



## Physicochemical properties, profile of volatiles, fatty acids, lipids and concomitants from four *Kadsura coccinea* seed oils

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### ABSTRACT

The *Kadsura coccinea* fruit is a wild fruit that may be eaten and used medicinally. Its seeds are rich in nutrients but are typically thrown away without processing. The physicochemical characterization, volatiles, fatty acids, lipids and concomitants of cold-processed seed oils from four kinds of *K. coccinea* were evaluated. The average kernel yield and oil yield of *K. coccinea* seeds were 68.21 % and 30.44 %, respectively. The seed oil contains a moderate level of total phenolics (368.99–503.99 mgGAE/100 g), total flavonoids (95.01–126.18 mg RE/100 g), and  $\beta$ -sitosterol (1498.8–1712.7 mg/kg) with higher iodine value, lower acid value, saponification value and shorter induction time. GC analysis reveals appreciable amounts of linoleic acid (64.91–68.05 %) and squalene in seed oil. GC–MS analysis showed that the major volatile compounds were  $\gamma$ -muurolene (27.25–31.7 %),  $\beta$ -himachalene (19.51–20.37 %) and  $\beta$ -curcumene (15.78–16.78 %). Moreover, 16 terpenoids, 14 phenolics were identified by UPLC-QTOF-MS/MS. These results suggest that *K. coccinea* seed seems an promising alternative oilseed with biological ingredients for food, cosmetics and pharmaceutical industries.

### 1. Introduction

*Kadsura coccinea* (Lem.) A.C. Smith (*K. coccinea*), also called Lengfantuan belongs to the family *Schisandraceae*, grows in mountain forests with an altitude of 1500–2400 m and is widely distributed in southern China, Vietnam, Thailand, etc. It is mainly composed of five parts: root, stem, leaf, flower and fruit, the roots and vines can be used as medicine, which can dispel wind and activate collaterals, regulate qi and relieve pain (Shi & Chen, 2013). The ripe fruit is red and consists of approximately 30 to 65 small berries, each berry contains three to six seeds, which are bitter and have thick shells and account for about 10 % of the *K. coccinea* berries (Liu et al., 2014). Each small berry has a fruity aroma similar to that of an apple, with a skin and pulp color similar to that of a

lychee. They are rich in amino acids, vitamin C, minerals and anthocyanins. These fruits have been used by ethnic minorities such as the Miao and Zhuang groups as a beneficial fruit and traditional folk medicine to treat gastritis (Li et al., 2022). In addition, *K. coccinea* contains lignans, triterpenes, sesquiterpenes, steroids, amino acids and other effective ingredients with anti-tumor, anti-HIV, anti-inflammatory, liver protective, antioxidant functions and other bioactivity (Li et al., 2020). It is a green cash crop with edible, medicinal and ornamental values. The *K. coccinea* fruit seeds exist in a small aggregate fruit; each cluster contains 3–5 seeds. The fruit seeds are composed of exotesta, endocarp and kernel. The exotesta is a thin layer of wood, which is fragile and peeled easily. The endocarp is a brown film close to the kernel and difficult to peel. Apart from the production of seedlings, the seed is

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generally discarded as waste. Whereas, the *K. coccinea* seeds are rich in nutrients such as protein, a variety of trace elements, polyphenols, sterols, vitamins, and other active ingredients with free radical scavenging activity, bacteriostasis and enzyme inhibition activity (Wei et al., 2016). Similar bioactive compounds were also found in much seed oil and attracted the attention of many researchers (Sun et al., 2024; Wang et al., 2023). However, the research information about the bioactive components of *K. coccinea* seeds are limited, such as fatty acid composition, phenols and other trace nutrients. As far as we know, there is no information on the comparative analysis of physicochemical properties and bioactive components of different undeveloped *K. coccinea* seed oils.

In this investigation, the seeds sourced from four main varieties of *K. coccinea* that are recognized for geographical indication products in China are used as the research object. The study's scope encompasses a systematic comparative analysis of these seeds' nutritional profiles, the physicochemical properties of the derived seed oil, the composition of fatty acids, volatile components, lipid concomitants, and other functional components. This study is expected to provide experimental data for the development and utilization of its seeds, and breeding of different varieties of *K. coccinea*.

## 2. Materials and methods

### 2.1. Plant materials

Four varieties of ripe fruit Dahong, Zihei, Fenhong, Hulu were obtained from Tongdao County, Hunan Province. These fruits were identified by Professor Lizhonghai in the College of Food Science and Engineering, and their voucher samples (No.DH202012 for Dahong; No.ZH202012 for Zihei; No.FH202012 for Fenhong; No. HL202012 for Hulu) were deposited in our laboratory of Central South University of Forestry and Technology. Their seeds were manually peeled from sarcocarp, and then cleaned with water, followed by dried in the shade, and stored in a black plastic bag at room temperature until use.

### 2.2. Chemical and reagents

Gallic acid, methanol, sodium carbonate, potassium hydroxide, toluene and n-hexane (analytical grade) was procured from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China). Folin-ciocalteu (analytical grade) was obtained from Shanghai Yuanye Biotechnology Co., LTD. The HPLC grade solvents (acetonitrile, methanol) were purchased from Merck (Darmstadt, Germany). HPLC grade formic acid and ammonium acetate were obtained from AMPO CNW (Shanghai, China).

### 2.3. The preparation of pressed *K.Coccinea* seed oil

Firstly, the shells of *K.coccinea* seeds were manually peeled to get the kernels, then the kernels were put into the SUN-2 oil press for pressing at room temperature to get the crude oil. Next, the *K. coccinea* seed oil was obtained after centrifuging the crude oil at 4000 rpm and 8000 rpm for 15 min. The kernel yield and oil yield of *K.coccinea* seed oil were calculated according to the following equation.

$$\text{Kernel yield} / \% = \frac{\text{kernel weight}}{\text{seed mass}} \times 100 \quad (1)$$

$$\text{Oil yield} / \% = \frac{\text{oil weight}}{\text{kernel weight}} \times 100 \quad (2)$$

### 2.4. Fat, protein, ash and minerals of *K.Coccinea* seed

The fat, crude protein, ash and minerals content of the *K.coccinea* seeds were determined according to the methods of AOAC (2016).

## 2.5. Physicochemical properties of *K.Coccinea* seed oil

### 2.5.1. Color determination of *K.Coccinea* seed oil

Color measurements were performed by an automatic multi-function colorimeter (UltraScan PRO, HunterLab, USA). Use the oil sample without dilution and pour it into a 10 mm quartz cuvette. In the tricolor coordinate system, the L \* value is a measure of brightness, ranging from 0 (black) to +100 (white), a \* value ranging from -100 (green) to +100 (red), and b \* value ranging from -100 (blue) to +100 (yellow). The chromaticity value data is the average value of the measurement results from different positions of the sample.

### 2.5.2. Physicochemical properties analysis of *K.Coccinea* seed oil

The chemical properties of *K.coccinea* seeds, such as acid value, peroxide value, iodine value, and saponification value, were analyzed by the methods of AOAC (2016). The oxidation induction period was determined by 743 Rancimat oxidation stability teste (Metrohm, China), and the specific steps were appropriately modified according to Morelló et al. (2004). Take an appropriate oil sample and put it into the Rancimat oxidation stability tester. Set the heating temperature to 110 °C and the airflow rate to 20 L/h to accelerate the oxidation of oil and fat and generate volatile organic acids. The air will bring volatile organic acids into the conductivity chamber, and the deionized water in the conductivity chamber will dissolve volatile organic acids, ionize ions and cause changes in the conductivity of the deionized water. At the same time, the computer records the induction time of accelerated oxidation of oil and fat, and the test results are expressed in h.

### 2.6. Detection of volatile components in *K.Coccinea* seed oil

Using a headspace solid phase microextraction (HS-SPME) in conjunction with a QP-2010 Plus chromatograph mass spectrometer, the volatile components of *K. coccinea* seed oil were identified. The method conditions were referred to Yang et al. (2021) with slight modification. Briefly, *K. coccinea* seed oil (1.0 g) was poured into a 20 mL headspace vial before the extraction head (Supelco 50/30 m DVB/CAR/PDMS) was inserted. The extraction head was then held at 60 °C for equilibrium (30 min), followed by 5 min of desorption. Following were the gas chromatography conditions: HP-5MS column, temperature program: 35 °C for oven, 250 °C for injection, a split injection mode, split ratio of 80:1. The column temperature was first set at 35 °C and increased to 100 °C at a rate of 5 °C per min, holding that temperature for 0.1 min. The temperature was then raised by 10 °C/min for an additional 5 min to 158 °C. After that, it was elevated to 230 °C at a rate of 15 °C/min and maintained there for 2.0 min. The following were the mass spectrometry conditions: In full scan mode with a *m/z* range of 40 to 500, the ion source and interface temperatures were 230 °C and 250 °C, respectively. By comparing the compounds to the NIST11 database, those that had matching degrees above 80 were chosen, and the relative concentration of each component was then determined using the peak area normalization method.

