Contents lists available at ScienceDirect

Food Chemistry: X

## ELSEVIER



journal homepage: www.sciencedirect.com/journal/food-chemistry-x

### Comparative study on quality characteristics of *Bischofia polycarpa* seed oil by different solvents: Lipid composition, phytochemicals, and antioxidant activity

Yongjin Wang<sup>a,b</sup>, Yijia Su<sup>b,c</sup>, Qayyum Shehzad<sup>d</sup>, Le Yu<sup>b,e</sup>, Ailing Tian<sup>a</sup>, Shihao Wang<sup>a</sup>, Lukai Ma<sup>f</sup>, Lili Zheng<sup>g</sup>, Lirong Xu<sup>a,\*</sup>

<sup>a</sup> Institute of Nutrition and Health, Qingdao University, Qingdao 266071, China

<sup>b</sup> National Engineering Research Center for Functional Food, State Key Lab of Food Science and Technology, Collaborative Innovation Center of Food Safety and Quality

Control in Jiangsu Province, School of Food Science and Technology, Jiangnan University, Wuxi 214122, China

<sup>c</sup> College of Fashion and Art Design, Minnan University of Science and Technology, Fujian 362700, China

<sup>d</sup> National Engineering Laboratory for Agri-product Quality Traceability, Beijing Technology and Business University, Beijing 100048, China

<sup>e</sup> Jiahe Foods Industry CO., LTD, Suzhou 215000, China

<sup>f</sup> Key Laboratory of Green Processing and Intelligent Manufacturing of Lingnan Specialty Food of Ministry and Rural Affairs, College of Light Industry and Food, Zhongkai University of Agriculture and Engineering, Guangzhou 510225, China

<sup>g</sup> National Engineering Research Centre for Intelligent Electrical Vehicle Power System (Qingdao), College of Mechanical & Electronic Engineering, Qingdao University, Qingdao 266071, China

### ARTICLE INFO

### Keywords: Bischofia polycarpa seed oil Solvent extraction Chemical composition Phytochemicals Antioxidant activity Correlations analysis Chemical compounds studied in this article: n-Hexane (PubChem CID: 8058) Isopropanol (PubChem CID: 3776) Methanol (PubChem CID: 887) Chloroform (PubChem CID: 6212) Ethyl acetate (PubChem CID: 8857) Acetone (PubChem CID: 180)

### ABSTRACT

*Bischofia polycarpa* seed oil is rich in nutrition and positively affects on human health. We analyzed and compared the chemical compositions, antioxidant activities, and quality characteristics of *Bischofia polycarpa* seed oils using different solvents and cold-pressing. Hx: Iso (*n*-hexane/isopropanol, 3:2 v/v) had the highest lipid yield (35.13 %), while Folch (chloroform/methanol, 2:1 v/v) had the highest linolenic acid (50.79 %), LnLnLn (43.42 %), and LnLnL (23.43 %). Tocopherols (2108.99 mg/kg) were extracted most efficiently with Folch, whereas phytosterols (3852.97 mg/kg) and squalene (55.21 mg/kg) were extracted most efficiently with petroleum ether. Although the lower phytosterol was obtained using isopropanol, the polyphenol content (271.34 mg GAE/kg) was significantly higher than other solvents, showing the best antioxidant ability. Additionally, polyphenols were observed to be the most significant factor predicting antioxidant activity from the correlation analysis. The above information can provide a useful reference for manufacturers to obtain satisfactory *Bischofia polycarpa* seed oil.

### 1. Introduction

*Bischofia polycarpa* (Levl.) Airy Shaw (*Bischofia polycarpa*) is a deciduous tree belonging to the Dicotyledonous class, Euphorbiaceae, and the genus Autumn Maple. It is a native tree species in China and has been widely cultivated in Jiangsu, Zhejiang, Anhui, Gansu, and Guangzhou. Many articles on *Bischofia polycarpa* have focused primarily on ornamental attributes, the medicinal properties of its roots and leaves, and the pulp for producing wine (Zhang, Yan, & Wu, 2016). In addition, according to traditional Chinese medicine records, the oil content of *Bischofia polycarpa* seed is about 30 %, which has medicinal value and can be safely consumed by humans (Li, Yin, Jing, Wang, Shen, & Li, 2019; Zhang et al., 2016).

