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Research Article

Effects of post-harvest natural drying on seed quality and endogenous hormones of *Camellia oleifera*

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Abstract

Camellia oleifera Abel. (*C. oleifera*), one of the four woody oil-producing plants in the world produces edible oils with high percentage of unsaturated fatty acid content in seeds. The mature *C. oleifera* seeds continue to undergo a series of physiological changes after harvest. To this end, the dynamic changes in nutrients, oil content, fatty acid composition, and endogenous hormone content in *C. oleifera* seeds under different natural drying times after harvest were investigated. The content of soluble sugar and soluble protein of *C. oleifera* seeds increased with the extension of natural drying, especially soluble sugar content increased nearly 2-fold at 30 d after post-harvest natural drying compared with that of the control group. The content of oil reached a peak (23.6%) at 30 d after post-harvest natural drying. During the post-harvest natural drying process, the relative content of palmitic acid and oleic acid increased, while the relative content of palmitic acid and linoleic acid decreased. Furthermore, the levels of unsaturated fatty acids (oleic acid, linoleic acid, linolenic acid, and arachidonic acid) increased significantly with increasing natural drying time. The overall trend of endogenous hormones SA, SL, and ACC concentrations increased with the post-harvest natural drying process. Furthermore, the concentration of SA, SL, and ACC were positively correlated with oil content. Altogether, post-harvest natural drying for 30 days significantly promoted the anabolism of oil and improved the quality of *C. oleifera* seeds. These findings provide a scientific basis for reasonable post-harvest treatment to improve *Camellia* oil yield.

Keywords: *Camellia oleifera*; endogenous hormones; fatty acid composition; oil content; post-harvest natural drying

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Introduction

Camellia oleifera Abel. (*C. oleifera*), a small evergreen tree or shrub, known as the world's four largest woody oil plants with olive, oil palm, and coconut (Wen *et al*., 2018; Wu *et al*., 2020). *C. oleifera* has a long history of cultivation and utilization in China, and the cultivation of *C. oleifera* has reached 453.33 hectares by 2020 (Qin *et al.*, 2018). For *C. oleifera*, most of the nutrients are concentrated in the seeds (Zhang *et al.*, 2021), and the *Camellia* oil is obtained by pressing *C. oleifera* seeds. *Camellia* oil is high-quality edible oil in the world, with higher unsaturated fatty acid content than peanut oil, soybean oil, and other herbal oils (Ma *et al.*, 2011). *Camellia* oil is also easily absorbed by the human body, for example, promoting the absorption of fat-soluble fiber and trace elements such as calcium, iron, and zinc. *Camellia* oil also contains various bioactive substances, which can improve cardiovascular health, reduce cholesterol and fasting glucose, and maintain normal blood triglyceride levels, etc. (Lee *et al.*, 2006; Di *et al.*, 2017). Additionally, *Camellia* oil has a wide range of uses in skincare and industry with a growing potential for market development (Zhang, 2013; Luo *et al.*, 2015). These beneficial efficacies depend on the genetic characteristics and post-harvest treatment measures of *C. oleifera* seeds (Gao *et al*., 2013; Zhang *et al*., 2021). Therefore, reasonable harvest and scientific post-harvest treatment technology are key to ensuring the quality of *C. oleifera* seeds.

Drying treatment is very important for post-harvest seeds. Seeds need to be stored after harvest, which has a high moisture content, so they should be dried. Furthermore, for some oil crops and fruits, post-harvest drying is also conducive to the transformation of oil or other beneficial substances (Baud *et al*., 2002; Borek *et al*., 2009; Ojediran *et al*., 2021; Wu *et al*., 2022). In hazelnut, the protein content has a gradual reduction, and the monounsaturated fatty acid content has a gradual increase with the increase in post-harvest drying time (Ali, 2018). The sugar content increased with the increase of drying time in jujube (Gao and Wang, 2013). Solar drying has been proven to convert ergosterol into vitamin D and maximize the nutritional value of mushrooms (Jiang *et al*., 2020). In mango and pineapple, the content of dry matter, carbohydrates, and total sugar increased significantly after drying (Mohammed *et al*., 2020). For *C. oleifera*, when the assimilation of green organs and the storage substances in the pericarp are rapidly transferred to the seed, the content of nutrients in the seed changes significantly, and a large amount of oil will form during this period (Peng *et al*., 2020). Natural drying could promote the oil accumulation in the seeds of the two *C. oleifera* species (Feng *et al*., 2017). In addition, the change of endogenous hormone affects the conversion of nutrients (Khavari *et al*., 2021) and has the same effect on *C. oleifera* seeds (Cao *et al*., 2015). Studies have shown that drying treatment has significant effect on the changes of endogenous hormone (Jing *et al.*, 2017). Xie *et al*. (2013) reported that the air-drying affected the changes of endogenous hormones in Intertidal Macro-Alga *Ulva pertusa*. In addition, the different drying processing times might play different roles (Mohammed *et al.*, 2020). Therefore, it is necessary to further study the post-harvest drying time of *C. oleifera* seeds.

To understand the effect of post-harvest natural drying time on the substances in *C. oleifera* seeds, the contents of moisture and oil, and fatty acid composition, as well as the endogenous hormone content were analyzed in the present study. This study will provide a suitable raw material treatment method for processing high-quality *Camellia* oil and promoting technological research and development to improve the yield and quality of *C. oleifera*.

Materials and Methods

Plant materials

The plant materials used 6-year-old high-crown-grafted *C. oleifera* cultivar '*Hua Shuo*', which was planted in the *C. oleifera* experimental field of Central South University of Forestry and Technology, located in Wangcheng District, Changsha City, Hunan Province (113° 31′ E, 28° 05′ N). The planting area is a subtropical monsoon humid climate with average annual precipitation (1,380 mm), an average annual temperature (19.3 °C), and an average annual frost-free period (271-291 days). A sufficient number of mature fruits were picked randomly from the different directions of each *C. oleifera* tree without diseases and insect pests. The collected fruits were placed on open-ended paper box with multiple air holes around in a shaded place, and seeds were collected at 0 days (d), 5 d, 10 d, 15 d, 20 d, 25 d, and 30 d after natural drying, respectively. Subsequently, frozen the seeds in liquid nitrogen quickly and stored at -80 °C (the sample of 0 d was set as the control group and each group had three biological replicates.

Analyzed of soluble sugar content and soluble protein content

The content of soluble sugar was determined according to John's description (John *et al*., 1950). Briefly, one-half-gram frozen samples were ground with liquid nitrogen and homogenized with 4 mL of 80% alcohol (v/v). The mixture was incubated in shaking water bath at 80 °C for 30 min. The supernatant was collected after centrifuging for 10 min (4000 r min^{-1}), and the residue was re-extracted with 80% alcohol (4 mL) as described above. The resulting supernatants were combined, and activated carbon (0.1%) was added, decolorized at water bath (80 °C) for 30 min, and then filtered. The obtained extract was added to 10 mL of distilled water to obtain the sugar extract. Add 5 mL enthrone reagent (0.1%) in 0.1 mL sugar extraction and place in a water bath at 100 °C for 10 min. After cooling, the OD value was measured at 620 nm with three times repetitions. The standard curve uses 1000 ug mL^{-1} glucose standard solution. Each experiment with three biological repetitions.

