the transcriptomes were obtained using KEGG Automatic Annotation Server (KAAS) (Moriya et al., 2007). The coding sequences (CDSs) were predicted according to the results of the BLAST comparison and TransDecoder. Bowtie2 (Langmead et al., 2012) was used to compare the effective data of the samples to the transcripts obtained by splicing, and statistical mapping information was obtained.

Gene expression quantification, differential expression analysis, and functional enrichment

Salmon (Patro et al., 2017) was used for expression statistics, and gene expression was represented in transcripts per million (TPM). DESeq2 (Love et al., 2014) was used for DEG analysis with the screening standards set as |Fold Change| > 2 and q-value < 0.05. Hierarchical clustering was performed for the DEGs. The GO and KEGG functional enrichment analysis of DEGs was carried out using clusterProfiler (Yu et al., 2012).

Real-time quantitative PCR (RT-qPCR) verification

Eight genes were selected for verification, with the glyceraldehyde 3-phosphate dehydrogenase (*GAPDH*) gene of *C. oleifera* used as the internal reference gene. There were three biological and technical replicates for each reaction, PCR primers were designed using Oligo 7 *(Table 2)*. Total RNA was extracted using a Trizol kit (Invitrogen). For each sample, 1 g of total RNA was used for cDNA synthesis using the PrimeScript kit (Takara Biotechnology, China). Then, a LightCycler® FastStart DNA Master^{PLUS} SYBR Green I Kit (Roche) was used to conduct the RT-qPCR reaction on a LightCycler ® 480 Instrument II (Roche). The reaction conditions were as follows: 30 s of preheating at 95°C, 5 s of thermal denaturation at 95°C, and 34 s of annealing at 55°C for 40 cycles. The relative expression level was determined using the $2^{-\Delta\Delta CT}$ method (Schmittgen et al., 2008).

Table 2. *Primer sequences*

Sequence Definition	Forward primer	Reverse primer
TRINITY_DN114864_c1_g3	TGTTCCTCTTGGTCTCTCTTCTG	GATTGAAGGCTCCTCTGTTCTG
TRINITY_DN120072_c0_g2	GCTCTCAAACCCAGTCACCGTC	CACCCAGCCGAAGATAATAAAG
TRINITY_DN116370_c0_g1	GTGGTAAGACAGGGATGGAAAG	GTTGAGGTTGTGTTTGTGTGTGA
TRINITY_DN121015_c2_g2	TCTCTCTATACAGCACTCGG	TCTCCCCATAGTTCTGCTTA
TRINITY_DN117920_c1_g1	ATTGTTGGACTGGCAGATTTCA	TTGGCTGGGGAATGTATATTGT
TRINITY_DN120929_c0_g1	TCACCGCCAATCAAGCAATCC	AGGTTATCTCTCACCAATCTC
TRINITY_DN118625_c2_g1	CCCCATCTTCATCTCCTCCAC	TCAAAACCATACTCTTCCTCAG
TRINITY_DN112121_c0_g1	TCAAAGATGGGGAAGGAGGG	TTGAACACGCTCCTCCACAC

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Results and analysis

Analysis of nutrient element content differences between clones

The analysis of leaf nutrient elements in the three plant groups (*Figure 1*) showed that the contents of N, P, K, Mg, Zn, and Fe were all lower in high-yield group (Group A) than the low-yield group (Group B) and the control group (Group C). There was a significant difference in the P content between Group A and C. There were also significant differences in the P and Ca contents among Groups A, B, and C.