Bischofia polycarpa seed abundantly contains polyunsaturated fatty acids, specifically  $\alpha$ -linolenic acid (C18:3), up to 45 % (Li et al., 2019), comparable to flaxseed oil (30–50 %) (Yang et al., 2021) and peony seed oil (40–47 %) (Chang et al., 2020), and much higher than common plant seed oils such as rapeseeds (9–11 %) and soybean (3–13 %) (Abdelghany et al., 2020; Chew, 2020). Numerous health benefits are associated with C18:3, including its ability to protect against chronic inflammation.

https://doi.org/10.1016/j.fochx.2023.100588

Received 30 October 2022; Received in revised form 15 December 2022; Accepted 24 January 2023 Available online 25 January 2023

<sup>\*</sup> Corresponding author. *E-mail address:* lirongxu@qdu.edu.cn (L. Xu).

<sup>2590-1575/© 2023</sup> The Author(s). Published by Elsevier Ltd. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

According to the dietary guidelines, the daily intake of long-chain n-3 polyunsaturated fatty acids is not meeting the requirements of a healthy lifestyle (Yue et al., 2021). Thus, researchers are always looking for bulky sources of these essential fatty acids to meet the growing demands of an increasing population. Additionally, *Bischofia polycarpa* seed oil is rich in C18:3 but also contains other active dietary phytochemicals, including a wide range of phenolic compounds, tocopherols, and phytosterols. *Bischofia polycarpa* seed oil has the potential superiority as a natural source of supplementary lipid nutrients, particularly C18:3.

Cold pressing and solvent extraction are the most common methods of extracting plant oils for commercial use. Cold pressing is a safe and straightforward mechanical process, which can effectively preserve the natural properties of raw materials, such as color, flavor, and other quality attributes (Lee, Kim, Cho, Choi, In, & Kim, 2013). Solvent extraction is a mature vegetable oil extraction technology, which has also been used on an industrial scale because of its simple process and high oil extraction efficiency. However, numerous studies have indicated that multiple solvents can be adopted to attain seed oil. Still, the solvent type may affect the oil's quality characteristics and chemical composition (Tir, Dutta, & Ahmed, 2012; Uoonlue & Muangrat, 2018). Particularly in the oil production process, the minor phytochemicals (such as tocopherols, polyphenols, phytosterols, etc.) need to be extracted simultaneously since their presence can affect the antioxidant activity and nutritional value of the final oil product. Therefore, selecting a suitable solvent is critical to getting satisfactory oil quality.

When selecting a solvent, the influential factors to consider are extraction efficiency, safety, and energy efficiency. Non-polar solvents are suitable for extracting edible oil because of their aliphatic structure and are conducive to extracting lipid nutrients from raw materials (Gao, Liu, Jin, & Wang, 2019). As non-polar solvents are flammable and nonrenewable, they are limited in their use. Oils have also been extracted with alternative polar solvents, such as short-chain alcohols. Several studies indicated that oils obtained from non-polar solvents might show weaker antioxidant activity than those with alcoholic solvents (Bhatnagar & Krishna, 2013; Xie et al., 2017). Although alcoholic solvents are required in high amounts, they possess safer production characteristics. A biodegradable solvent with low evaporation heat is ketones, which have similar physicochemical properties to alcohols. Since alcohols and ketones have relatively strong polarity, they are the better choice for obtaining more polar substances from plant seeds. Also, the combination of non-polar and polar solvents has been applied to yield better oil quality during extraction. The addition of polar solvents effectively enhanced the polarity of the co-solvent system, allowing more compounds with different polarities to be collected. Particularly, Hx: Iso and Folch with low toxicity show advantages in extraction efficiency and energy consumption, which have been applied at the laboratory scale (Tir et al., 2012; Xie et al., 2017). However, to the best of our knowledge, Bischofia polycarpa seed oil obtained by various extraction solvents and cold pressing was not studied in terms of its chemical composition, antioxidant activity, or overall quality attributes.

The present study compared the extraction of *Bischofia polycarpa* seed oil using seven solvent systems, including traditional ethers, alkanes, and new leaching solvents, short-chain alcohols, and ketones, and compared them with cold-pressing. The overall quality of the oil obtained from different extraction solvents was analyzed in detail. This provides a powerful insight for the further development and utilization of *Bischofia polycarpa* seed.

### 2. Materials and methods

### 2.1. Materials and chemicals

We received *Bischofia polycarpa* seeds from Jiangsu, China, and stored them at -80 °C. A mixture of 37 fatty acid methyl esters, phytosterol mixtures, tocopherol mixtures, squalene, gallic acid, and  $5\alpha$ -cholestane were obtained from Sigma Aldrich (Belleforte, PA, USA).