The content of soluble protein was determined according to the method (Bradford, 1976). The frozen sample (1 g) was ground with liquid nitrogen, and then 8 mL of distilled water was added. The mixtures were left for 0.5-1 h at room temperature (20-25 °C) to facilitate full extraction and then centrifuged for 20 min (4,000 r min^{-1}). Collected all the pooled supernatant, add distilled water to 10 mL, and then extracted to be tested. Then 0.1 mL extraction was added to 5 mL of Coomassie brilliant blue G250, and let stand for 2 min after fully mixing. The OD value at 595 nm was measured (zero is the blank value) and repeated three times. The standard curve was drawer using 1 mg mL^{-1} glucose standard solution. Each experiment with three biological repetitions.

Analyzed of moisture content, oil content, and fatty acid composition

To investigate the moisture content, the fresh seeds were weighed (W1) and then dried to constant weight (W2) in a 55 °C oven. Repeated three times.

$$\text{Moisture content (\%)} = (W1-W2) \times 100 / W1 \quad (1)$$

The oil content was analyzed according to the method of Gong et al. (2020). The oil of samples was extracted using Soxhlet extraction with organic solvent (petroleum ether). Specifically, the fresh seeds were dried to constant weight in an oven at 60 °C. Then grind the dried seeds into powder with a pulverizer. Accurately weighed 5.0 g of powder into a folded filter paper tube, which was tightened with a fire cotton thread and inserted into a Soxhlet extractor. About 50 mL of petroleum ether was added to the cup connected to the Soxhlet extractor. In each experiment, a weighed sample was extracted with a required solvent in a Soxhlet extractor for 30 min. The machine temperature was maintained at 75 °C during the extraction process,

and then rinsed for 150 min, recuperated the solvent for 60 min. The extracted oil was stored in the 4 °C refrigerators for further analysis. Repeated three times.

$$\text{Oil content} = [(\text{weight of cup and oil-weight of the empty cup}) / \text{weight of powder}] \times 100\% \quad (2)$$

The fatty acid content was analyzed followed with the guidelines of the Determination of Fatty Acids in Food of National Food Safety Standard in China (GB5009.168, 2016). The oil sample (60.0 mg) was accurately weighed and dissolved in 2 mL glycerol undecanoate solution and 4 mL isooctane. The oil solution is methylated by transesterification with 200 μL potassium hydroxide methanol solution, and the mixture was agitation for 30 s and stand for clarification at room temperature. About 1 g of sodium bisulfate was added to neutralize potassium hydroxide. After the salt is precipitated, move the upper solution to the upper bottle for testing. The gas chromatograph adopts a hydrogen flame ionization detector and chromatographic column (60 m × 0.25 mm × 0.2 μm). The carrier gas is nitrogen with a split ratio of 1:50. The automatic injection was 1 μL. The oven temperature was programmed from 50 °C (2 min hold) to 170 °C with 20 °C min^{-1} (10 min hold), then to 108 °C with 2 °C min^{-1} (10 min hold), finally to 220 °C with 4 °C min^{-1} (22 min hold). The fatty acids relative content was then calculated as follows: fatty acids relative content (%) = (fatty acid methyl esters peak area × conversion coefficient of fatty acid methyl esters to fatty acids) / (sum of peak areas of all fatty acid methyl esters × conversion coefficient of fatty acid methyl esters to fatty acids). All fatty acid content data in this work were presented as the mean ± SD of three biological repetitions.

Analyzed of endogenous hormones

In brief, 0.5 g frozen sample were ground with liquid nitrogen and homogenized with 9 mL PBS buffer (PH = 7.4). Centrifuged for 30 min (4 °C, 8,000 r min^{-1}). The supernatant was stored at 4 °C. The contents of strigolactone (SL), salicylic acid (SA), 1-aminocyclopropanecarboxylic Acid (ACC), brassinosteroid (BR) and methyl jasmonate (MeJA) were determined using an Enzyme-Linked ImmunoSorbent Assay (ELISA) kit according to instructions of the manufacturer. The plant hormone strigolactone (SL) ELISA kit, plant hormone salicylic acid (SA) ELISA kit, plant hormone 1-aminocyclopropanecarboxylic acid (ACC) ELISA kit, plant hormone brassinosteroid (BR) ELISA kit, and plant hormone methyl jasmonate (MeJA) ELISA kit were used to determine the endogenous hormone content of SL, SA, ACC, BR, and MeJA, respectively, following the manufacturer's instructions. The reaction was terminated by the addition of a sulfuric acid solution, and the color change was measured spectrophotometrically at 450 nm using a microplate reader (BioTek, Winooski, Vermont, USA). Each experiment with three biological repetitions.

Statistical analysis

Data were analyzed with a one-way ANOVA using the Statistical Package for the Social Sciences (SPSS 11.0, SPSS Inc., Chicago, Illinois, USA) and Origin 9.0 software (OriginLab, Northampton, Massachusetts, USA). Means were compared using Duncan's honestly significant difference test at $p < 0.05$. All data had three biological replicates, and were analyzed and represented as the mean ± SD ($n = 3$). The corrplot package in RStudio 1.3.959 (RStudio, Boston, Massachusetts, USA) was used for the correlation analysis.

Results

Dynamic changes of soluble sugar and protein content in C. oleifera seeds after post-harvest natural drying

The changes of soluble sugar content were shown in Figure 1A. Compared with 0 d, the soluble sugar content in 10-30 d increased to different degrees, especially in 20 d and 30 d, and the content of soluble sugar after 20 and 30 d at post-harvest drying treatment increased by 1.77- and 1.89-fold, respectively. The soluble

sugar content of *C. oleifera* reaching 156.2 mg g^{-1} at 30 d of treatment and increased nearly 2-fold compared with that of the control group. These results indicated that the soluble sugar content of *C. oleifera* increased with the extension of natural drying time.

The content of soluble protein decreased first but then increased. The observed maximum soluble protein reached 13.9 mg g^{-1} at 20 d, which was approximately 1.54-fold higher than the control group (Figure 1B). Compared with the control group, the soluble protein content increased slightly after 30 d. These results showed that post-harvest natural drying time had little effect on the soluble protein content of *C. oleifera* seeds.