### 2.7. Detection of fatty acid composition of *K.Coccinea* seed oil

Fatty acid composition was studied according to the method of Xu et al. (2022). GC 2030 equipment (Shimadzu, Japan) with a split/splitless injector was used to analyze the fatty acid composition of *K. coccinea* seed oil. Specifically, a 5 mL test tube was filled with 60 mg of oil, 4 mL of n-hexane, and 0.2 mL of a 2 mol/L potassium hydroxide-methanol solution. The test tube was then shaken rapidly for 30 s and left motionless until the combined solution turned clear. Then, 1 g of sodium bisulfate was added to the sample solution to shaking vigorously for 30 s and standing still until the mixed solution became clear. The supernatant was collected for gas chromatography analysis using a FID detector on a GC-2030 column with an SP-2560 column (100 m × 0.25 mm × 0.20 m) (Shimadzu, Japan). The column temperature was

planned to rise from 120 °C to 230 °C at 3 °C/min, then kept at 120 °C for 3 min and 230 °C for 10 min before rising to 235 at 5 °C/min and remaining at 235 °C for 28 min. The inlet temperature was kept at 230 °C, the detector temperature was kept at 250 °C, and the split ratio was 40:1. Inlet carrier gas was high quality nitrogen at a flow rate of 1.0 mL/min. Tail blowing, air, and hydrogen are 30, 400, and 40 respectively, quantified by the area normalization method.

## 2.8. Detection of lipids and other concomitants of *K.Coccinea* seed oil based on UPLC-QTOF-MS/MS

The UPLC-QTOF-MS/MS settings were created using the techniques described by Fan et al. (2018). Put 200 µL of *K.coccinea* seed oil into the centrifuge tube, add 600 µL of methanol and mix thoroughly. After vacuum extraction of methanol, add 100 µL of 50 % methanol aqueous solution and centrifuge for 15 min under 17,000 g. Use an UltiMate 3000 ultra-high-performance liquid chromatography (THERMO) in conjunction with a 5600 quadrupole time-of-flight mass spectrometer (AB SCIEX) to analyze the supernatant as the detection liquid and identify the multicomponent characterization and metabolomics of the *K. coccinea* seed oils. To be more precise, the chromatography conditions were column temperature of 40 °C, sample loading of 3 µL, and positive and negative ions were used to detect the samples. Positive ion mode: 0.1 % formic acid water (A), 0.1 % formic acid acetonitrile (B); Negative ion mode: A: water (2 mmol/L ammonium acetate), B: acetonitrile, mobile phase flow rate of 0.4 mL/min, elution gradient as follows: 0–1.5 min 5–5 % (B), 1.5–2.5 min 5–10 % (B), 2.5–14 min 10–40 % (B), 14–22 min 40–95 % (B), 22–25 min 95–95 % (B), 25–26 min 95–5 % (B), 26–30 min 5–5 % (B). The primary acquisition range for mass spectrometry was 50–1200, the bombardment energy was 30 eV, and 10 secondary spectra were obtained every 50 ms. The atomizing air pressure (GS1), auxiliary air pressure (60 Psi), air curtain air pressure (35 Psi), temperature (650 °C), and spray voltage (5000 V (positive ion mode) or – 4000 V (negative ion mode) model) are the settings for the ESI ion source. Data analysis was employed with the software Analyst TF 1.7. AB Sciex. Firstly, the gathered data were converted into abf format by AnalysisBaseFileConverter, and then, MSDIAL ver 4.6 software was used to perform data processing such as peak search and peak alignment on the converted abf files. Meanwhile, the databases of Metlin, MassBank, MoNA and HMDB were independently integrated based on the first-level and second-level map search (version V6.0).

## 2.9. Detection of sterol content in *K.Coccinea* seed oil

Based on the reference provided by Xiong et al. (2019), the sterol content of *K. coccinea* seed oil was calculated. In a 50 mL test tube, add 25 mL of 2 mol/L KOH-C<sub>2</sub>H<sub>5</sub>OH solutions and 5 g of *K. coccinea* seed oil. After 60 min of saponification at 75 °C in a water bath kettle, add 5 mL of distilled water for chilling, then complete vortex extraction with 5 mL of n-hexane. The upper organic phase was collected to centrifuge at 3000 r/min for 20 min to obtain the supernatant to be analyzed by Shimadzu GC-MS-QP2010 equipped with Agilent HP-5MS UI capillary column (30 m × 0.250 mm × 0.25 µm). A 300 °C inlet temperature was maintained. The temperature of the column was programmed to rise from 120 to 300 °C at a rate of 12 °C/min and to stay there for 20 min. High purity helium was used as inlet carrier gas and a flow rate of 1 mL/min. Each sample was injected in a volume of 1.0 mL utilizing the splitless mode, with a 7-min solvent delay. The following settings were applied to the mass spectrometry: 230 °C for the ion source, 70 eV for the ionization voltage and the EI ionization source, 300 °C for the transmission line and 150 °C for the quadrupole, and 35–500 amu for the complete scan mode.

## 2.10. Determination of total phenolics content and total flavonoids content

The total phenolics content of *K.coccinea* seed oil was estimated by

the Folin-Ciocalteu method (Bubonja-Sonje et al., 2011) with slight modification. Briefly, 0.1 g of *K.coccinea* seed oil was mixed well with 1 mL of 80 % methanol by vortex blender and supernatant was collected. Combine three extracts as the detection solution. After that, combine 200 µL of Folin reagent with 100 µL of the sample solution, and let the mixture react for 30 min in the dark before incorporating 2 mL of a 10 % sodium carbonate solution. Then, being heated for 30 min at 70 °C after the mixed solution was diluted to a level of 10 mL with ultrapure water, and the absorbance was gauged at 760 nm, gallic acid was used as the standard control, the amount of total phenolics was determined, and the results were represented as mg of gallic acid equivalents (mg GAE/100 g) per 100 g of oil.

The total flavonoids content of *K.coccinea* seed oil was estimated according to Ji (n.d.). Briefly, 2 g *K.coccinea* seed oils were added with ethanol to 25 mL, shaken well before ultrasonic extraction for 20 min. Then, let stand to place 1 mL supernatant into the evaporating dish, add 1 g polyamide powder for adsorption, volatilize ethanol in the water bath and transfer it to the chromatographic column, which first elutes with 20 mL toluene and discard the toluene solution and then elute with methanol, combine the eluent and fix the volume to 25 mL before the absorbance was determined at 360 nm. Rutin was used as the standard control, and the result were expressed as mg rutin equivalents (RE) per 100 g of oil (mg RE / 100 g).

## 2.11. Determination of squalene content in *K.Coccinea* seed oil

According to Ye et al. (2021), the squalene concentration of *K.coccinea* seed oil was determined using Shimadzu GC-2010 gas chromatograph equipment with a flame ionization detector (GC-FID) using squalane as the internal standard. Briefly, 1 g of *K.coccinea* seed oil was added to 50 mL potassium hydroxide-ethanol solution for saponification. Then followed by extraction with 50 mL n-hexane and washed using 50 mL ethanol. Finally, the extract was concentrated with rotary evaporators and nitrogen-blowing instrument and adjusted with distilled water to a final volume of 10 mL as the detection solution. For analysis, an HP-5 column (30 m × 0.32 mm i.d × 0.25 mm) was used. For one minute, the temperature of the column was fixed at 160 °C. The temperature of the column was designed to rise from 160 to 220 °C at a rate of 15 °C/min, then to 280 °C at a rate of 5 °C/min, to be kept at 280 °C for 20 min, then to 300 °C for two min. Each sample was injected in 1.0 µL utilizing the split mode with a 40:1 split ratio. 1.0 mL/min of high quality nitrogen was employed as the inflow carrier gas. The FID detector temperature was kept at 300 °C, while the inlet temperature was kept at 250 °C. Squalene content was calculated as follows Eqs. (3):

$$X = \frac{A_1 \times m_s}{A_s \times m} \quad (3)$$

X—the content of the component to be measured in the sample, mg/kg; A<sub>1</sub>—the peak area of the component to be measured in the sample. As—the peak area of the internal standard squalane in the sample; m<sub>s</sub>—the internal standard squalane added to the sample mass, µg; m—the sampling amount, g.

## 2.12. Statistic analysis

Sample analyses were performed in triplicate. The SPSS22 (version 22.0, IBM Corp. Armonk, NY, USA) program was used to determine the mean and standard deviations. A statistically significant difference was indicated by the  $p < 0.05$ .

## 3. Results and discussions

### 3.1. The kernel yield, oil yield and the nutrients of *K.Coccinea* seeds

The dried *K.coccinea* seeds have a thin layer of light khaki shells and brown *K.coccinea* kernel. All of the oils were clear and transparent by

pressing and centrifuging. As shown in Table 1, the average kernel yield of the four varieties of *K.coccinea* seeds is 68.21 % with a variation coefficient of 1.02 %, and the average oil yield is 30.44 % with a variation coefficient of 7.89 %, which indicates that the kernel yield and oil yield of the four varieties of *K.coccinea* seeds are highly consistent. The crude fat content of *K.coccinea* seed kernels varied from 47.8 to 54.9 g/100 g indicating that the *K.coccinea* seed kernels are rich in oil, our finds are higher than that of *Cucumis melo* var. *agrestis* seeds ( $27.96 \pm 2.15\%$ ) (Jiang et al., 2022).