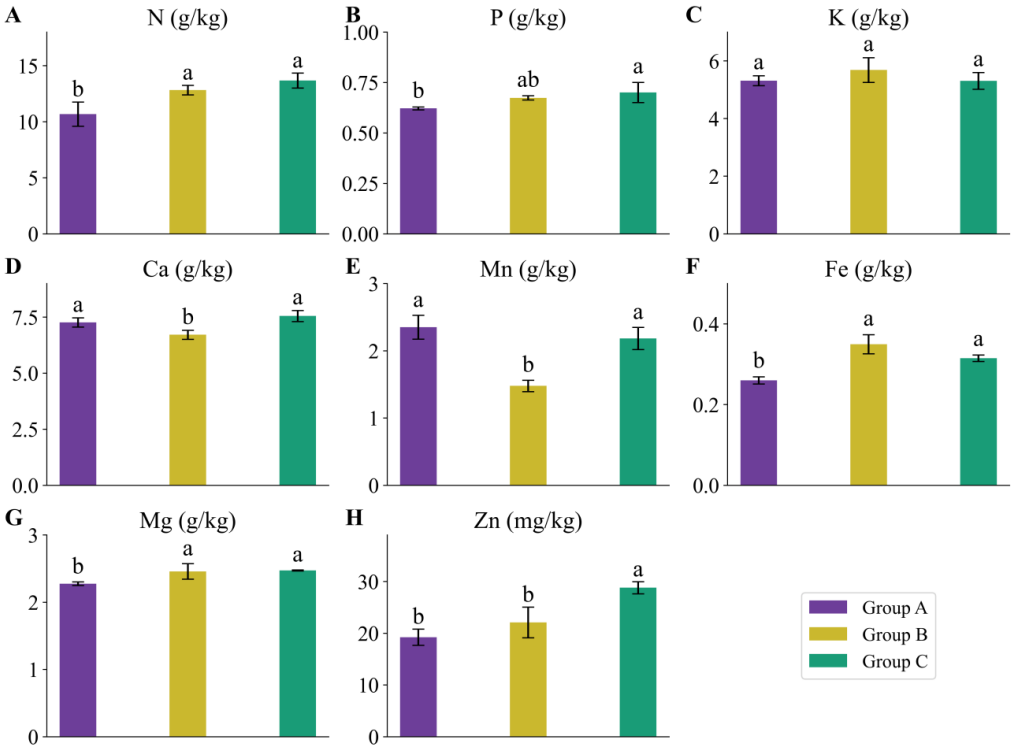

Figure 1. *Leaf nutrient element content of Camellia oleifera*

Data quality control, splicing, annotation, and comparison

After the sequencing data were obtained, the raw read quality was controlled. The Q20 value of the samples was above 98.38–98.64%, the Q30 was 94.29–94.91%, the GC value was 46.79–52.31%, and the proportion of unknown N bases was below 0.01%. The GC content of each sample was distributed horizontally, indicating that the sequencing quality was generally good and the data were reliable, and thus suitable for further analysis. After splicing, a total of 275,805 sequences with an average length of 452.65 bp were obtained. Of these sequences, 60,757 were larger than 500 bp, and 23,343 were larger than 1000 bp. The number of functionally annotated Unigenes obtained using the NR database was 71,515 (25.93%), the Swiss-Prot database was 114,527 (41.52%), GO annotation was 113,490 (41.15%), and KEGG annotation was 10,605 (3.85%). The reads were relatively evenly distributed across the genes (*Fig. 2A*).

The mapping results showed that the sample mapping rate was high, at between 90.18% and 91.44%. The analysis of the distribution frequency of redundant sequences showed that the overall trend was linear, smooth, extended, and spread, and that the

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database was expanded evenly (*Fig. 2B*). The quantitative saturation curve reached saturation, indicating that the sequencing data volume met the quantitative requirements (*Fig. 2C*).

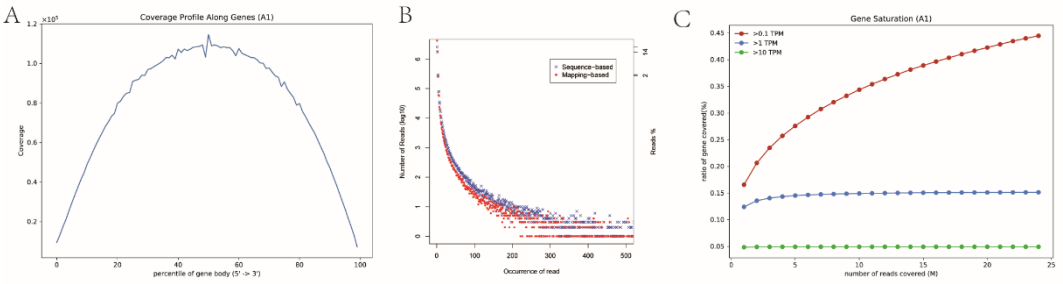

Figure 2. *Sequencing quality analysis. 2A Distribution of sequenced reads on Unigene; 2B Distribution frequency diagram of sample redundancy sequence; 2C Sample sequencing saturation graph. Note: 2A The horizontal axis represents the 5'->3' of genes, the vertical axis indicates the coverage of reads; 2B The horizontal axis represents data subsets of different scales, and the vertical axis represents the proportion of genes with quantitative error within 15% to the total number of genes. 2C Different colored lines represent different RPKM intervals*