HPLC grade *n*-hexane, isopropanol, ethanol, dichloromethane, ethyl acetate, methanol, and other inorganic reagents were delivered by Sinopharm Medicine Holding Co., Ltd. (Shanghai, China). The supplementary material provides detailed information regarding chemicals used during the study.

### 2.2. Cold pressing

Screw pressing was done using the method described by Xu et al. (2022). In brief, 500 g of *Bischofia polycarpa* seed was pressed with the ZY-22A screw press (Daxiang, Inc., Guangzhou, China) at room temperature. The crude oil temperature was 40 °C, and the oil was centrifuged at 10,000 rpm for 8 min. We immediately transferred the supernatant oils to dark bottles and stored at -20 °C for further analysis.

### 2.3. Solvent extraction

Solvent extraction was carried out according to the method of Gao et al. (2019). Briefly, 40 g of *Bischofia polycarpa* seed power was supplemented with 200 mL of solvents (petroleum ether, *n*-hexane, ethyl acetate, Folch (chloroform/methanol, 2:1 v/v), Hx: Iso (*n*-hexane/isopropanol, 3:2 v/v), acetone and isopropanol, respectively). After homogenizing the mixture (7000 revolutions per minute) at room temperature (25–30 °C) for 8 min, the filtrate was collected using Whatman #4 filter paper. The rotary-evaporator (RE-201, Kexing, China) removed the solvent under vacuum control and a constant temperature water bath (30 °C). The residual solvent was then removed with nitrogen. After obtaining all oil samples, we transferred them into brown bottles and kept at -20 °C until analysis.

### 2.4. Lipid yield

The weight of the *Bischofia polycarpa* seeds and oils were recorded to assess the lipid yield. The lipid yield of the *Bischofia polycarpa* seed was measured according to the given equation: Lipid yield (%) =  $y/a \times 100$ %. where a is the *Bischofia polycarpa* seed weight (g) and y is the *Bischofia polycarpa* seed oil (g) mass.

### 2.5. Quality characteristics

According to the Association of Official Analytical Chemists AOAC (Official, 2000) (AOAC), the acid value, saponification value, iodine value, and peroxide value of *Bischofia polycarpa* seed oil were determined.

### 2.6. Fatty acid composition

The fatty acid composition was detected according to the method described by Xu et al. (2022). 50 mg of oil sample was combined with 2 mL hexane and dissolved with 500  $\mu$ L of 2 mol/L methanolic potassium hydroxide. Then, FAMEs were determined by a gas chromatography (GC) system (7820A; Agilent Technologies, CA, USA) equipped with a flame ionization detector (FID). The trace TR-FAME capillary column (60 m × 0.25 mm, 0.25  $\mu$ m, Thermo Fisher, Shanghai, China) was used for separation. The injector and FID temperature were set as 250 °C, and the injection volume was 1  $\mu$ L. The oven temperature was set as follows: 3 min at 60 °C, from 60 °C to 175 °C at 5 °C/min, from 175 °C to 220 °C at 2 °C/min, 10 min at 220 °C. Fatty acids were determined by comparing retention times with the 37 FAMEs standards, and their contents were expressed as relative percentages of total area.

### 2.7. Triacylglycerol

Triacylglycerol composition was conducted according to the method of Gao et al. (2019). Agilent 7890A GC was equipped with an FID detector. The temperature of the FID and injector were set at 360 °C and

375 °C, respectively. The DB-17HT capillary column (0.15  $\mu m$ , 0.25 mm  $\times$  15 m, Agilent) was used for separation. The temperature rise of the chromatographic column was from 250 °C to 340 °C at 5 °C/min and then kept at 340 °C for 32 min. The split ratio was 1:100. Nitrogen was used as carrier gas at a rate of 1 mL/min. The injection volume was 1.0  $\mu L$ . The contents of each triacylglycerol were according to the relative proportion of the total area.

### 2.8. Tocopherol

Tocopherols were identified by following the method of Xu et al. (2022) with some modifications. Oil samples (0.5 g) were mixed with 1.5 mL of *n*-hexane and filtered with a 0.22 µm nylon syringe. Then, 10 µL of the sample was injected into an LC system (Waters 1525, Waters, Massachusetts, USA) with a photodiode array detector. For separation, we used a silica column (4.6 mm  $\times$  250 mm, 5 µm; Hanbon, Jiangsu, China), and the column temperature was set at 40 °C. *n*-Hexane/isopropanol (98.5:1.5 v/v) was used at a flow rate of 0.8 mL/min. Peaks on chromatograms were discovered at 295 nm. Further, tocopherols were identified and quantified by comparing with their standards, and their levels were expressed as mg/kg.