Meanwhile, the protein content was also shown to some extent higher than *Combretum micranthum* seed oil (11.95 g/100 g) and *Morinda citrifolia* seed oil (7.1 g/100 g), with the content varied from 16.5 g/100 g to 17.3 g/100 g (Bougma et al., 2021; Jahurul et al., 2022), and the ash content was 2.9–3.0 g/100 g. The protein content of *K.coccinea* seeds is higher than that of common grains, and the fat content is comparable to that of higher than most of edible oilseeds such as peanuts, sesame seeds, sunflower seeds and other oil (Wang et al., 2021). There are 11 mineral elements in *K.coccinea* seed kernels in all four varieties with variation coefficients of less than 10 % except sodium (21.88 %), indicating that there is general uniformity in the elemental composition of the four kinds of *K.coccinea* seeds. The five macroelements are potassium, phosphorus, magnesium, calcium, and sodium in descending order of content. These high-potassium and low-sodium elements are very beneficial to the human body. In addition, the contents of four trace elements, manganese, iron, copper and zinc, were also detected. Among these elements, the highest level was found in manganese with an average value of 122.12 mg/kg, followed by iron, these relatively higher levels of trace elements also contribute to the nutrition value of *K.coccinea* seeds.

### 3.2. The physicochemical properties of *K.Coccinea* seed oil

#### 3.2.1. Color

Color is one of the important quality indicators that affect

**Table 1**

The kernel yield, kernel oil yield, fat, protein, ash and mineral element content of *K.coccinea* seed.

Variety	DH	ZH	FH	HL
Kernel yield %	68.70 ± 0.38	67.23 ± 1.54	68.20 ± 0.77	68.72 ± 0.29
Oil yield %	27.54 ± 0.20	27.80 ± 2.43	33.36 ± 0.49	31.10 ± 0.52
Fat g/100 g	56.20 ± 1.83	49.05 ± 1.76	50.68 ± 2.23	52.73 ± 1.17
Protein g/100 g	16.6 ± 0.26	16.5 ± 0.13	16.8 ± 0.18	17.3 ± 0.16
Ash g/100 g	3.1 ± 0.07	3.1 ± 0.14	3.0 ± 0.07	3.1 ± 0.14
K(mg/kg)	7777.96 ± 217.04 a	7268.39 ± 190.77 ab	6830.12 ± 125.09 b	7861.50 ± 194.72 a
Na(mg/kg)	11.33 ± 2.49 a	7.68 ± 0.54 a	8.85 ± 1.41 a	7.77 ± 1.01 a
Ca(mg/kg)	564.57 ± 7.17 a	576.31 ± 40.02 a	501.61 ± 21.32 a	506.85 ± 26.72 a
Mg(mg/kg)	3401.96 ± 62.11 a	3303.74 ± 93.25 a	3017.89 ± 52.36 b	3354.25 ± 76.36 a
P(mg/kg)	4464.45 ± 96.36 a	4192.05 ± 141.12 ab	3931.55 ± 10.63 b	4519.25 ± 127.25 a
Mn *(mg/kg)	126.45 ± 2.62 a	124.65 ± 4.42 a	109.89 ± 2.01 b	127.50 ± 3.46 a
Fe *(mg/kg)	47.97 ± 0.18 a	41.86 ± 0.89 b	41.41 ± 0.37 b	48.01 ± 1.46 a
Cu *(mg/kg)	15.58 ± 0.23 a	13.40 ± 0.41 b	13.73 ± 0.15 b	15.80 ± 0.33 a
Zn *(mg/kg)	15.41 ± 0.29 a	13.52 ± 0.41 b	13.37 ± 0.27 b	14.93 ± 0.37 a

Notes: \* is trace element. Different letters in the same row indicate significant differences among different *K.coccinea* varieties ( $p < 0.05$ ). DH: Dahong variety of *K.coccinea* seed oil; ZH: Zihei variety of *K.coccinea* seed oil; FH: Fenhong variety of *K.coccinea* seed oil; HL: Hulu variety of *K.coccinea* seed oil.

consumers' choice of edible oil. The four *K.coccinea* seed oils are all light yellow. Their color measurement results are shown in Table 2. The luminance coefficient L is 81.10–83.73 indicating that oils have a bright appearance, their chromaticity coordinate  $a^*$  has some negative value from  $-11.91$  to  $-13.15$ , and the chromaticity coordinate  $b^*$  has positive values varied from 59.62 to 65.78. These differences may be due to the variety and some pigment components such as natural polyphenol pigments and carotenoids in vegetable oils, which are the dominant components affecting the color of vegetable oil.

#### 3.2.2. The acid value, peroxide value, iodine value and saponification value

The acid value, peroxide value, iodine value and saponification value of the four varieties of *K.coccinea* seed oil are shown in Table 2. Acid value is one of the important indicators to measure the antioxidant properties of oils. The acid value of *K.coccinea* seed oil varied from 0.70 to 0.85 mg/g, which is far below the 4 mg/g, stipulated in the Chinese national food safety standard for vegetable oil, indicating that the free fatty acid content of *K.coccinea* seed oil is low and not easy to oxidize. Peroxide value is an indicator to measure the degree of oil oxidation and is generally positively correlated with the degree of oil rancidity. The peroxide value of *K.coccinea* seed oils is 0.013–0.027 g/100 g, which is far lower than the China national standard of 0.25 g/100 g, indicating that the degree of oil oxidation is extremely low.

The iodine value reflects the degree of oil unsaturation. In general, the higher the iodine value, the greater the degree of unsaturation of oil. Here, the iodine value of four kinds of *K.coccinea* seed oil varied from 135.32 to 177.15 g/100 g, all above 130 g/100 g, which is higher than pumpkin seed oil and *Argemone mexicana* seed oil that content of 105.33–109.67 g/100 g and 141.2 g/100 g, respectively (Singh & Kumar, 2023), indicating that they are all dry oil. The saponification value is generally related to the purity of oil and the molecular weight of fatty acids. The saponification value of these four kinds of *K.coccinea*

**Table 2**

Physicochemical and lipid concomitants content of *K.coccinea* seed oil.

Variety	DH	ZH	FH	HL
Color				
L*	81.95 ± 0.01 c	82.84 ± 0.13 b	81.10 ± 0.09 d	83.73 ± 0.02 a
a*	-13.02 ± 0.01 c	-13.15 ± 0.00 d	-11.91 ± 0.01 a	-12.74 ± 0.02 b
b*	61.11 ± 0.01 c	63.91 ± 0.19 b	59.62 ± 0.14 d	65.82 ± 0.01 a
acid value(mg/g)	0.70 ± 0.005 c	0.75 ± 0.001 b	0.70 ± 0.000 c	0.85 ± 0.001 a
peroxide value(g/100 g)	0.013 ± 0.0002 d	0.018 ± 0.0001 b	0.015 ± 0.0001c	0.027 ± 0.0003a
iodine value (g/100 g)	135.32 ± 0.45 c	168.35 ± 1.50 b	166.45 ± 1.35 b	177.15 ± 0.50 a
saponification value (mg/g)	178.05 ± 0.25 a	177.95 ± 0.05 a	156.20 ± 0.80 c	169.05 ± 0.18 b
accelerated oxidation (h)	3.19 ± 0.05 a	1.58 ± 0.03 b	3.14 ± 0.02 a	1.36 ± 0.03 c
total phenolics (mg GAE/100 g)	393.22 ± 6.92 c	430.15 ± 3.26 b	368.99 ± 3.46 d	503.99 ± 5.65 a
total flavonoids (mg RE/100 g)	101.67 ± 1.49 bc	117.46 ± 5.46 ab	95.01 ± 0.59 c	126.18 ± 6.82 a
β-sitosterol(mg/kg)	1593.7 ± 134.1c	1790.4 ± 109.9a	1578.0 ± 138.9c	1621.7 ± 100.2b
cholesterol(mg/kg)	95.2 ± 1.9	94.7 ± 2.3	96.4 ± 2.4	95.8 ± 2.1
campesterol(mg/kg)	ND	ND	ND	ND
stigmasterol(mg/kg)	ND	ND	ND	ND
Total sterolamount (mg/kg)	1688.8 ± 136.1b	1885.1 ± 112.2a	1674.4 ± 136.5b	1212.4 ± 102.3c
squalene(mg/kg)	ND	14.4 ± 0.0	15.1 ± 0.1	14.7 ± 0.1

Notes: ND:Undetectable or below the limited of detection. Different letters in the same row indicate significant differences among different *K.coccinea* varieties ( $p < 0.05$ ). DH: Dahong variety of *K.coccinea* seed oil; ZH: Zihei variety of *K.coccinea* seed oil; FH: Fenhong variety of *K.coccinea* seed oil; HL:Hulu variety of *K.coccinea* seed oil.