Gene expression analysis and functional enrichment analysis

After the quantification of gene expression, correlation coefficient analysis and principal component analysis (PCA) were carried out (*Fig. 3*). Correlation heat map analysis showed that there was high correlation between samples in Group A. The PCA results showed that the value of PCA1 and PCA2 was 20.97% and 15.5%, respectively. After identifying differential expression, 234 DEGs were found in Group A, which included 48 upregulated genes and 186 downregulated genes, as compared to Group C. In Group B, there were 296 DEGs, including 163 upregulated genes and 133 downregulated genes, as compared to Group C. There were 233 DEGs between Group A and Group B, among which, 142 were upregulated and 91 were downregulated.

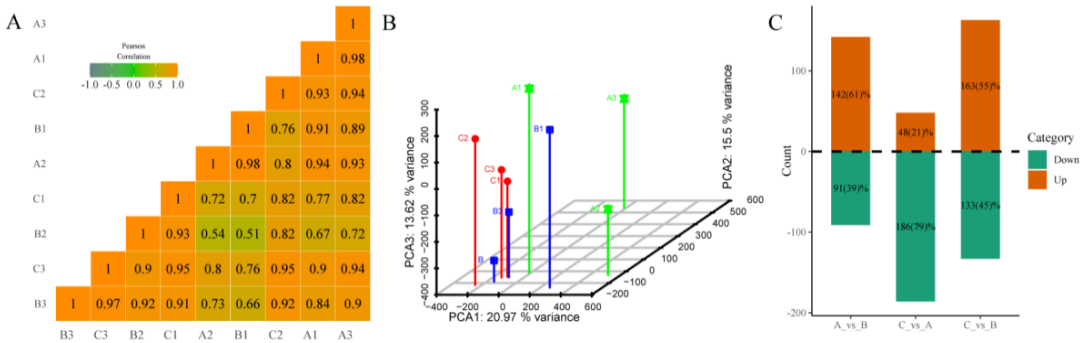

Figure 3. *Correlation of gene expression and differentially expressed genes. 3A Correlation heat map; 3B Three-dimensional diagram of principal component analysis; 3C A histogram of the number of differentially expressed genes*

GO enrichment analysis was performed for DEGs between Group A and B. The biological process (BP) entries showed that the response to hormone (GO:0009725), response to endogenous stimulus (GO:0009719), cellular response to hormone stimulus

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(GO:0032870), chloroplast (GO:0009507), plastid (GO:0009536), and senescence-associated vacuole (GO:0010282) were enriched. In item GO:0009725, TRINITY_DN120072_c0_g2 (auxin-responsive protein SAUR3), and TRINITY_DN116370_c0_g1 (glutathione S-transferase [GST]) were upregulated in Group A relative to Group B (*Fig. 4A*).