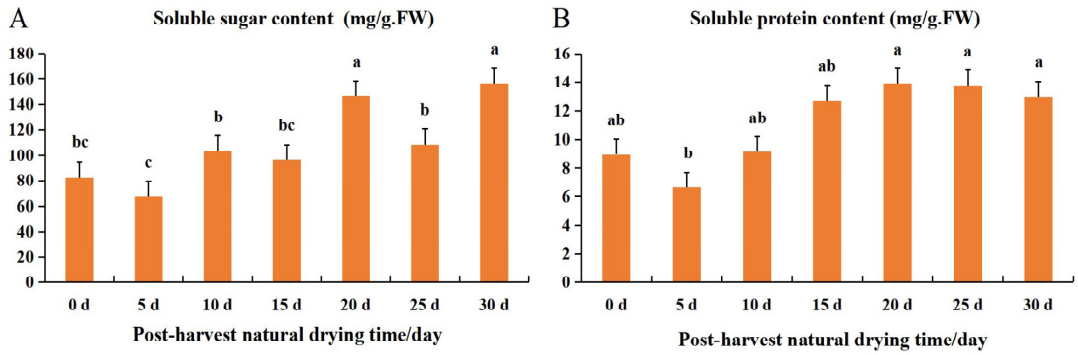

Figure. 1. Effect of post-harvest natural drying time on the content of soluble sugar (A) and soluble protein (B) in *C. oleifera* seeds

Values as shown as means ± SDs of three biological replicates. Different letters between treatments denote significant differences (ANOVA test, $P < 0.05$)

Dynamic changes of moisture and oil content in C. oleifera seeds after post-harvest natural drying

With the increase in natural drying time, the moisture content of *C. oleifera* seeds decreased from 60.2% (0 d) to 15% (30 d) (Figure 2A). The moisture content of *C. oleifera* seeds tended to be stable around 20 d after post-harvest natural drying.

As shown in Figure 2B, drying time after harvest has a more obvious effect on the oil content of *C. oleifera* seeds. The content of the oil increased from 18.2% to 22.9% during 0-5 d of natural drying. However, the seed oil content decreased significantly from 5 to 15 d, reaching the lowest value of 16.7% at 15 d. Subsequently, the content of oil increased significantly from 15 to 30 d, reaching a peak (23.6%) at 30 d after postharvest natural drying (Figure 2B).

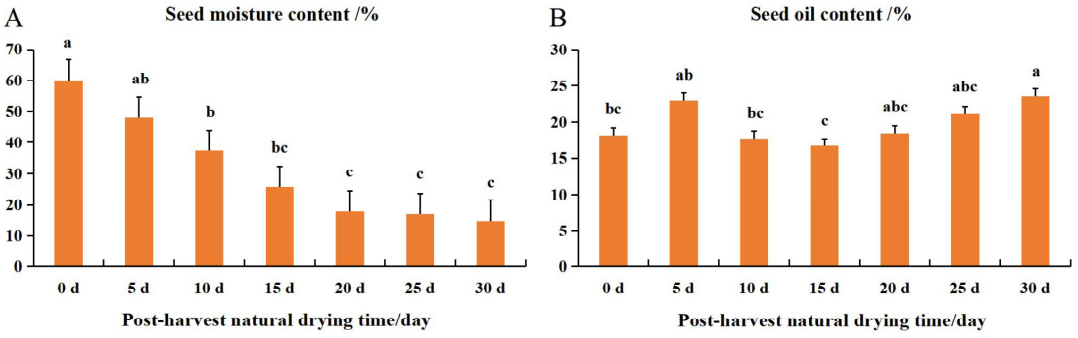

Figure 2. Effect of post-harvest natural drying time on the content of moisture (A) and oil (B) in *C. oleifera* seeds

Values as shown as means ± SDs of three biological replicates. Different letters between treatments denote significant differences (ANOVA test, $P < 0.05$)

Moreover, there was an opposite trend between moisture content and oil content. Therefore, the optimum time of natural drying after harvest was 30 d, which can effectively improve the oil production of *C. oleifera* seeds.

The changes of fatty acid compositions in C. oleifera seeds after post-harvest natural drying

The fatty acids of *C. oleifera* were mainly composed of palmitic acid, stearic acid, oleic acid, linoleic acid, linolenic acid, and arachidonic acid (Figures 3-5). The relative content of oleic acid accounted for about 80% in *C. oleifera* seeds and increased with the natural drying time (Figures 5A and 4B). The overall trend of palmitic acid content decreased with natural drying time (Figure 4A). The stearic acid content was about 2% and the overall trend was similar to linolenic acid (Figures 3B and 4B). A small amount of linolenic acid (0.3%) was determined in *C. oleifera* seeds (Figure 3B). The trend of linolenic acid content decreased from 5 to 15 d; but recovered after 15 d (Figure 4E) and the content of arachidonic acid increased after treatment compared with 0 d (Figure 4F).

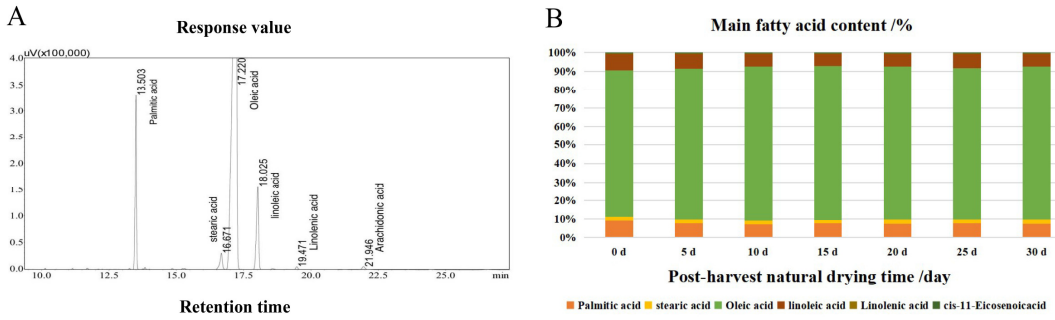

Figure 3. The fatty acids of *C. oleifera* (A) and effects of post-harvest natural drying time on fatty acid composition (B) in *C. oleifera* seeds

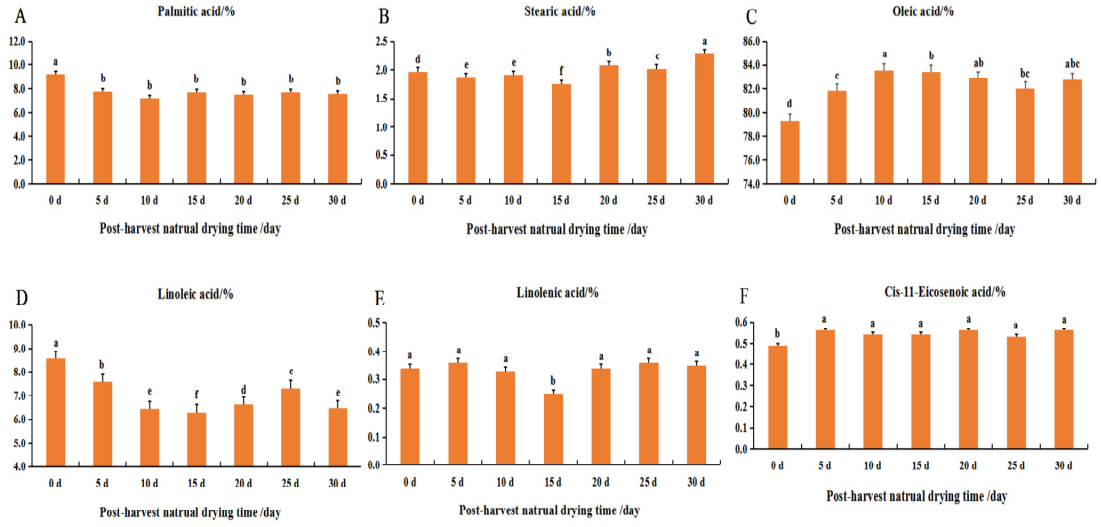

Figure 4. Effects of post-harvest natural drying time on the contents of Palmitic acid (A), stearic acid (B), Oleic acid (C), linoleic acid (D), Linolenic acid (E) and cis-11-eicosenoicacid (F) in *C. oleifera* seeds