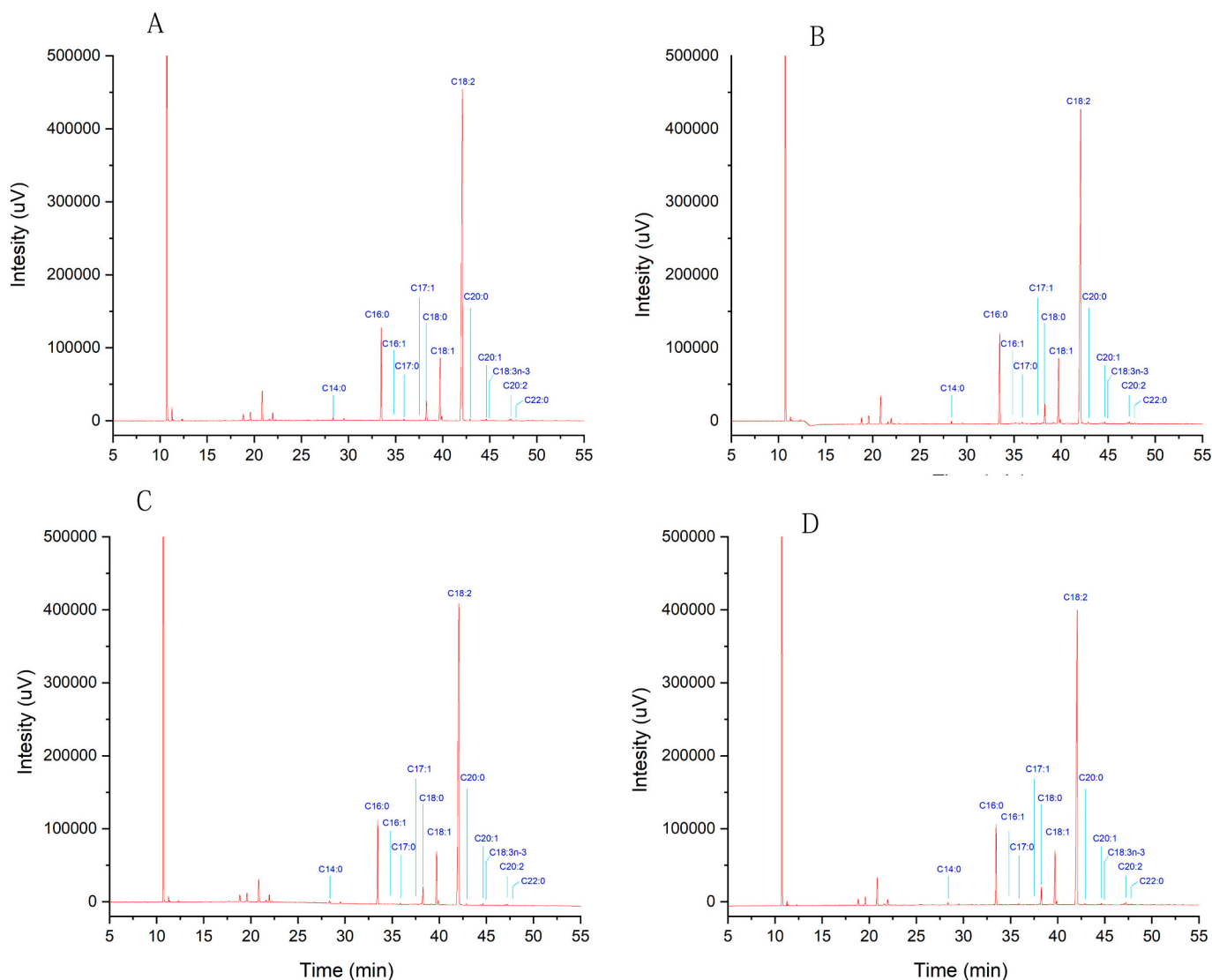
seed oil ranged from 156.20 to 178.05 mg/g with the Fenhong varieties being the lowest. The lower the saponification value, the fewer free fatty acids and glycerides in the oil, and the more impurities. On the contrary, the higher the saponification value, the better the oil quality.

For oil accelerated oxidation experiments, the Rancimat method is widely used because of its good reproducibility and easy operation. The Rancimat Oxidation Stabilizer reflects the oil oxidation induction time by detecting the change of the conductivity of the solution in the measuring cell. Under the oxidation induction temperature was 120 °C, the induction periods of Dahong and Fenhong were 3.19 h and 3.14 h, respectively, which is longer than that of Zihei (1.58 h) and Hulu (1.36 h), indicating that the Dahong and Fenhong have relatively stronger stability. This difference may be caused by the different levels of linoleic acid in oils as the higher content of linoleic acid, the shorter time of the induction time (Zhang et al., 2015). Moreover, the induction time was probably close to the level of acid value, as the acid value of Zihei and Hulu were slightly higher than those of Dahong and Fenhong.

### 3.3. Volatile components of *K.Coccinea* seed oil

Volatile substances are also one of the important indicators for the sensory characteristics of vegetable oils. Different oils often have

different flavors and volatile substances in oils give them unique smells and characteristics. The total ion chromatography of volatile components detected in the four varieties of *K.coccinea* seed oil was shown in S. Fig. 1, and the compositions are shown in Table 3. A total of 60 volatile components have been detected in these seeds and 52, 54, 48 and 52 volatile components were detected in Dahong, Zihei, Fenhong and Hulu, respectively, which accounted for 96.58 %, 95.66 %, 95.37 % and 93.80 % of the corresponding total peak area, respectively. Among them, most of the volatile components were alkenes, and a small amount of alcohols, aldehydes, esters, ketones and other substances. There were 44 common components in the volatile substances of *K.coccinea* seed oil, accounting for the majority of the total peak area, indicating that the volatile components of the four varieties of *K.coccinea* seed oil were highly consistent. Of these ingredients, 11 volatile substances have a content of 1 % of the total, which are also common components of the four *K.coccinea* seed oils. The compounds listed below, in descending order of content, make up 87.19–89.88 % of the total peak area:  $\gamma$ -Cadinene,  $\beta$ -Himachalene,  $\beta$ -Curcumene,  $\alpha$ -Longipinene, Cuparene Bicycl, Caryophyllene, 1,5-dimethyl-8-(1-methylethenyl)-[S-(Z,E)],  $\alpha$ -Zingiberene, Selina-3,7(11)-diene, Bergamotol, Z.- $\alpha$ .trans-, (+)-epi-Bicyclosesquiphellandrene. These volatile components of *K.coccinea* seed oil are mainly sesquiterpenes. Among the 11 most important



**Fig. 1.** The total ion chromatogram of volatile components detected in the four varieties of *K.coccinea* seed oil. A: Dahong variety of *K.coccinea* seed oil; B: Zihei variety of *K.coccinea* seed oil; C: Hulu variety of *K.coccinea* seed oil; D: Fenhong variety of *K.coccinea* seed oil.