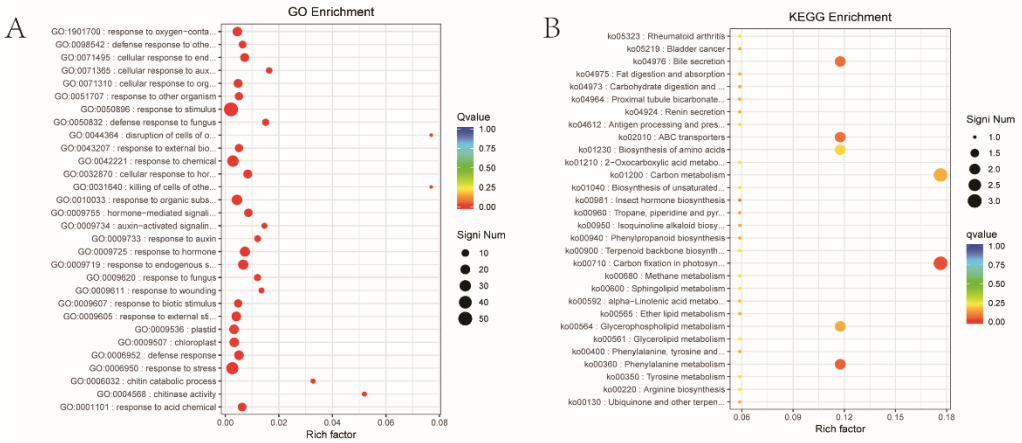

Figure 4. *Functional enrichment map of differentially expressed genes. A: GO enrichment (group A vs group B); B: KEGG enrichment (group A vs group B)*

KEGG enrichment analysis was also performed for DEGs between Group A and B. The results showed that the pathways of carbon fixation in photosynthetic organisms (ko00710) and phenylalanine metabolism (ko00360) were enriched (*Fig. 4B*). In the carbon fixation in photosynthetic organisms pathways, the TRINITY_DN121015_c2_g2 (malate dehydrogenase [oxaloacetate-decarboxylating][NADP+]; MDH; EC 1.1.1.40) and TRINITY_DN117920_c1_g1 (aspartate aminotransferase, cytoplasmic; AspaT; EC 2.6.1.1) genes were upregulated in Group A.

RT-qPCR

To verify the reliability, 8 genes related to the yield were selected from the transcriptome data for RT- qPCR. The results showed that the expression trends of these 8 genes were basically consistent between the transcriptome data and the RT-qPCR results, indicating that the RNA-seq results have high reliability (*Figure 5*).