Values as shown as means ± SDs of three biological replicates. Different letters between treatments denote significant differences (ANOVA test, $P < 0.05$)

In order to reveal the changing trend of saturated fatty acids and unsaturated fatty acids more intuitively, we conducted a comprehensive analysis of the relative content of palmitic acid with stearic acid, oleic acid, linoleic acid, linolenic acid, and arachidonic acid. The changes of the total content of saturated fatty acids (palmitic acid and stearic acid) and unsaturated fatty acids (oleic acid, linoleic acid, linolenic acid, and arachidonic acid) were shown in Figures 5C and B. The results showed that with the increase of natural drying time, the total content of unsaturated fatty acids significantly increased, while saturated fatty acids were the opposite. Thus, post-harvest natural drying could increase the total content of unsaturated fatty acids, thereby improving the quality of the oil.

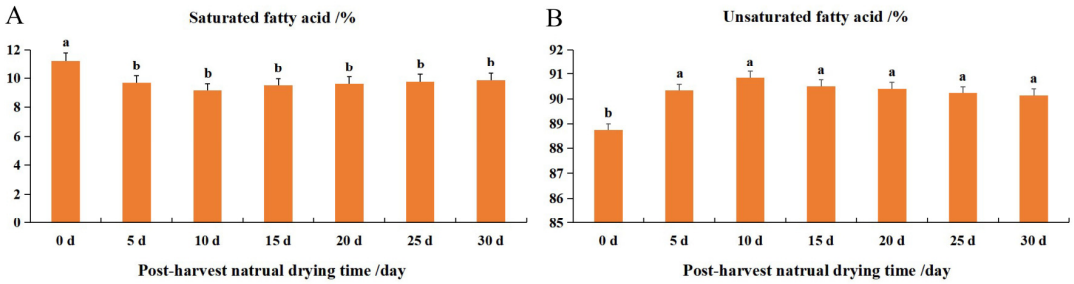

Figure 5. Effect of post-harvest natural drying time on the content of saturated acid (A) and unsaturated acid (B) in *C. oleifera* seeds after harvest

Values as shown as means ± SDs of three biological replicates. Different letters between treatments denote significant differences (ANOVA test, $P < 0.05$)

The change of endogenous hormones contents in C. oleifera seeds after post-harvest natural drying

The concentration of SL, SA, and ACC fluctuated first from 0 to 15 d and then increased with post-harvest drying time from 20 to 30 d. Compared with 0 d, the concentration of SL, SA, and ACC were highest at 30 d, increasing 125.14% (0.58→1.30 ng g^{-1}), 134.80% (13.03→30.60 ng g^{-1}), and 194.90% (1.39→4.09 ng g^{-1}), respectively (Figures 6A-C).

Compared with the control group, BR concentration increased sharply at first, but then decreased, and then increased to 3.26 ng g^{-1} at 20 d. After 20 d, the content of BR decreased gradually and became stable (Figure 6D).

As shown in Figure 6E, the concentration of MeJA significantly increased from 0 to 10 d, reaching a maximum value of 4.53 ng g^{-1}, which increased 1.68-fold compared with 0 d, but then sharply decreased to 15 d (4.53⟶2.03 ng g^{-1}), and then tended to be stable. There was no significant difference between the control group after 30 d and 0 d.

Correlation analysis of nutrients contents and endogenous hormones after post-harvest natural drying

To investigate the relationship between the content of various substances (moisture, soluble sugar and protein, oil, fatty acid components, and endogenous hormone) in *C. oleifera* seeds after post-harvest natural drying, the correlations of them were analyzed. As shown in Figure 7, there was a strong negative correlation between soluble sugar and soluble protein and moisture content (correlation coefficients were -0.80 and -0.88). Soluble protein was negatively correlated with MeJA. Oil content has a significant positive correlation with SL, SA, and ACC (correlation coefficient > 0.8), which correlation coefficients were 0.85, 0.85 and 0.82. Stearic acid showed a strong positive correlation with SL, SA, and ACC. In addition, five endogenous hormones were positively correlated with oleic acid, linolenic acid, cis-11-Eicosenoicacid, and unsaturated fatty acid, but negatively correlated with saturated fatty acid and palmitic acid.

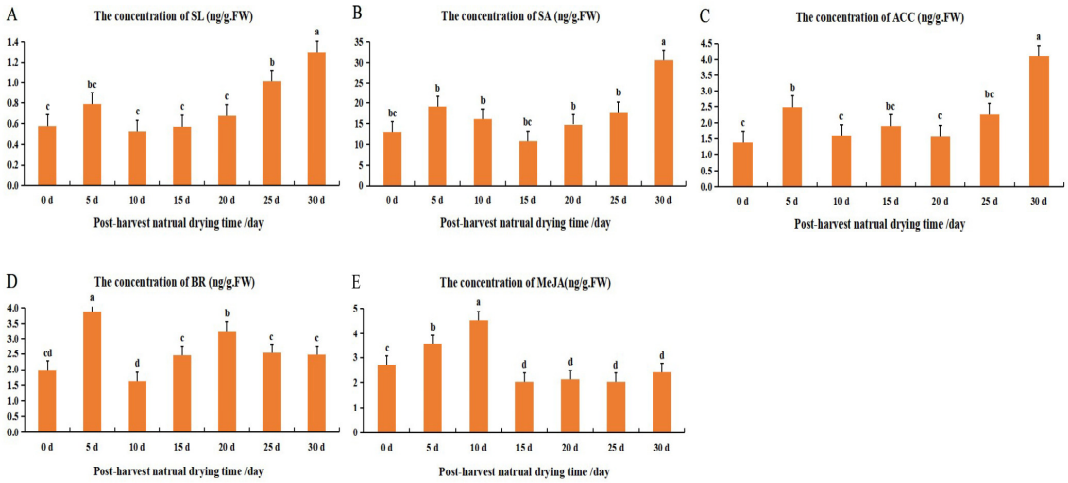

Figure 6. Effects of post-harvest natural drying time on the endogenous hormones contents of SL (A), SA (B), ACC (C), BR (D) and MeJA (E) in *C. oleifera* seeds

Values as shown as means ± SDs of three biological replicates. Different letters between treatments denote significant differences (ANOVA test, $P < 0.05$)