**Table 3**  
Volatile components of *K.coccinea* seed oil.

Retention time (min)	Compound name	Retention index	CAS	Classification	Relative content/%			
					DH	ZH	FH	HL
10.235	(+)-Dipentene	1018	5989-27-5	alkene	ND	ND	0.01	ND
10.332	Eucalyptol	1059	470-82-6	alcohols	0.01	0.01	0.00	ND
17.994	δ-Elemene	1377	20,307-84-0	alkene	0.11	0.10	0.14	0.13
18.206	α-cubebene	1344	17,699-14-8	alkene	0.01	0.01	0.02	0.01
18.294	2-Isopropenyl-4a,8-dimethyl-1,2,3,4,4a,5,6,8a-octahydronaphthalene	1474	473-13-2	alkene	0.01	0.01	ND	ND
18.600	Ylangene	1221	14,912-44-8	alkene	ND	ND	0.02	0.02
18.634	α-Elemene	1410	5951-67-7	alkene	0.05	0.05	0.03	0.03
18.676	α-Copaene	1221	3856-25-5	alkene	0.02	0.02	0.03	0.03
18.737	γ-Cadinene	1435	39,029-41-9	alkene	0.38	0.37	0.39	0.35
18.805	Di-epi-.alpha.-cedrene	1403	50,894-66-1	alkene	0.05	0.05	0.05	0.05
18.927	β-Elemene	1398	515-13-9	alkene	0.20	0.21	0.22	0.20
18.998	α-Cubebene	1344	17,699-14-8	alkene	0.04	0.04	0.04	0.03
19.100	(+)-Ledene	1419	21,747-46-6	alkene	0.59	0.57	0.61	0.58
19.235	(+)-Sativen	1339	3650-28-0	alkene	0.03	0.03	0.04	0.03
19.302	Spiro[2.4]heptane,1,2,4,5-tetramethyl-6-methylene-	1065	74,792-98-6	alkene	ND	ND	0.38	0.35
19.325	Cedrene	1403	469-61-4	alkene	0.42	0.35	ND	ND
19.443	Caryophyllene	1494	87-44-5	alkene	3.63	3.52	2.31	1.95
19.542	α-Guaiene	1474	3691-12-1	alkene	0.42	0.42	0.46	0.43
19.658	cis-Thujopsene	1416	470-40-6	alkene	0.06	0.05	0.05	0.05
19.733	α-Chamigrene	1512	19,912-83-5	alkene	0.08	0.08	0.12	0.12
19.785	β-sesquiphellandrene	1446	20,307-83-9	alkene	0.03	0.02	ND	ND
19.900	(-)-Isolodene	1419	95,910-36-4	alkene	0.87	0.66	0.82	0.69
19.982	Bicyclo[7.2.0]undecane,10,10-dimethyl-2,6-bis(methylene)-, [1S-(1R*,9S*)]-	1489	136,296-38-3	alkene	0.30	0.29	0.34	0.34
20.096	Selina-3,7(11)-diene	1507	6813-21-4	alkene	1.30	1.31	1.32	1.23
20.163	Acoradien	1474	24,048-44-0	alkene	0.78	0.70	0.72	0.62
20.238	(+)-epi-Bicyclosesquiphellandrene	1435	54,274-73-6	alkene	1.18	0.85	1.06	0.90
20.300	α-Bisabolene	1518	25,532-79-0	alkene	0.16	0.18	0.16	0.16
20.508	β-Himachalene	1528	1461-03-6	alkene	19.90	20.37	19.85	19.51
20.559	(+)-Cuparene	1556	16,982-00-6	alkene	4.47	5.49	4.61	4.28
20.735	Bicyclo[5.2.0]nonane, 2-methylene-4,8,8-trimethyl-4-vinyl-	1407	242,794-76-9	alkene	ND	0.47	ND	ND
20.797	α-Zingiberene	1451	495-60-3	alkene	2.47	1.55	2.71	2.75
20.850	(1-Methoxy-4-methyl-3-pentenyl)benzene	1387	68,705-86-2	ethers	ND	0.84	ND	ND
20.921	β-Panasinsene	1411	56,684-97-0	alkene	0.90	0.86	1.08	1.01
21.033	α-Longipinene	1403	5989-08-2	alkene	6.85	5.79	5.17	5.13
21.221	β-Curcumene	1480	28,976-67-2	alkene	15.78	17.68	16.23	16.42
21.375	γ-Muurolene	1435	30,021-74-0	alkene	30.77	27.25	31.70	31.30
21.470	δ-Cadinene	1469	483-76-1	alkene	ND	0.02	ND	ND
21.605	Bergamotol,Z-.alpha.-trans-	1673	88,034-74-6	alcohols	1.02	1.16	1.13	1.15
21.764	1,5-Cyclodecadiene,1,5-dimethyl-8-(1-methylethenyl)-, [S-(Z,E)]-	1570	75,023-40-4	alkene	2.51	2.70	2.50	2.57
22.050	cubedol	1484	100,374-15-9	alcohols	0.03	ND	ND	0.02
22.188	β-Vatirenene	1489	0-00-0	alkene	0.10	0.17	0.12	0.13
22.277	10s,11s-Himachala-3(12),4-diene	1494	60,909-28-6	alkene	0.14	0.15	0.17	0.17
22.610	Isolongifolene, 4,5,9,10-dehydro-	1380	156,747-45-4	alkene	0.05	0.07	0.05	0.06
22.685	p-(Pentyloxy)acetophenone	1616	5467-56-1	ketones	0.05	0.08	0.07	0.07
22.767	α-curcumene	1524	644-30-4	alkene	0.17	0.22	0.17	0.20
23.051	Caryophyllene oxide	1507	1139-30-6	oxide	0.01	0.01	0.01	0.01
23.568	geranyl-.alpha.-terpinene	1962	0-00-0	alkene	0.05	0.09	0.07	0.07
23.751	7-epi-cis-sesquisabinene hydrate	1523	0-00-0	containing hydroxyl	0.03	0.04	0.03	0.04
24.228	(-)-Globulol	1530	489-41-8	alcohols	ND	ND	ND	0.02
24.933	α-Cadinol	1580	481-34-5	alcohols	0.28	0.29	0.18	0.25
25.155	1,5-Heptadien-4-ol, 3,3,6-trimethyl-	1068	27,644-04-8	alcohols	0.11	0.15	0.09	0.14
25.538	α-Bisabolol	1625	515-69-5	alcohols	0.01	0.08	ND	0.03
25.639	Aromadendrene oxide-(1)	1462	94,020-95-8	oxide	0.01	0.04	ND	0.02
25.724	Tumerone	1616	180,315-67-7	ketones	0.04	0.07	0.03	0.05
25.921	(-)-Spathulenol	1536	77,171-55-2	alcohols	0.02	0.02	0.02	0.02
26.052	trans-Geranylgeraniol	2192	24,034-73-9	alcohols	0.02	0.03	0.02	0.03
26.501	2,6,10-Dodecatrienal, 3,7,11-trimethyl-	1656	19,317-11-4	aldehyde	ND	ND	ND	0.01
26.624	Longiverbenone	1574	64,180-68-3	ketones	0.02	0.02	ND	0.01
27.546	Farnesol, acetate	1834	29,548-30-9	esters	0.02	0.03	0.01	0.02
27.730	(-)-Isolongifolol	1635	1139-17-9	alcohols	0.02	0.02	0.01	0.01

Notes:ND:Undetectable or below the limited of detection. Different letters in the same row indicate significant differences among different *K.coccinea* varieties ( $p < 0.05$ ). DH: Dahong variety of *K.coccinea* seed oil; ZH: Zihei variety of *K.coccinea* seed oil; FH: Fenhong variety of *K.coccinea* seed oil; HL:Hulu variety of *K.coccinea* seed oil.

substances, only borneol is a monoterpene, and the other 10 substances are sesquiterpenes. Among the natural products, sesquiterpenes are a large and important group, and the basic skeleton of sesquiterpenes is acacia pyrophosphate composed of three isoprene groups, which is formed through a series of complex transformations in living organisms, becoming the largest type and number of structural skeletons among terpenes components, and most sesquiterpenes have good biological activity. Sesquiterpenes and monoterpenes are also the main components of volatile oils, giving them various properties such as antibacterial, antifungal, antiviral, anti-inflammatory, insecticidal, and antioxidant while imparting a strong and unique odor (da Silva Gündel et al., 2018).

### 3.4. Fatty acids of *K.Coccinea* seed oil

The fatty acid composition, sterol, and squalene content of the four kinds of *K.coccinea* seed oils are shown in Table 4. Their fatty acid composition is similar with a total of 13 fatty acids (Fig. 1), 6 saturated fatty acids, and 7 unsaturated fatty acids detected. Linoleic acid among them had the highest content of 68.32%. This is comparable to the high content of linoleic acid in the widely used grapeseed oil (67%) and slightly lower than that of the “king of linoleic acid” safflower oil (78.54% ~ 82.45%). However, it is significantly higher than that of many common vegetable oils, including corn oil (57%), soybean oil (54%) and linseed oil (Liang et al., 2021; Xu et al., 2022).  $\alpha$ -Linolenic acid and linoleic acid are essential fatty acids that must be obtained from dietary sources, which can promote the conversion of excess cholesterol in the body into bile salts, prevent the deposition of cholesterol in the arterial wall, reduce serum cholesterol content and prevent arteriosclerosis. Palmitic acid (12.2–12.8%) and oleic acid (2.68–2.92%) were next to the linoleic acid in abundance in *K.coccinea* seed oils. *K.coccinea* seed oil can be exploited as a new edible oil source due to its high amount of seed

**Table 4**  
Fatty acid composition and content of *K.coccinea* seed oil.

fatty acid(%)	DH	ZH	FH	HL
Myristic C <sub>14:0</sub>	0.36 ± 0.00	0.36 ± 0.01	0.37 ± 0.01	0.35 ± 0.00
Palmitic C <sub>16:0</sub>	12.82 ± 0.04	12.80 ± 0.10	12.20 ± 0.60	12.47 ± 0.01
	0.11 ± 0.01b	0.08 ± 0.00c	0.07 ± 0.00c	0.13 ± 0.01a
Palmitoleic C <sub>16:1</sub>	0.21 ± 0.00	0.21 ± 0.00	0.22 ± 0.01	0.21 ± 0.01
Margaric C <sub>17:0</sub>	0.06 ± 0.00	0.06 ± 0.00	0.05 ± 0.00	0.07 ± 0.00
Ginkgolic C <sub>17:1</sub>	2.92 ± 0.01	2.80 ± 0.02	2.77 ± 0.09	2.80 ± 0.02
Stearic C <sub>18:0</sub>	9.19 ± 0.05a	8.85 ± 0.08b	9.19 ± 0.03a	8.38 ± 0.03b
	67.65 ± 0.45b	68.05 ± 0.45b	67.18 ± 0.33b	70.38 ± 0.01a
Linoleic C <sub>18:2</sub>	0.22 ± 0.00	0.21 ± 0.00	0.21 ± 0.01	0.20 ± 0.00
Arachidic C <sub>20:0</sub>	0.17 ± 0.00a	0.12 ± 0.00c	0.12 ± 0.00c	0.15 ± 0.02b
	0.26 ± 0.00b	0.28 ± 0.00b	0.25 ± 0.00b	0.22 ± 0.00a
Linolenic C <sub>18:3</sub>	0.00b	0.02a	0.01c	0.01d
	0.05 ± 0.00b	0.04 ± 0.00b	0.04 ± 0.00b	0.07 ± 0.00a
Eicosapentaenoic C <sub>20:2</sub>	0.00b	0.00b	0.00b	0.00a
Behenic C <sub>22:0</sub>	0.10 ± 0.00	0.10 ± 0.00	0.10 ± 0.00	0.11 ± 0.00
∑SFA	16.66 ± 0.06	16.48 ± 0.13	15.86 ± 0.67	16.14 ± 0.01
∑MUFA	9.52 ± 0.06a	9.48 ± 0.45a	9.43 ± 0.02a	8.73 ± 0.01b
∑PUFA	67.96 ± 0.45b	68.37 ± 0.47b	67.46 ± 0.33b	70.67 ± 0.00a
	77.48 ± 0.51b	77.85 ± 0.92b	76.90 ± 0.31b	79.39 ± 0.01a

Notes: Different letters in the same row indicate significant differences among different *K.coccinea* varieties ( $p < 0.05$ ). SFA: saturated fatty acid; MUFA: monounsaturated fatty acid; PUFA: polyunsaturated fatty acids; USFA: unsaturated fatty acid; DH: Dahong variety of *K.coccinea* seed oil; ZH: Zihei variety of *K.coccinea* seed oil; FH: Fenhong variety of *K.coccinea* seed oil; HL: Hulu variety of *K.coccinea* seed oil.

oil and unsaturated fatty acids.