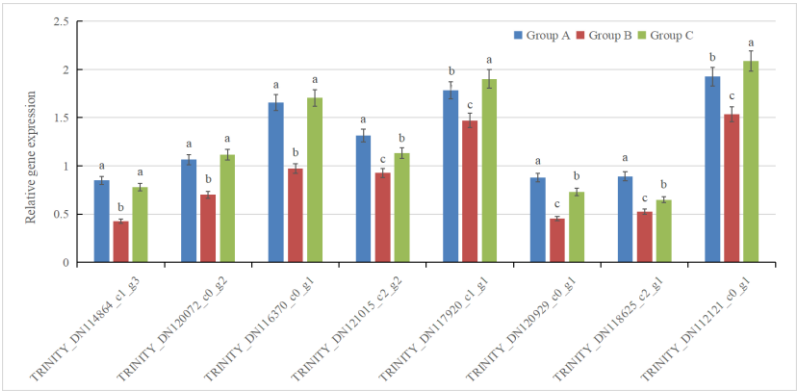

Figure 5. *RT-qPCR validation*

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Discussion

The leaf is the main site of photosynthesis. Nutrients accumulate and are transferred between branches and leaves, which results in branch, leaf, and fruit growth. The fruit yield and leaf characteristics on different types of branches are correlated (Liu et al., 2020a). N, P, and potash fertilizers are conducive to the flower bud differentiation of *C. oleifera*. In addition, the contents of N, P, and K in *C. oleifera* leaves have been shown to display a trend of "increasing–decreasing–increasing" along with the annual growth and development processes of the plant (Mo et al., 2013). In the pre-differentiation stage, N fertilizer has been shown to significantly promote the elongation of *C. oleifera* mixed buds. At sepal forming stage, N fertilizer significantly promoted bud elongation (Luo, 2018). N is one of three basic elements in higher plants. Increasing N can increase yield, chlorophyll content, and photozyme activity (Yan et al., 2018; Liu et al., 2020b; Li et al., 2020). N and P are essential for the formation of chlorophyll and enzymes related to photosynthesis, influencing the photosynthetic activity of plants. A deficiency in N and P can lead to a decrease in photosynthetic activity, reduced synthesis of chlorophyll, and impact plant growth and development (Cheng et al., 2024). When there is a nitrogen deficiency, the leaves of *Moringa oleifera* seedlings become smaller, and the whole plant appears light yellow (Li et al., 2015); *Musa nana* also exhibits yellowing of the leaves (Wang et al., 2023); The expression levels of chlorophyll catabolism genes *MdNYC1*, *MdPAO*, and *MdSGR1* in the leaves of apple (*Malus pumila*) significantly increased, inhibiting chlorophyll synthesis. As a result, the chlorophyll content markedly decreased, causing the leaf color to change from dark green to light green, and finally to yellowish-green (Wen et al., 2022). Wang (2019) discussed the stoichiometric characteristics and distribution of N, P, and K in the organs of *C. oleifera* at different ages and found that N was the limiting element affecting the growth of *C. oleifera*. Gao (2016) selected 20 representative *C. oleifera* plantations and analyzed the mineral nutrient elements during the fruit enlargement and oil conversion phase. The results showed that the leaf nutrient contents in October were in the following order: Mn > Fe > N > P > K. Hu (2016) measured the contents of nutrient elements in the leaves at different development stages of *C. oleifera*. It was found that the Mn content was in high demand in the sapling and initial fruits stages. In another study, nutrient element determination and Diagnosis and Recommendation Integrated System (DRIS) analysis were carried out in 40-year-old Mengjiang *C. oleifera* with high and low yield in Sanjiang County, Guangxi. The results showed that N, P, and Fe deficiencies were the most important factors limiting *C. oleifera* yield. Trace elements are essential for plant growth, and so in the process of *C. oleifera* fertilization, attention should be paid to element collocation in order to avoid single or excessive fertilization (Tang et al., 2015). It was suggested that all nutrient elements should be supplemented to varying degrees before winter dormancy. Additionally, nitrogen, phosphorus, potassium, copper, calcium, zinc, and magnesium should be appropriately supplemented during the summer for optimal nutrition regulation of *C. oleifera* forests (Zeng et al., 2020). In the present study, the N, P, K, Mg, Zn, and Fe contents were lower in the high- yield group (Group A) than the low-yield group (Group B). This may be related to the constant transport of nutrients to fruits during the fruiting period, which may lead to a large decrease in nutrient contents in other organs. Meanwhile, the unbalanced distribution of nutrients resulted in fewer flower buds and yellowing leaves in the high yield group. Plants within the high yield group showed a certain degree of deficiency symptoms. Aspartic acid biosynthesis is mediated by AspaT, which plays a key role in the metabolic regulation of C and N metabolism in all

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organisms. As a key enzyme for primary N assimilation, AspaT can improve plant N utilization efficiency (de la Torre et al., 2014). Thus, the upregulated expression of TRINITY_DN117920_c1_g1 (AspaT) in Group A may have been related to N conversion, absorption, and demand.

Anthocyanins are water-soluble pigments that appear as colors such as red, pink, blue, and purple, in leaves (Ren et al., 2015). GST is associated with anthocyanin transport, and in purple tea, both CsGSTF1 and the transcription factor, CsMYB75, are related to the color of buds and leaves. CsGSTF1 can complement the anthocyanin deletion phenotype of the Arabidopsis mutant *tt19-8* but cannot restore the color of mature seeds (Wei et al., 2019). *FeGST1* transcripts of *Fagopyrum esculentum* have been found in the stems of wild-type cultivars, but not in the stems of green-type cultivars (Matsui et al., 2018). It has also previously been found that with the degradation of anthocyanins and the accumulation of chlorophyll, the two-color leaf phenotype (green margin and red center) of *Brassica oleracea* L. var. *acephala* gradually forms (Ren et al., 2019). Anthocyanins are important secondary metabolites and antioxidants in plants. Under nitrogen deficiency conditions, the anthocyanin content in the stems and petioles of *Manihot esculenta* increases as the nitrogen concentration decreases; similar changes are observed in the leaves of *Arabidopsis thaliana* (Cheng et al., 2024). The leaves in the high-yield group were yellow, and the leaves in the low-yield group were emerald green. TRINITY_DN116370_c0_g1 (GST) was upregulated in the high-yield group (relative to the low-yield group), which may be the reason for the difference in leaf color.