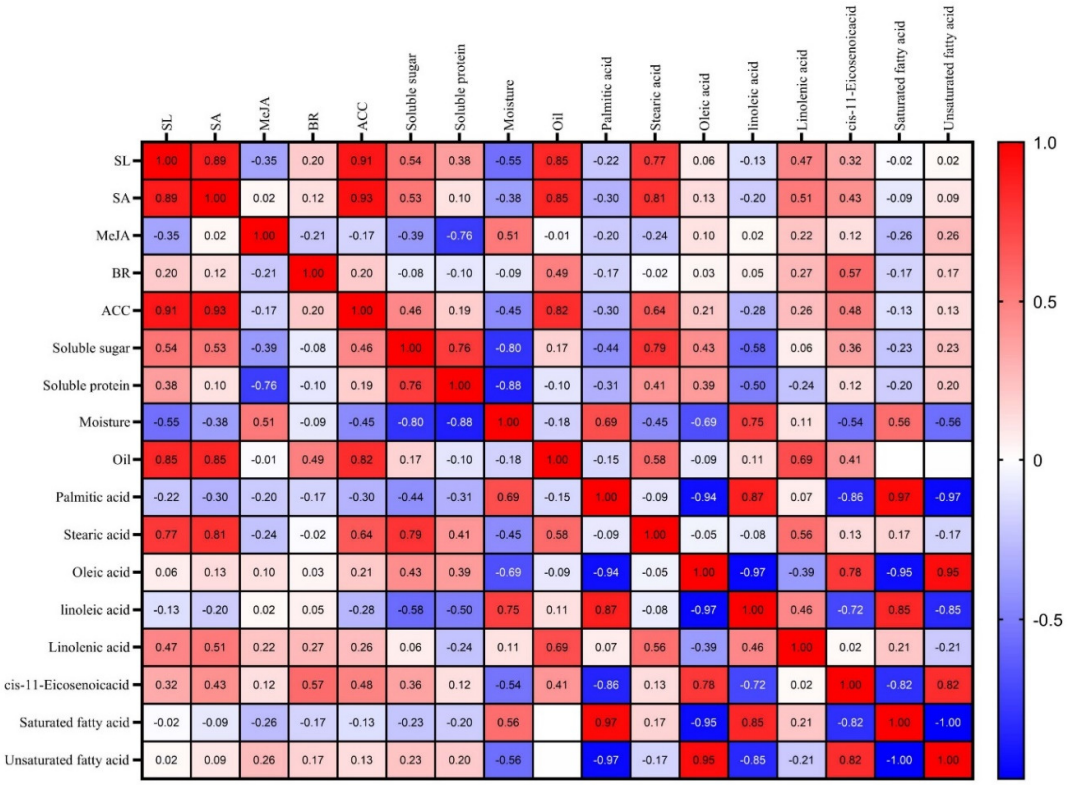

Figure 7. Correlation analysis

Correlation analysis of each substance content, using Pearson method (GraphPad Prism 9 software test)

Discussion

Effects of post-harvest natural drying on oil content and quality in C. oleifera seeds

For oil crops, the accumulation of seed storage materials began to tilt in the direction of oil during post ripening, and post-harvest natural drying will promote this transformation (Borisjuk *et al*., 2004; Cao *et al*., 2015; Eliášová *et al*., 2022; Hernández-Clemente *et al*., 2019; Xu *et al*., 2020). The total oil content increased during the drying of *Cupuaçu* seeds (Souza *et al.*, 2020). In this study, the accumulation of oil was closely related to the increase of drying time and reached the peak value after 30 d of post-harvest natural drying. These findings were consistent with the result of Luo *et al.* (2015). Unsaturated fatty acids are important bioactive ingredient in *C. oleifera* seeds (accounting for more than 90%), which affects the oil quality and is beneficial to the human body (Xiao *et al.*, 2017; Zhou *et al.*, 2020). Many studies indicated that drying treatment not only promoted the conversion of fatty acids but also enhanced the continuous conversion of saturated fatty acids into unsaturated fatty acids in seeds (Curtidor *et al.*, 2020; Khan *et al.*, 2011; López-Fernández *et al.*, 2018; Wang *et al.*, 2021). Wang *et al.* (2020) found that air-drying is more favorable to produce fatty acids. In this study, the unsaturated fatty acids level increased, and finally stabilized from 0 to 30 d natural drying, especially oleic acid accumulation level, while the saturated fatty acids of *C. oleifera* seeds decreased. Our results were consistent with the previous report (Wang *et al*. 2017). In conclusion, the optimal post-harvest natural drying time was 30 d could ensure the oil content and quality of *C. oleifera* oil.

The contains substances content change of C. oleifera seeds under post-harvest natural drying treatments

C. oleifera seeds are the main source of economic benefits because they are rich in a large number of nutrients (sugar, protein, oil, etc.) (Zeng and Endo, 2019). A large number of studies have shown that post-harvest drying will change the content of carbohydrates in fruits (Abano *et al.*, 2021; Marzec *et al.*, 2020). In *dewandaru* fruit, the content of soluble sugar significantly increased with the drying time, especially at 9 h of drying, the sugar content reached the highest level (Asri *et al.*, 2021). In the present study, the content of soluble sugar of *C. oleifera* seeds had the highest level at 30 d of post-harvest natural drying, which was consistent with the report of Brenac *et al.* (1997). Furthermore, Ling and Zheng (2012) reported that the oil content increased but the soluble sugar content gradually decreased during the seed development of *C. oleifera*, which were consistent with our results, that is, the soluble sugar content decreased, but the oil content increased significantly at 20-25 d. At the same time, there is a positive correlation between soluble sugar and stearic acid. These results suggested that soluble sugar would be gradually converted into oil during the drying process of *C. oleifera* seeds.

In addition, the accumulation of sugar and protein can provide a carbon frame and energy for the synthesis and accumulation of fatty acids**Error! Reference source not found.**. Roesler *et al.* (2016) revealed that with the decrease of sucrose and soluble sugar content, the oil content increased by at least three percentage points, and the protein content also increased slightly in soybean seeds. In the present study, the content of soluble sugar increased at a certain rate after post-harvest natural drying. There was an obvious trade-off between oil and soluble sugar. These findings were supported by Song *et al.* (2021).

The change of endogenous hormone content may be due to post-harvest natural drying

Plant endogenous hormones refer to trace organic substances synthesized by plants themselves that can significantly regulate the physiological process of plants even at very low concentrations (Wasternack and Hause, 2013; Nguyen *et al*., 2016). Studies have proved that plant endogenous hormones play important roles in the distribution and regulation of carbon assimilation in seeds **Error! Reference source not found.**Srivastava and Handa, 2005). Moreover, drying treatment had significant effects on the content of endogenous hormones in plants (Chojnowski *et al.*, 1997; Yang *et al.*, 2008). Adams and Yang (1979) reported

that ACC is a precursor of ethylene and the ACC content exhibited the change of ethylene level (Yang and Hoffman, 2003; Pech *et al*., 2010). The research by Chojnowski *et al.* (1977) indicated that drying enhanced the ability of ACC to convert to ethylene. Ethylene promotes oil content and the process of conversion of saturated fatty acids to unsaturated fatty acids in *C. oleifera* seeds (Bhushan and Mandava, 1988). In this study, the ACC content of *C. oleifera* seeds significantly increased after 20 d, and ACC content was positively correlated with oil content, which coincided with the reports by Adams and Yang (1979). These results suggested that the treatment of natural drying after harvest promoted the transformation of ACC in *C. oleifera* seeds, thereby improving the oil content and unsaturated fatty acid content. SL is a group of carotenoid-derived plant hormones that play a key role in plant growth and development (Crawford *et al*., 2010). Lee and Yoon (2020) found that SL could promote the synthesis of ethylene. In this study, the content of SL was significantly positively correlated with ACC level. These results suggested that SL affected substance transformation by promoting the increase of ACC. SA is a phenolic compound, which is involved in regulating plant growth and development (Raskin, 1992). Yu *et al*. (2021) mentioned that SA and ethylene synergistically promoted leaf senescence in *Arabidopsis*. In this study, there was a positive correlation between the SA and ACC and oil content, which were consistent with the reported by Yu *et al*. (2021). Altogether, the content of endogenous hormones of SL, SA, and ACC mainly changed after post-harvest natural drying, and these three endogenous hormones could affect the transformation of substances in *C. oleifera* seeds.