### 2.9. Phytosterol and squalene

Phytosterols and squalene were determined based on the method (Xu, Zhu, Liu, Karrar, Ouyang, & Li, 2022) with some modifications. Thermo Scientific ISQ<sup>TM</sup> GC–MS equipped with ion source (T = 250 °C), injector (T = 280 °C), and FID (T = 280 °C) was applied. DB-5 capillary column (0.25 µm, 30 m × 0.25 mm; Agilent) was adopted for separation. The initial column temperature was set as follows: 200 °C (1 min) – 300 °C (10 °C/min) – 300 °C (18 min). The ionization mode was the electron impact ion source, and the mass range (*m*/*z*) was 20–550 Da. The injection volume was 1.0 µL. Helium (99.999 % purity) was used as a carrier gas (1.2 mL/min), and the split ratio was 100:1. 5α-Cholestane was used as the internal standard for quantification, and phytosterol and squalene contents were presented as mg/kg for each sample.

### 2.10. Polyphenol

A modified Folin-Ciocalteau approach was used to detect the polyphenols (Gao et al., 2019). Oil samples (1.0 g) were dissolved with 6 mL hexane. Polyphenols were extracted using a Sepax Generik Diol tube (Sepax Technologies, Inc., Newark, DE, USA) pretreated with 6 mL of *n*-hexane and methanol. Then, the solid-phase extraction (SPE) was washed using 4 mL of *n*-hexane and *n*-hexane/ethyl acetate (v/v = 9:1). Further, methanol was used as an eluent to obtain the *Bischofia polycarpa* seed oil polar extract solution from the SPE column in 10 mL dark volumetric flasks. Folin Ciocalteu reagent (0.5 mL) was added to the solution (5 mL). Finally, 1 mL Na<sub>2</sub>CO<sub>3</sub> (10 %) was added, and the volume was adjusted with ultrapure water to 10 mL. After 2 h in darkness, the absorbance at 765 nm was measured. The quantitative results were reported in mg of gallic acid equivalents per kg of *Bischofia polycarpa* seed oil sample (mg/kg).

### 2.11. Antioxidant activity

We explored the oxidative stability index and antioxidant activities of *Bischofia polycarpa* seed oil by following the procedure previously done with some modifications (Wang et al., 2021; Gao et al., 2019). The Supplementary Materials provide detailed information about these methods.

### 2.12. Data analysis

All experiments were conducted in triplicate, with values expressed as means  $\pm$  SD, and analyzed the data by SPSS (23.0) and Origin (2019).

Obtained values were statistically analyzed using one-way ANOVA with Tukey's test, and significant differences were established at P < 0.05. Pearson's correlation analysis and multiple linear regression (MLR) were used to analyze the correlation between antioxidant activity and minor phytochemicals.

### 3. Results and discussion

### 3.1. Lipid yield

As illustrated in Fig. 1, the lipid yield of *Bischofia polycarpa* seeds extracted by various solvents and pressing methods were significantly different, ranging from 19.12 to 35.13 % (P < 0.05). Following the results of vegetable oil processing, the lipid yield of solvent extraction was significantly higher than cold-pressing. In cold-pressing, oil was not extracted fully from raw materials, which led to relatively low oil extraction efficiency. Nonetheless, the *Bischofia polycarpa* seeds in this study produced higher lipid yields than other C18:3-rich seeds, such as peony seeds (Wang, Xu, Wu, Zhou, Ren, & Yang, 2015), comparable to flaxseeds and hempseeds (Zeng et al., 2022).