### 3.5. Analysis of lipids and their concomitants in *K.Coccinea* seed oil

#### 3.5.1. The lipids in *K.Coccinea* seed oil

The study utilized UPLC-QTOF-MS/MS tandem technology to analyze the biological components present in four varieties of *K.coccinea* seed oil. The results, including heat maps of compositions are presented in Fig. 2. A total of 74 substances were identified through comparative analysis of the contrast database, with 34 of them being fatty acyl compounds such as stearic acid, myristic acid, palmitic acid, heptadecanoic acid, linoleic acid, and eicosanoic acid. The majority of these fatty acids were consistent with those detected using gas chromatography. Furthermore, pinolenic acid greatly reduced lipid accumulation, oxidative stress, and inflammatory responses generated by oleic acid in HepG2 cells (Zhang et al., 2019). Moreover, 9(S)-HODE and 9(S)-HOTRE are chemicals in the metabolic pathways of linoleic acid and  $\alpha$ -linolenic acid, respectively. Both are positioned in the downstream locations of the metabolic processes. In addition, the study discovered that *K.coccinea* seed oil also contains some rich phytosterols, such as  $\beta$ -sitosterol and cholesterol. Of these,  $\beta$ -sitosterol is the predominant phytosterols in *K.coccinea* seed oil, with a level of 1498.8–1712.7 mg/kg of oil (Table 2). This was greater than the levels found in *Balanites roxburghii* seed oil (52.5–147 mg/kg) (Yadav et al., 2023), and some edible oil such as olive oil (1200–1330 mg/kg) (Bai et al., 2021), palm oil (150.6–434.7 mg/kg), coconut oil (415.14–422.79 mg/kg), etc. These triterpene molecules have a similar structure to cholesterol and provide a variety of health benefits such as cardiovascular and hypocholesterolemic activity prevention.

#### 3.5.2. The terpenoids in *K.Coccinea* seed oil

In addition, 16 terpenoids (sesquiterpenes, diterpenes, and triterpenes) were identified. The four varieties of *K.coccinea* seed oil contained two types of sesquiterpenes, seven types of diterpenes, and seven types of triterpenes, which differed from the volatile components, which were primarily sesquiterpenes. *K.coccinea* seed oil contains terpenoids such as 5-Hydroxyiminoisocaryophyllene, abscisic acid, geraniol, pristanoic acid, carnosic acid, sclareol, rafenolide, glycyrrhetic acid, madecassoside, betulinic acid,  $\alpha$ -boswellic acid, 11-Oxoursolic acid acetate, and others. These compounds like prenyl lipids all possess similar 5-carbon precursors, isopentenyl diphosphate. Thus, they coexist with fatty acyl molecules found in seed oils. The majority of these terpenoids have good biological activity, for example, sclareol has antibacterial action (Tran et al., 2019), glycyrrhetic acid possesses anti-inflammatory, antioxidant, antitoxin, gastroprotective, antiviral, cardioprotective, anti-inflammatory effects. Asiaticoside has various activities such as antitumor, neuroprotection and liver protection (Ming & Yin, 2013), and madecassoside has activities such as treatment of skin trauma, anti-inflammation, tumor suppressor, and anti-oxidative stress (Kunjumon et al., 2022). Geraniol is an intermediary in the production of vitamin E and vitamin K, with active properties such as antibacterial, anti-inflammatory, anti-tumor, and neuroprotective effects (Ho et al., 2018). Carnosic acid has a phenolic hydroxyl group, which is a lipophilic antioxidant molecule with strong antibacterial action as well as tumor suppressor, anti-inflammatory, and fat-reducing properties. It is often utilized in the food, nutrition, healthcare, and cosmetics sectors (Birtić et al., 2015).

#### 3.5.3. The phenolic compounds in *K.Coccinea* seed oil

Besides the active ingredients mentioned above compounds, a total of 14 phenolic substances including 9 phenolic acids, 1 flavonoid and 4 lignans were identified in four varieties of *K.coccinea* seed oil. Ferulic acid, 4-Hydroxy-3-methoxycinnamaldehyde, hydroquinone, catechol, dextranquinic acid, coniferyl aldehyde, 3-methoxycatechol, syringaldehyde, isoeugenol, chrysanthemine, etc., among which the last five are phenolic substances common to the four *K.coccinea* seed oils. Among

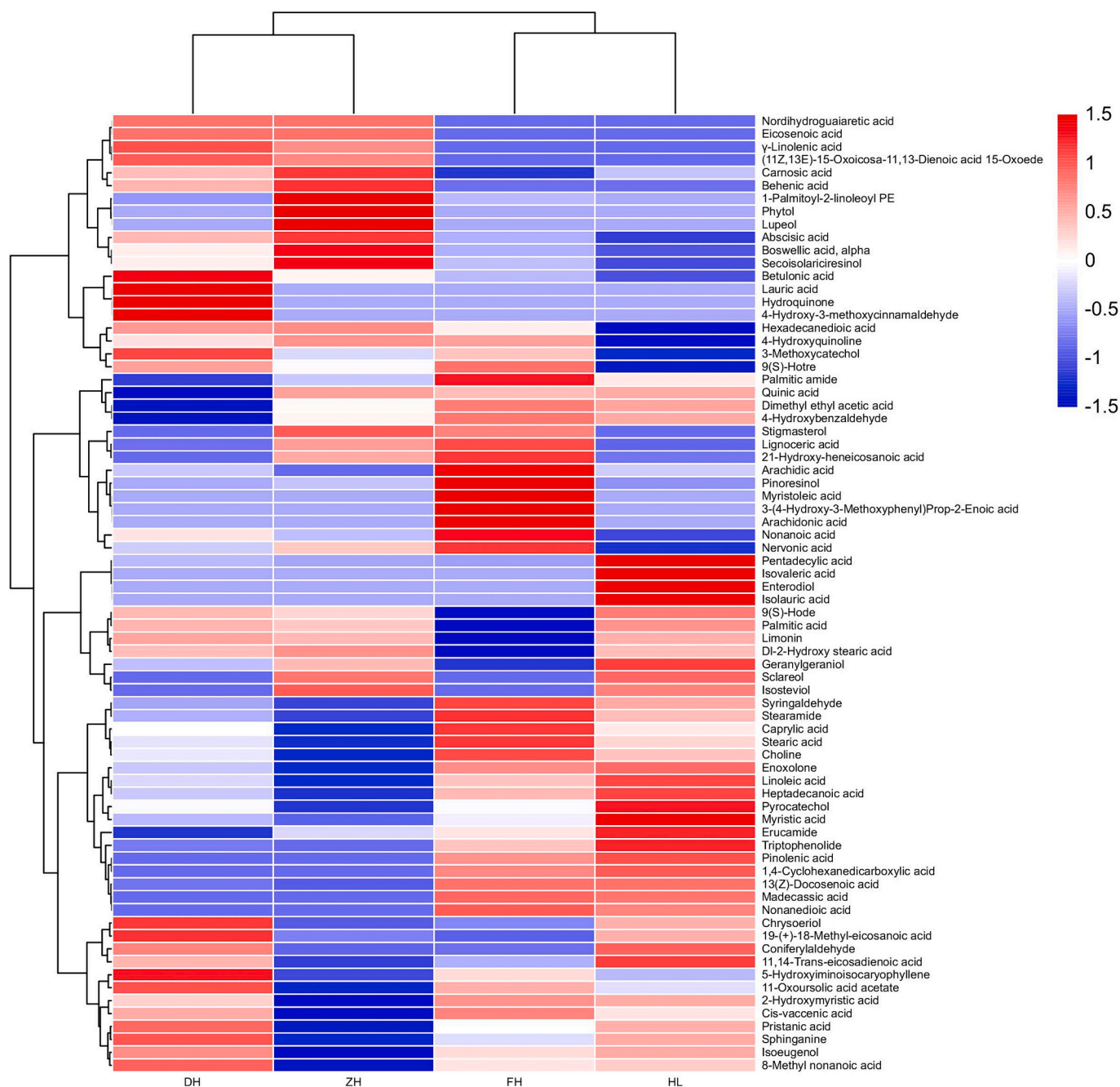


Fig. 2. Heatmap clustering of metabolites in seed oils of four *K.coccinea* species.