Conclusions

In this study, the content changes of soluble sugar, soluble protein, moisture, oil, and endogenous hormones of SL, SA, ACC, BR, and MeJA of *C. oleifera* seeds under the different post-harvest natural drying were analyzed. The results suggested that the moisture content decreased with the increase of natural drying days, leading to the promotion of post-ripening process. On 30 d, the contents of soluble sugar and oil in *C. oleifera* seeds reached the highest, which soluble sugar content reaching 156.2 mg g^{-1} (30 d) and the oil content reaching 23.6%. The soluble protein reached the highest on 20 d (13.9mg g^{-1}), which was 1.54-fold higher than the control group (9.03 mg g^{-1}) and changed little, and unsaturated fatty acids such as oleic acid reached the peak and tended to be stable on 30 d. Therefore, drying for 30 d after picking can increase the contents of soluble sugar, soluble protein, oil and unsaturated fatty acid of *C. oleifera*. In addition, the endogenous hormones SL, SA, and ACC were positively correlated with the oil content (correlation coefficient > 0.8), and these three hormones might promote the formation of oil. Therefore, the balance between endogenous hormones after 30 d of post-harvest natural drying is most conducive to the accumulation of large amounts of oil. In conclusion, natural drying 30 d after harvests is an effective method to improve the oil content and oil quality of *C. oleifera* seeds.

Authors' Contributions

All authors participated in planning this paper. Conceptualization (XY and QS); Data curation (YL and YZ); Formal analysis (RX and QS); Methodology (GH); Software (KJ and LC); Writing-original draft (XY and QS); Writing-review and editing (WG); Material supply (DY). All authors read and approved the final manuscript.

Ethical approval (for researches involving animals or humans)

Not applicable.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

References

Abano EE, Akanson J, Kizzie-Hayford N (2021). Drying kinetics and quality of whole, halved, and pulverized tiger nut tubers (*Cyperus esculentus*). International Journal of Food Science 2021:8870001. *https://doi.org/10.1155/2021/8870001*

Adams DO, Yang SF (1979). Ethylene biosynthesis: identification of 1-aminocyclopropane-1-carboxylic acid as an intermediate in the conversion of methionine to ethylene. Proceedings of the National Academy of Sciences 76(1):170-174. *https://doi.org/10.1073/pnas.76.1.170*

Ali T (2018). Effect of drying methods on nut quality of hazelnuts (*Corylus avellana* l.). Journal of Food Science & Technology 55:4554-4565. *https://doi.org/10.1007/s13197-018-3391-8*

Asri NKT, Devi M, Soekopitojo S (2021). Effect of drying on antioxidant capacity, sugar content, water content, physical and organoleptic properties of dried candied dewandaru fruit (*Eugeia uniflora* L.). Journal of Physics: Conference Series 1882(1): 012110 (6pp). *https://iopscience.iop.org/article/10.1088/1742-6596/1882/1/012110*

Baud S, Boutin JP, Miquel M, Lepiniec L, Rochat C (2002). An integrated overview of seed development in *arabidopsis thaliana* ecotype ws. Plant Physiology & Biochemistry 40(2):151-160. *https://doi.org/10.1016/S0981-9428(01)01350-X*

Bhushan N, Mandava (1988). Plant growth-promoting brassinosteroids. Annual review of plant Physiology & Plant Molecular Biology 39(1):23-52. *https://doi.org/10.1146/annurev.pp.39.060188.000323*

Borek S, Pukacka S, Michalski K, Ratajczak L (2009). Lipid and protein accumulation in developing seeds of three lupine species: *Lupinus luteus* L. *Lupinus albus* L. and *Lupinus mutabilis* sweet. Journal of Experimental Botany 60(12):3453-3466. *https://doi.org/10.1093/jxb/erp186*

Borisjuk L, Rolletschek H, Radchuk R, Weschke W, Wobus U, Weber H (2004). Seed development and differentiation: a role for metabolic regulation. Plant Biology 6(4):375-386. *https://doi.org/10.1055/s-2004-817908*

Bradford MM (1976). A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Analytical Biochemistry 72(1-2):248-254. *https://doi.org/10.1016/0003-2697(76)90527-3*

Brenac P, Horbowicz M, Downer SM, Alexandra (1997). Raffinose accumulation related to desiccation tolerance during maize (*Zea mays* L.) seed development and maturation. Journal of Plant Physiology 150(4):481-488. *https://doi.org/10.1016/S0176-1617(97)80102-2*

Cao YQ, Yao XH, Ren HD, Wang KL (2015). Changes in contents of endogenous hormones and main mineral elements in oil-tea *Camellia* fruit during maturation]. Journal of Beijing Forestry University 37(11):76-81. [in Chinese] *https://www.cnki.net/kcms/doi/10.13332/j.1000-1522.20140407*

Chojnowski M, Corbineau F, D Côme (1997). Physiological and biochemical changes induced in sunflower seeds by osmopriming and subsequent drying, storage, and aging. Seed Science Research 7(4):323-332. *https://doi.org/10.1017/S096025850000372X*

Crawford S, Shinohara N, Sieberer T, Williamson L, George G, Hepworth J, ... Leyser O (2010). Strigolactones enhance competition between shoot branches by dampening auxin transport. Development (Cambridge, England) 137(17):2905. *https://doi.org/1 0.1242/dev.051987*

Curtidor CM, Annunziata MG, Gupta S, Apelt F, Richard SI, KAragler F, ... Olas JJ (2020). Physiological profiling of embryos and dormant seeds in two *Arabidopsis* accessions reveals a metabolic switch in carbon reserve accumulation. Frontiers in Plant Science 11:588433. *https://doi.org/10.3389/fpls.2020.588433*

Di TM, Yang SL, Du FY, Zhao L. Xia T, Zhang XF (2017). Cytotoxic and hypoglycemic activity of triterpenoid saponins from *Camellia oleifera* Abel. Seed Pomace. Molecules (Basel, Switzerland) 22(10):1562. *https://doi.org/10.3390/molecules22101562*

Eliášová K, Konrádová H, Dobrev PI, Motyka V, Lomenech AM, Fischerová L, ... Teyssier C (2022). Desiccation as a post-maturation treatment helps complete maturation of Norway spruce somatic embryos: carbohydrates, phytohormones and proteomic status. Frontiers in Plant Science 13:823617. *https://doi.org/10.3389/fpls.2022.823617*