Among these seven solvents, Hx: Iso method had the highest lipid vield, followed by Folch, and acetone exhibited the lowest value (seen in Fig. 1). The lipid yield of *n*-hexane and petroleum ether was not significantly different (P > 0.05). Overall, the polar solvents (acetone or isopropanol) extracted lower lipid yields than non-polar solvents (nhexane or petroleum ether), which was in line with the theory of similarity and mutual solubility. However, it was observed that the lipid yield of solvent mixtures (Hx: Iso and Folch) exceeded that of the nonpolar solvents. Tir et al. (2012) reported that slightly polar solvents might accelerate the cell membrane destruction, allowing seed oil to be extracted more thoroughly, as well as some specific interactions involving hydrogen bonding with the ester groups of triglycerides. Parallel results were also described by Bhatnagar and Krishna (2013), who discovered that increasing the polarity of the solvent may be involved in obtaining higher lipid yields from plant seeds. Polar solvents reduce the surface tension difference of phase boundary and improve the phase separation (Berezin, Tur'yan, Kuselman, & Shenhar, 1996). However, single polar solvents (acetone and isopropyl alcohol) showed the lowest lipid yield in this paper, possibly because the excessive increase in solvent polarity would limit the solubility of lipids and lead to the hydrolysis (solvolysis) of some lipids (Russin, Boye, Arcand, & Rajamohamed, 2010).

### 3.2. Quality characteristics

As displayed in Table 1S, the acid value of *Bischofia polycarpa* seed oil ranged from 2.08 to 5.01 mg KOH/g, with a significant difference among solvents (P < 0.05). The acid value of the oil extracted by ethyl acetate was the highest, whilst the oil samples extracted by *n*-hexane revealed the lowest values. Overall, the acid value of strong polar solvents was higher than that of weak polar solvents. Acid value mainly reflects the refining degree of oil and the preservation of raw materials. In addition to free fats, pigments, and phospholipids, other substances may be extracted during extraction of *Bischofia polycarpa* seeds. Meanwhile, increasing the solvent polarity accelerated the destruction of lipids and free fatty acids binding to lipoproteins or cell membranes (Zhang, Li, Zheng, Liu, Ge, & Zhang, 2017). Hence, the levels of free fatty acids, fatty alcohols, etc., in oils extracted with stronger polar solvents might be higher, resulting in higher acid values.

There was also a significant difference in the saponification value of *Bischofia polycarpa* seed oil prepared with different solvents, ranging from 156.04 to 165.30 mg KOH/g (P < 0.05) (Table 1S). An oil's saponification value is the potassium hydroxide required to saponify 1 g of oil. According to previous studies, low-molecular-weight fatty acids have higher saponification values, meaning they contain more free fatty acids in the oil (Zhang et al., 2017; Zhu, Zhang, Xie, & Liu, 2016). In this



**Fig. 1.** Lipid yield of different extraction solvent from studied samples. Different letters on the top of data bars indicate significant differences (Tukey's test, P < 0.05) between mean values ( $\pm$  SD, n = 3).

study, *Bischofia polycarpa* seed oil samples with high acid values also confirmed these results.

The peroxide value of *Bischofia polycarpa* seed oil obtained with nine solvents ranged from 7.02 to 11.10 mmol/kg (Table 1S). The peroxidation value of oil extracted with mixed solvents (Folch and Hx: Iso) was lower than the traditional single solvent. Moreover, the iodine value of *Bischofia polycarpa* seed oil varied from 177.60 to 185.02 g/100 g, except that the iodine value of oil extracted with acetone was low, the other kinds of differences were not significant (P > 0.05). Oil has a certain iodine value based on the number of unsaturated double bonds in it, and the greater the number, the higher the value. In some vegetable oils, iodine values range from 90 to 130, for example, soybean oil, corn oil, and canola oil (Li et al., 2018), which shows that *Bischofia polycarpa* seed oil contains many unsaturated double bonds and can be used as a natural source of unsaturated fatty acids.

### 3.3. The composition of fatty acid and triacylglycerol

Table 1 lists the fatty acid compositions of Bischofia polycarpa seed oil extracted by different methods. Linolenic acid (C18:3) (45.47-50.79%), linoleic acid (C18:2) (23.60-27.61 %), oleic acid (C18:1) (11.15-13.31 %), and palmitic acid (C16:0) (6.17-6.79 %) were the major fatty acids. C18:2 and C18:3 of Bischofia polycarpa seed oil accounted for more than 70 %, indicating that it can serve as a natural dietary source of essential fatty acids supplement. In contrast, gadoleic acid (C20:1), palmitoleic acid (C16:1), arachidonic acid (C20:0), and heneicosylic acid (C21:0) were detected in smaller amounts. The unsaturated fatty acid (UFAs) content of Bischofia polycarpa seed oil obtained with different solvents was significantly different (P < 0.05). Values of C18:3 have declined in the order of Folch > petroleum ether > n-hexane > Hx: Iso > ethyl acetate > acetone > isopropanol > cold pressing. Meanwhile, n-hexane extracts presented the maximum C18:2, whereas the oil achieved using Folch exhibited the minimum. Additionally, Hx: Iso extracts showed a significantly higher C18:1 than other methods. Similar results were