In the carbon fixation pathway, GAPDH, ribulose-1,5-bisphosphate carboxylase/oxygenase (RuBisCO), MDH, and other genes directly related to yield, have been shown to be highly expressed as a result of hybridization. Higher expression levels of genes in energy-related pathways at the beginning of panicle differentiation in rice may provide an explanation for hybrid line yield advantage (Katara et al., 2020). The overexpression of an apple cytoplasmic MDH gene (*MdcyMDH*) has been shown to significantly improve the photosynthetic rate, root weight, and total fresh weight of transgenic individuals (Wang et al., 2015). In the present study, within the carbon fixation in photosynthetic organisms pathways, TRINITY_DN121015_c2_g2 (MDH) and TRINITY_DN117920_c1_g1 (AspaT) were upregulated in Group A. The high expression of MDH in the high-yield may be directly related to the yield.

At the flower bud morphological differentiation stage of *C. oleifera*, the abscisic acid (ABA) content has been shown to decrease under fertilization treatment. Meanwhile, the zeatin riboside (ZR), indole acetic acid (IAA), and gibberellic acid (GA_3) contents showed an increasing trend (Yuan et al., 2019). It has been suggested that low concentrations of exogenous GA_3 may affect the ABA content by regulating the endogenous ZR, IAA, ABA, and GA_3 contents in the floral organs, thus affecting the fruit setting rate of *C. oleifera* (Ge et al., 2020). Different concentrations of GA treatment have been shown to change the distribution of the ^{13}C produced by the leaves of *C. oleifera* 'Xianglin 27' in various organs. In particular, different GA treatment concentrations significantly increased the distribution of ^{13}C in flower buds and fruit (Wen, 2019). IAA is a key factor affecting the yield and SPAD value of *C. oleifera*, and the interaction between IAA and GA is considered to be a major factor affecting the seed yield and kernel yield (Hu, 2011). Seed size is a crucial factor influencing the final seed yield, phytohormones related to regulating seed size have also been studied, Brassinolide (BR), auxins and salicylic acid (SA) are considered to be important participants (Ji et al., 2022). The higher expression of gibberellin 20 oxidase (GA20ox), lower expression of

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gibberellic acid insensitive (GAI), and the increase in 9-cis-epoxycarotenoid dioxygenase (NCED) at the mRNA level could contribute to oil accumulation (Song et al., 2021). In the present study, GO enrichment analysis was performed for genes that were differentially expressed between Group A and B. As a result, BP items, such as response to hormone (GO:0009725), response to endogenous stimulus (GO:0009719), and cellular response to hormone stimulus (GO:0032870) were enriched. The auxin response factor SAUR3, TRINITY_DN120072_c0_g2 (auxin response factor 6), TRINITY_DN118625_c2_g1 (SAUR22), and TRINITY_DN112121_c0_g1 (auxin repressed protein [ARP]) were upregulated in Group A. This indicates that IAA has a big influence on high *C. oleifera* yield. In addition, the expression of the TRINITY_DN120929_c0_g1 (*GRAS*) gene was significantly different between Group A and B. It has previously been found in the study of fruit trees such as apple, that the total number of seeds increases due to increase in the number of fruits, and GA was synthesized in large quantities in the seed embryo, which inhibited the formation of flower buds. Endogenous hormones such as ethylene, auxin, kinetin, and ABA, jointly control vegetative and reproductive growth, and play a significant role in the formation of alternate bearing (Zhang, 2016).

Conclusion

As flowering and fruit growth occur in the same period in *C. oleifera*, nutrients are distributed unevenly. This restricts the development of *C. oleifera.*This study showed that the contents of N, P, K, Mg, Zn and Mg were lower in the high-yield group than the-low yield and control groups. Further, it was found that the differential expression of SAUR3, GRAS and other related genes may be associated with the yield of *C. oleifera*. *C. oleifera* clones of the same age were used as the study materials to avoid the influence of environmental factors and individual genetic factors on the yield. This clone is an important material for the further study of the causes of alternate bearing. The DEGs that existed between the high-yield and low-yield groups were preliminarily screened. These DEGs will provide a theoretical basis for the study of alternate bearing in *C. oleifera*, the causes of which remain to be understood.

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