Feng JL, Yang ZJ, Chen SP, El-Kassaby A, Chen H (2017). High throughput sequencing of small rnas reveals dynamic micrornas expression of lipid metabolism during *Camellia oleifera* and *C. meiocarpa* seed natural drying. BMC Genomics [electronic resource] 18(1):546. *https://doi.org/10.1186/s12864-017-3923-z*

Gao QH, Wang M (2013). Effect of drying of jujubes (Ziziphus jujuba mill.) on the contents of sugars, organic acids, α-tocopherol, β-carotene, and phenolic compounds. Journal of Agricultural & Food Chemistry 61(19):4665-4665. *https://doi.org/10.1021/jf400098v*

Gao W, He XS, Sun Y, Zhan ZY, Huang JJ, Lei XL (2013). Effects of different harvest methods on oil content in *Camellia oleifera*. Nonwood Forest Research 31(04):177-181. [in Chinese]

Gong WF, Song QL, Ji K, Gong SF, Wang LK, Chen L, ... Yuan DY (2020). Full-length transcriptome from *Camellia oleifera* seed provides insight into the transcript variants involved in oil biosynthesis. Journal of Agricultural & Food Chemistry 68 (49):14670-14683. *https://doi.org/10.1021/acs.jafc.0c05381*

Grosso NR, Guzmán CA (2010). Lipid, protein, and ash contents, and fatty acid and sterol compositions of peanut (*Arachis hypogaea* L.) seeds from Ecuador. Peanut Science 22(2):84-89.

Hernández-Clemente R, Hornero A, Mottus M, Penuelas J, González-Dugo V, Jiménez JC... Zarco-Tejada PJ (2019). Early diagnosis of vegetation health from high-resolution hyperspectral and thermal imagery: lessons learned from empirical relationships and radiative transfer modelling. Current Forestry Reports 5:169-183. *https://doi.org/10.1007/s40725-019-00096-1*

Huang AY, Wu SC, Gu WH, Li YX, Xie XJ, Wang GC (2019). Provision of carbon skeleton for lipid synthesis from the breakdown of intracellular protein and soluble sugar in *Phaeodactylum tricornutum* under high CO_2. BMC Biotechnology [electronic resource] 19(1):53-60. *https://doi.org/10.1186/s12896-019-0544-4*

Jiang QY, Zhang M, Arun SM (2020). UV induced conversion during drying of ergosterol to vitamin d in various mushrooms: effect of different drying conditions. Trends in Food Science & Technology 105:200-210. *https://doi.org/10.1016/j.tifs.2020.09.011*

Jing DL, Zhang JW, Xia Y, Kong LS, OuYang FQ, Zhang SG, ... Wang JH (2017). Proteomic analysis of stress-related proteins and metabolic pathways in *Picea asperata* somatic embryos during partial desiccation. Plant Biotechnology Journal 15(1):27-38. *https://doi.org/10.1111/pbi.12588*

John A, Barnett G, Miller TB (1950). The determination of soluble carbohydrate in dried samples of grass silage by the anthrone method. Journal of the Science of Food & Agriculture 1(11):336-339. *https://doi.org/10.1002/jsfa.2740011106*

Kende H (1993). Ethylene biosynthesis. Annual Review of Plant Physiology & Plant Molecular Biology 44:283-307. *https://doi.org/10.1146/annurev.pp.44.060193.001435*

Khan NA, Cone JW, Pellikaan WF, Khan MA, Struik PC, Hendriks WH (2011). Changes in fatty acid content and composition in silage maize during grain filling. Journal of the Science of Food & Agriculture 91(6):1041-1049. *https://doi.org/10.1002/jsfa.4279*

Khavari M, Fatahi R, Zamani Z (2021). Salicylic acid and kaolin effects on pomological, physiological, and phytochemical characters of hazelnut (*Corylus avellana*) at warm summer condition. Scientific Reports 11(1):4568. *https://doi.org/10.1038/s41598-021-83790-0*

Lee CP, Yen GC (2006). Antioxidant activity and bioactive compounds of tea seed (*Camellia oleifera* Abel.) oil. Journal of Agricultural & Food Chemistry 54(3):779-784. *https://doi.org/10.1021/jf052325a*

Lee HY, Yoon GM (2020). Strigolactone elevates ethylene biosynthesis in etiolated *Arabidopsis* seedlings. Plant Signaling & Behavior 15(11):1805232. *https://doi.org/10.1080/15592324.2020.1805232*

Lei J, Wang T, Han C, Chen L, Hao Q (2013). The effect of different drying methods on the quality of white seedless raisins. Xinjiang Agricultural Sciences 50(2):254-259.

Ling HY, Zheng DY (2012). Changes of the functional components in *Camellia oleifera* seed during mature process. Journal of Fujian College of Forestry 32(1):89-92. [in Chinese] *http://qikan.cqvip.com/Qikan/Article/Detail?id=40980811*

López-Fernández MP, Moyano L, Correa MD, Vasile F, Burrieza HP, Maldonado S (2018). Deterioration of willow seeds during storage. Scientific Reports 8(1):17207. *https://doi.org/10.1038/s41598-018-35476-3*

Luo F, Fei X, Guo S, Chen J, Li KX (2015). Effects of harvest time and drying methods on quality of oil-tea *Camellia* seed oil. China Oils and Fats 40(11):69-73. [in Chinese]

Ma JL, Hang Y, Rui YK, Chen GC, Zhang NY (2011). Fatty acid composition of *Camellia oleifera* oil. Journal Für Verbraucherschutz und Lebensmittelsicherheit 6(1):9-12. *https://doi.org/10.1007/s00003-010-0581-3*

Marzec A, Kowalska H, Kowalska J, Domian E, Lenart A (2020). Influence of pear variety and drying methods on the quality of dried fruit. Molecules (Basel, Switzerland) 25(21):5146. *https://doi.org/10.3390/molecules25215146*

Mohammed HA, Al-Omar MS, Mohammed S, Aly M, Alsuqub A, Khan RA (2020). Drying induced impact on composition and oil quality of rosemary herb, *Rosmarinus Officinalis* Linn. Molecules (Basel, Switzerland) 25(12):2830. *https://doi.org/10.3390/molecules25122830*

Mohammed S, Edna M, Siraj K (2020). The effect of traditional and improved solar drying methods on the sensory quality and nutritional composition of fruits: a case of mangoes and pineapples - sciencedirect. Heliyon 6(6):e04163. *https://doi.org/10.1016/j.heliyon.2020.e04163*

National health and family planning commission of the people's republic of China. (2016). National food safety standard, determination of fat in foods: GB 5009.6-2016[S]. Standards press of China, 2017, Beijing. [in Chinese]

Nguyen QT, Kisiala A, Andreas P, Neil Emery RJ, Narine S (2016). Soybean seed development: fatty acid and phytohormone metabolism and their interactions. Current Genomics 17(3):241-260. *https://doi.org/10.2174/1389202917666160202220238*