them, hydroquinone has a good antioxidant and whitening activity. Syringaldehyde has antioxidant and anti-inflammatory activities. Isoeugenol is mainly used as a fragrance in industry. Chrysoeriol is a natural flavonoid with various benefits such as anti-oxidant, anti-inflammatory, anti-tumor, anti-osteoporosis, and cardioprotective (Limboonreung et al., 2020). Coniferylaldehyde is a plant phenol found in several medicinal plants such as sage, eugenol, and cinnamon, with anti-inflammatory and antioxidant activities (Akram et al., 2016). Polyphenolic compounds not only have antioxidant effects but also have physiological functions of lowering blood lipids and preventing cardiovascular and cerebrovascular diseases, which are a class of active substances of great importance to human health. For lignan compounds, four lignans were detected in *K.coccinea* seed oil with significant differences between the varieties. Only Pinoresinol and open-loop isolapsin resin phenol all exist in the four *K.coccinea* seed oil. Pinoresinol is in the

upstream position of the open-loop isolapsin resin phenol in the metabolic pathway, and the conversion is completed by the action of pinoresinol-lapsinol reductase. Nordihydroguaiaretic acid was only detected in Dahong and Zihei, and only enterodiol was detected in Hulu.

Due to lack of reference material of phenolics compounds, the total phenolics content and total flavonoids of *K.coccinea* seed oil were spectrophotometry detected by are summarized in Table 2. of the four varieties, Hulu had the highest total phenolics content with the value of  $503.99 \pm 5.65$  mg GAE/100 g, followed by Zihei ( $430.15 \pm 3.26$  mg GAE/100 g), Dahong ( $393.22 \pm 6.92$  mg GAE/100 g) and Fenhong ( $368.99 \pm 3.46$  mg GAE/100 g), which significantly higher than that of nutmeg seed oils ( $3.21$  mg GAE/g) (Kozłowska et al., 2016). Phenolic compounds can protect lipids from peroxidation and maintain lipid stability. The abundance of phenolic components found in *K. coccinea* seed oil suggests that the oil may possess superior antioxidant



properties. The total flavonoid content of the four varieties of *K.coccinea* seed oil varied from  $95.01 \pm 0.59$  mg RE/100 g to  $126.18 \pm 6.82$  mg RE/100 g. Consistent with the level of total phenolics content, the total flavonoid content of Hulu and Zihei varieties was significantly higher than that of Dahong and Fenhong varieties, all higher than that of pressed perilla seed oil (43.6 mg/kg) (Jia et al., 2022).

### 3.5.4. The squalene in *K.Coccinea* seed oil

Aside from the above compounds, another unsaturated fatty acid, squalene, was also detected in *K.coccineas* seed oil. Squalene is a kind of bioactive molecule with antihypoxia activity, which is a precursor molecule for the synthesis of phytosterols and steroid hormones, widely found in various seed oils such as olive oil, camellia oil, peanut oil, soybean oil, peony seed oil, and linseed oil with a certain amount of squalene (Zhu, 2019). As shown in Table 2, a relatively higher level of squalene (14.4–15.1 mg/kg) has also been found in *K.coccinea* seed oil except Dahong variety. These values were much higher than many other plant seed oils, such as grape seed oil (Beveridge et al., 2005), pomegranate seed oil (Liu et al., 2022), etc. It was only 1 to 2 % lower than those found in olive oil (Cert et al., 2000), indicating that *K.coccinea* seed oil has a good application potential.

## 4. Conclusion

This present work is the first attempt to thoroughly assess the physicochemical properties and compositions of seed oils derived from four *K. coccinea* types with the goal of developing them into a possible industrial oil source. The nutritional composition of the seed oil showed that the seed was rich in phenolics and flavonoids, and also showed high iodine value, low acid value, peroxide value, saponification value and other good quality parameters. *K.coccinea* seed oil is rich in linoleic acid, terpenoids, phenols and lignans. In addition, the content of  $\beta$ -sitosterol is high, and a small amount of cholesterol and squalene. These findings clearly indicate that *K.coccinea* seeds may contribute to a variety of biological and nutritional applications. These bioactive ingredients are the biological material basis for *K.coccinea* seed oil to exert its antioxidant, antibacterial and other activities, indicating that *K.coccinea* seed oil has the potential to be processed into important edible oil, pharmaceutical and cosmetic oil in industrial mainstream.

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## CRediT authorship contribution statement

**Jing Wang:** Writing – original draft, Visualization, Software, Data curation. **Yingying Cheng:** Writing – review & editing, Methodology, Formal analysis. **Liying Fang:** Writing – review & editing, Methodology, Formal analysis. **Ao Yang:** Conceptualization. **Feijun Luo:** Validation, Resources. **Jun Lu:** Writing – review & editing. **Jiali Ren:** Validation, Resources.

## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Data availability

No data was used for the research described in the article.

## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.fochx.2024.101765>.

## References

- Akram, M., Kim, K.-A., Kim, E.-S., Shin, Y.-J., Noh, D., Kim, E., Kim, J.-H., Majid, A., Chang, S.-Y., & Kim, J.-K. (2016). Selective inhibition of JAK2/STAT1 signaling and iNOS expression mediates the anti-inflammatory effects of coniferyl aldehyde. *Chemico-Biological Interactions*, 256, 102–110. <https://doi.org/10.1016/j.cbi.2016.06.029>
- AOAC. (2016). Official methods of analysis of AOAC international. In W. Horwitz, & G. Latimer (Eds.), *Official methods of analysis of AOAC international* (20th ed.). Gaithersburg, MD: AOAC International.
- Bai, G., Ma, C., & Chen, X. (2021). Phytosterols in edible oil: Distribution, analysis and variation during processing. *Grain & Oil Science and Technology*, 4(1), 33–44. <https://doi.org/10.1016/j.gaost.2020.12.003>
- Beveridge, T. H., Girard, B., Kopp, T., & Drover, J. C. (2005). Yield and composition of grape seed oils extracted by supercritical carbon dioxide and petroleum ether: Varietal effects. *Journal of Agricultural and Food Chemistry*, 53(5), 1799–1804. <https://doi.org/10.1021/jf040295q>
- Birtić, S., Dussort, P., Pierre, F.-X., Bily, A. C., & Roller, M. (2015). Carnosic acid. *Phytochemistry*, 115, 9–19. <https://doi.org/10.1016/j.phytochem.2014.12.026>
- Bougma, A., Sere, A., Bazie, B. S. R., Sangare, H., Ouilly, J. T., & Bassole, I. H. N. (2021). Composition and physicochemical properties of Combretum collinum, Combretum micranthum, Combretum nigricans, and Combretum niorense seeds and seed oils from Burkina Faso. *Journal of the American Oil Chemists' Society*, 98(11), 1083–1092. <https://doi.org/10.1002/aocs.12540>
- Bubonja-Sonje, M., Giacometti, J., & Abram, M. (2011). Antioxidant and antilisterial activity of olive oil, cocoa and rosemary extract polyphenols. *Food Chemistry*, 127(4), 1821–1827. <https://doi.org/10.1016/j.foodchem.2011.02.071>
- Cert, A., Moreda, W., & Pérez-Camino, M. (2000). Chromatographic analysis of minor constituents in vegetable oils. *Journal of Chromatography A*, 881(1–2), 131–148. [https://doi.org/10.1016/S0021-9673\(00\)00389-7](https://doi.org/10.1016/S0021-9673(00)00389-7)
- Fan, M., Fan, Y., Huang, W., Wang, L., Li, Y., Qian, H., Zhang, H., & Qi, X. (2018). Tentative characterization of precursor compounds and co-factors of pigment formation in production of 'wu mi' from *Vaccinium bracteatum* Thunb. Leaves. *Food Chemistry*, 262, 199–205. <https://doi.org/10.1016/j.foodchem.2018.04.101>
- Ho, H.-J., Shirakawa, H., Giriwono, P. E., Ito, A., & Komai, M. (2018). A novel function of geranylgeraniol in regulating testosterone production. *Bioscience, Biotechnology, and Biochemistry*, 82(6), 956–962. <https://doi.org/10.1080/09168451.2017.1415129>
- Jahurul, M., Jack, C., Syifa, A., Shahidul, I., Norazlina, M., Shihabul, A., & Zaidul, I. (2022). Physicochemical and antioxidant properties, total phenolic and nutritional contents of noni (*Morinda citrifolia*) seed and its oil cultivated in Sabah, Malaysia. *Food Chemistry Advances*, 1, Article 100079. <https://doi.org/10.1016/j.focha.2022.100079>
- Ji, Y. Technical Guiding Principles for the Inspection and Evaluation of Physical, Chemical and Hygienic Indicators of Health Food (2020 Edition), Third Expert Q&A Session on Functional Claims, Efficacy Determination, R&D and Registration, Processes and Technical Requirements of Health Foods after the New Regulation, Nanjing, Jiangsu Province, China (2021), 125–212.
- Jia, X., Zhang, Q., Liu, H., Wang, X., Cao, Y., & Zhong, G. (2022). Effect of infrared preheating treatment on quality and antioxidant activity of pressed perilla seed oil. *China Oils and Fats*, 47(02), 16–22. <https://doi.org/10.19902/j.cnki.zgyz.1003-7969.210187>
- Jiang, C., Li, X., Jiang, Q., Wang, X., Chen, H., Ma, Z., & Chen, J. (2022). Composition analysis and oxidative stability of *Cucumis melo* var. *agrestis* seed oil. *China Oils and Fats*, 47(02). <https://doi.org/10.19902/j.cnki.zgyz.1003-7969.210126>, 109–113+135.
- Kozłowska, M., Gruczyńska, E., Ścibisz, I., & Rudzińska, M. (2016). Fatty acids and sterols composition, and antioxidant activity of oils extracted from plant seeds. *Food Chemistry*, 213, 450–456. <https://doi.org/10.1016/j.foodchem.2016.06.102>
- Kunjumon, R., Johnson, A. J., & Baby, S. (2022). *Centella asiatica*: Secondary metabolites, biological activities and biomass sources. *Phytomedicine Plus*, 2(1), Article 100176. <https://doi.org/10.1016/j.phyplu.2021.100176>
- Li, L., Tang, L., Xu, Y., & Yan, Z. (2020). Research Progress on chemical constituents and functions of *Kadsura coccinea* (Lem.) a. C. Smith in recent ten years. *Journal of Chinese Medicinal Materials*, 43(01), 236–242. <https://doi.org/10.3969/j.issn.1674-9146.2021.11.079>
- Li, Y., Liang, J., He, Y., Huang, X., & Li, S. (2022). Study on the ultrasonic-assisted extraction technology and antioxidant activity of polyphenols from *Kadsura coccinea* peel. *China Food Additives*, 33(04), 48–54. <https://doi.org/10.19804/j.issn1006-2513.2022.04.007>
- Liang, H., Xu, L., Yu, Y., Tan, Z., Yang, H., Dong, W., Li, L., Li, C., Liu, X., & Zhang, S. (2021). Evaluation and analysis of fatty acid composition and contents in safflower oil. *Food Science*, 42(06), 244–249. <https://doi.org/10.7506/spkx1002-6630-20200218-183>
- Limboonreung, T., Tuchinda, P., & Chongthammakun, S. (2020). Chrysoeriol mediates mitochondrial protection via PI3K/Akt pathway in MPP+ treated SH-SY5Y cells. *Neuroscience Letters*, 714, Article 134545. <https://doi.org/10.1016/j.neulet.2019.134545>