Ojediran JO, Okonkwo CE, Olaniran AF, Iranloye YM, Adewumi AD, Erinle O, ... Adeyi A (2021). Hot air convective drying of hog plum fruit (Spondias mombin): effects of physical and edible-oil-aided chemical pretreatments on drying and quality characteristics. Heliyon 7(11):e08312. *https://doi.org/10.1016/j.heliyon.2021.e08312*

Pech JC, Latché A, Bouzayen M (2010). Ethylene Biosynthesis. In: Davies PJ (Ed). Plant Hormones. Springer, Dordrecht, pp115-136. *https://doi.org/10.1007/978-1-4020-2686-7_6*

Peng SF, Lu J, Zhang, Z, Ma L, Liu CX, Chen YZ (2020). Global transcriptome and correlation analysis reveal cultivar-specific molecular signatures associated with fruit development and fatty acid determination in *Camellia oleifera* Abel. International Journal of Genomics 2020:6162802. *https://doi.org/10.1155/2020/6162802*

Qin SY, Rong J, Zhang WJ, Chen JK (2018). Cultivation history of *Camellia oleifera* and genetic resources in the Yangtze River basin. Biodiversity Science 26(4):384-395. *https://www.biodiversity.science.net/EN/10.17520/biods.2017254*

Raskin I (1992). Role of salicylic acid in plants. Annual Review of Plant Physiology & Plant Molecular Biology 43:439-463. *https://doi.org/10.1146/annurev.pp.43.060192.002255*

Roesler K, Shen B, Bermudez E, Li CJ, Hunt J, Damude HG Meyer K (2016). An improved variant of soybean type 1 diacylglycerol acyltransferase increases the oil content and decreases the soluble carbohydrate content of soybeans. Plant Physiology 171(2):878-93. *https://doi.org/10.1104/pp.16.00315*

Song QL, Ji K, Mo WJ, Wang LK, Chen L, Gao L, ... Yuan DY (2021). Dynamics of sugars, endogenous hormones, and oil content during the development of *Camellia oleifera* Abel. fruit. Botany 99(8):515-529. *https://doi.org/10.1139/cjb-2021-0019*

Souza JML, Rocha JM, Cartaxo CBC, Vasconcelos MAM, Álvares VS, Nascimento MM, ... Kaefer S (2020). Monitoring and optimization of *Cupuaçu* seed fermentation, drying and storage processes. Microorganisms 8(9):1314. *https://doi.org/10.3390/microorganisms8091314*

Srivastava A, Handa AK (2005). Hormonal regulation of tomato fruit development: a molecular perspective. Journal of Plant Growth Regulation 24**:**67-82. *https://doi.org/10.1007/s00344-005-0015-0*

Wang D, Javed HU, Shi Y, Naz, S, Duan CQ (2020). Impact of drying method on the evaluation of fatty acids and their derived volatile compounds in '*thompson seedless*' raisins. Molecules 25(3):1-13. *https://doi.org/10.3390/molecules25030608*

Wang HO, Hu ZC, Chen SJ, Fu QQ, Zhang W, Wang RR, Xie HX (2017). Effects of different harvesting dates and drying methods on peanut quality. Transactions of the Chinese Society of Agricultural Engineering 33(22):292-300.

Wang YP, Yao XH, Yang L, Fei XQ, Cao YQ, Guo SH (2021). Effects of harvest time on the yield, quality and active substance of Torreya grandis nut and its oil. Journal of Oleo Science 70(2):175-184 *https://doi.org/10.5650/jos.ess20155*

Wasternack C, Hause B. (2013). Jasmonates: biosynthesis, perception, signal transduction and action in plant stress response, growth and development. an update to the 2007 review in *annals of botany*. Annals of Botany London Oup Then Academic Press Then Oxford University Press 111(6):1021-1058.

Wen Y, Su SC, Ma LY, Yang SY, Wang YW, Wang XN (2018). Effects of canopy microclimate on fruit yield and quality of *Camellia oleifera* - sciencedirect. Scientia Horticulturae 235:132-141. *https://doi.org/10.1016/j.scienta.2017.12.042*

Wu B, Ruan CJ, Shah AH, Li DH, Li H, Ding J, ... Du W (2022). Identification of miRNA–mRNA regulatory modules involved in lipid metabolism and seed development in a woody oil tree (*Camellia oleifera*). Cells 11(1):71 *https://doi.org/10.3390/cells11010071*

Wu LL. Li JA, Li Z, Zhang FH, Tan XF (2020). Transcriptomic analyses of *Camellia oleifera* 'Huaxin' leaf reveal candidate genes related to long-term cold stress. International Journal of Molecular Sciences 21(3):846. *https://doi.org/10.3390/ijms21030846*

Xiao XM, He LM, Chen YY, Wu LH, Wang L, Liu ZP (2017). Anti-inflammatory and antioxidative effects of *Camellia oleifera* Abel components. Future Medicinal Chemistry 9(17):2069-2079. *https://doi.org/10.4155/fmc-2017-0109*

Xie XJ, Gao S, Gu WH, Pan GH, Wang GC (2013). Desiccation induces accumulations of antheraxanthin and zeaxanthin in intertidal macro-alga *Ulva Pertusa* (Chlorophyta). PloS One 8(9)e72929. *https://doi.org/10.1371/journal.pone.0072929*

Xu PL, Tang GY, Cui WP, Chen GX, Ma CL, Zhu JQ, ... Wan SB (2020). Transcriptional differences in peanut (*Arachis hypogaea* L.) seeds at the freshly harvested, after-ripening and newly germinated seed stages: Insights into the regulatory networks of seed dormancy release and germination. PloS One 15(1):e0219413. *https://doi.org/10.1371/journal.pone.0219413*

Yang SF, Hoffman NE (2003). Ethylene biosynthesis and its regulation in higher plants. Annual Review of Plant Biology 35(1):155-189. *https://doi.org/10.1146/annurev.pp.35.060184.001103*

Yang, L, Suen HL, Liang LD, Liu CP (2008). Changes in endogenous hormone content in seeds of *Sorbus pohuashanensis* (hance) Hedl. during artificial drying and cold stratification. Plant Physiology Communications 44(4):682-688. [in Chinese]

Yu XD, Xu YR, Yan SP (2021). Salicylic acid and ethylene coordinately promote leaf senescence. Journal of Integrative Plant Biology 63(5):823-827. *https://doi.org/10.1111/jipb.13074*

Zeng W, Endo Y. (2019). Effects of cultivars and geography in China on the lipid characteristics of *Camellia oleifera* seeds. Journal of Oleo Science 68(11):1051-1061. *https://doi.org/10.5650/jos.ess19154*

Zhang FH, Li Z, Zhou JQ, Gu YY, Tan XF (2021). Comparative study on fruit development and oil synthesis in two cultivars of *Camellia oleifera*. BMC Plant Biology *21*(1):348. *https://doi.org/10.1186/s12870-021-03114-2*

Zhong WQ, Shen JL, Liao XD, Liu XL, Zhang, J, Zhou CH, Jin YC (2019). Camellia (*Camellia oleifera* Abel.) seed oil promotes milk fat and protein synthesis-related gene expression in bovine mammary epithelial cells. Food Science & Nutrition 8(1):419-427. *https://doi.org/10.1002/fsn3.1326*