- Liu, J., Chen, P., He, J., Deng, L., Wang, L., Lei, J., & Rong, L. (2014). Extraction of oil from *Jatropha curcas* seeds by subcritical fluid extraction. *Industrial Crops and Products*, 62, 235–241. <https://doi.org/10.1016/j.indcrop.2014.08.039>
- Liu, N., Ren, G., Faiza, M., Li, D., Cui, J., Zhang, K., Yao, X., & Zhao, M. (2022). Comparison of conventional and green extraction methods on oil yield, physicochemical properties, and lipid compositions of pomegranate seed oil. *Journal of Food Composition and Analysis*, 114, Article 104747. <https://doi.org/10.1016/j.jfca.2022.104747>
- Ming, L. J., & Yin, A. C. Y. (2013). Therapeutic effects of glycyrrhizic acid. *Natural Product Communications*, 8(3). <https://doi.org/10.1177/1934578X1300800335>, 1934578X1300800335.
- Morelló, J.-R., Motilva, M. A.-J., Tovar, M. A.-J., & Romero, M. A.-P. (2004). Changes in commercial virgin olive oil (cv Arbequina) during storage, with special emphasis on the phenolic fraction. *Food Chemistry*, 85(3), 357–364. <https://doi.org/10.1016/j.foodchem.2003.07.012>
- Shi, Y. F., & Chen, H. L. (2013). Progress in studies on pharmacological activities of *Kadsura coccinea*. *Strait Pharmaceutical Journal*, 25(07), 67–69.
- da Silva Gündel, S., Velho, M. C., Diefenthaler, M. K., Favarin, F. R., Copetti, P. M., de Oliveira Fogaça, A., ... Sagrillo, M. R. (2018). Basil oil-nanoemulsions: Development, cytotoxicity and evaluation of antioxidant and antimicrobial potential. *Journal of Drug Delivery Science and Technology*, 46, 378–383. <https://doi.org/10.1016/j.jddst.2018.05.038>
- Singh, A., & Kumar, V. (2023). Phyto-chemical and bioactive compounds of pumpkin seed oil as affected by different extraction methods. *Food Chemistry Advances*, 2, Article 100211. <https://doi.org/10.1016/j.focha.2023.100211>
- Sun, J., Li, D., Huyen, W., Hong, X., He, S., Huo, J., Jiang, L., & Zhang, Y. (2024). Blue honeysuckle seeds and seed oil: Composition, physicochemical properties, fatty acid profile, volatile components, and antioxidant capacity. *Food Chemistry: X*, 21, Article 101176. <https://doi.org/10.1016/j.fochx.2024.101176>
- Tran, Q. T., Wong, W. F., & Chai, C. L. (2019). The identification of naturally occurring labdane diterpenoid calcaratarin D as a potential anti-inflammatory agent. *European Journal of Medicinal Chemistry*, 174, 33–44. <https://doi.org/10.1016/j.ejmech.2019.04.023>
- Wang, L., Liao, S., Liang, J., Long, H., Xia, X., Chen, Q., & Wei, S. (2021). Analysis and evaluation of nutritional components of *Kadsura coccinea* seeds. *China Oils and Fats*, 46(12), 112–117. <https://doi.org/10.19902/j.cnki.zgyz.1003-7969.2021.03.100>
- Wang, Y., Qayyum, Y., Shehzad, O., Yu, L., Tian, A., Wang, S., Ma, L., Zheng, L., & Xu, L. (2023). Comparative study on quality characteristics of *Bischofia polycarpa* seed oil by different solvents: Lipid composition, phytochemicals, and antioxidant activity. *Food Chemistry: X*, 17, Article 100588. <https://doi.org/10.1016/j.fochx.2023.100588>
- Wei, X., Tao, Y., & Wenlin, Z. (2016). The functional components analysis and application Prospect of *Kadsura coccinea* seed. *Food Research and Development*, 37(12), 1–5. <https://doi.org/10.3969/j.issn.1005-6521.2016.12.001>
- Xiong, X., Yang, M., Fan, S., Luo, T., & Xu, X. (2019). Research on the distribution of phytosterols and fatty acids in 13 kinds of vegetable oils by GC-MS. *China Measurement & Testing. Technology*, 45(02), 64–71. <https://doi.org/10.11857/j.issn.1674-5124.2018040066>
- Xu, Z., Liu, S., Shen, M., Xie, J., & Yang, J. (2022). Evaluation of trans fatty acids, carbonyl compounds and bioactive minor components in commercial linseed oils. *Food Chemistry*, 369, Article 130930. <https://doi.org/10.1016/j.foodchem.2021.130930>
- Yadav, G. G., Manasa, V., Murthy, H. N., & Tumaney, A. W. (2023). Chemical composition and nutraceutical characterization of *Balanites roxburghii* seed oil. *Journal of Food Composition and Analysis*, 115, Article 104952. <https://doi.org/10.1016/j.jfca.2022.104952>
- Yang, K., Wei, Z., Wang, J., Luan, X., Duan, Z., Guo, M., Yang, Q., & Xue, Y. (2021). Optimization of HS-SPME-GC/MS analysis of volatile components in *Camellia* seed oil. *Science and Technology of Cereals, Oils and Foods*, 29(04), 108–115. <https://doi.org/10.16210/j.cnki.1007-7561.2021.04.016>
- Ye, M., Zhou, H., Hao, J., Chen, T., He, Z., Wu, F., & Liu, X. (2021). Microwave pretreatment on microstructure, characteristic compounds and oxidative stability of *Camellia* seeds. *Industrial Crops and Products*, 161, Article 113193. <https://doi.org/10.1016/j.indcrop.2020.113193>
- Zhang, J., Sun-dongWang, P. G., NaWang, W. Y., Li-pingYang, Q. E., ThomasJiao, J. F., & Yu-jie.. (2019). Pinolenic acid ameliorates oleic acid-induced lipogenesis and oxidative stress via AMPK/SIRT1 signaling pathway in HepG2 cells. *European Journal of Pharmacology: An International Journal*, 861. <https://doi.org/10.1016/j.ejphar.2019.172618>
- Zhang, Y., Gu, B., Chen, F., & Liang, A. (2015). Comparative study on high oleic sunflower oil and normal sunflower oil. *Cereals & Oils*, 28(07), 50–52. <https://doi.org/10.3969/j.issn.1008-9578.2015.07.013>
- Zhu, Y. (2019). Content of squalene in vegetable oils and its change during oil processing and utilization. *China Oils and Fats*, 44(12), 136–138. <https://doi.org/10.3969/j.issn.1003-7969.2019.12.026>