Table 1

Lifect of callecton solvent on the fatty acta compositions (70) and that visive of compositions (70) of <i>Discional Dality</i> and seed on	Effect of extraction solvent on the fatty	v acid compositions (%) and	triacylglycerol compositions (%	<ul> <li>of Bischofia polycarpa seed oils.</li> </ul>
---	---	-----------------------------	---------------------------------	---

Composition	$\text{Mean}\pm\text{SD}~(n=3)$							
	<i>n</i> -Hexane	Petroleum ether	Ethyl acetate	Hx: Iso	Folch	Acetone	Isopropanol	Cold pressing
FA (%)								
C16:0	$6.22\pm0.03^{\rm c}$	$6.20\pm0.07^{\rm bc}$	$6.17\pm0.06^{c}$	$6.23\pm0.01^{\rm c}$	$6.23\pm0.0^{2c}$	$6.25\pm0.01b^{c}$	$6.31\pm0.05^{\rm b}$	$6.79\pm0.09^{a}$
C18:0	$4.72\pm0.08^{\rm b}$	$4.75\pm0.04^{\rm b}$	$\textbf{4.76} \pm \textbf{0.03}^{b}$	$4.74 \pm \mathbf{0.06^{b}}$	$4.72\pm0.06^{\rm b}$	$4.78\pm0.15^{\rm b}$	$4.82\pm0.05^{\rm b}$	$\textbf{5.19} \pm \textbf{0.00}^{a}$
C20:0	$0.28\pm0.01^{\rm b}$	$0.32\pm0.00^{\rm b}$	$0.26\pm0.01^{\rm b}$	$0.21\pm0.02^{\rm c}$	$0.22\pm0.00^{\rm c}$	$0.23\pm0.02^{\rm bc}$	$0.11\pm0.01^{\rm d}$	$0.55\pm0.01^{a}$
C21:0	$0.06\pm0.00^a$	$0.07\pm0.00^{a}$	$0.08\pm0.01^{a}$	$0.05\pm0.02^a$	$0.06\pm0.00^a$	$0.08\pm0.01^a$	$0.05\pm0.00^{a}$	$0.06\pm0.00^{a}$
C16:1	$0.27\pm0.01^{\rm f}$	$0.67\pm0.02^{\rm e}$	$1.02\pm0.03^{\rm d}$	$0.13\pm0.02~^{g}$	$1.39\pm0.05^{\rm c}$	$1.72\pm0.06^{\rm ab}$	$1.91\pm0.13^{\rm a}$	$1.65\pm0.10^{\rm b}$
C20:1	$0.75\pm0.00^{\rm b}\rm c$	$0.77\pm0.02^{\rm b}$	$0.83\pm0.06^{\rm b}$	$0.65\pm0.03^{\rm c}$	$0.85\pm0.08^{\rm b}$	$1.37\pm0.10^{\rm a}$	$1.48\pm0.02^{a}$	$0.93\pm0.03^{\rm b}$
C18:1	$11.51\pm0.30^{\rm bc}$	$12.58\pm0.12^{\rm b}$	$13.02\pm0.11^{\text{a}}$	$13.31\pm0.20^{\rm a}$	$12.14\pm0.18^{\rm b}$	$11.25\pm0.13^{\rm c}$	$11.24\pm0.06^{c}$	$11.15\pm0.15^{\rm c}$
C18:2	$27.61\pm0.17^{\rm b}$	$25.79 \pm 0.09^{c}$	$26.28\pm0.10^{\rm c}$	$26.55\pm0.28^{\rm bc}$	$23.60 \pm 0.21^{d}$	$26.51\pm0.32^{\rm bc}$	$27.05 \pm 0.08^{b}$	$28.21 \pm 0.20^a$
C18:3	$48.58\pm0.10^{\rm b}$	$48.85\pm0.03^{b}$	$\textbf{47.58} \pm \textbf{0.14}^{c}$	$48.13 \pm \mathbf{0.07^{b}}$	$50.79\pm0.19^a$	$\textbf{47.45} \pm \textbf{0.10}^{c}$	$47.03 \pm \mathbf{0.30^c}$	${\bf 45.47 \pm 0.25^{d}}$
SFA	$11.28\pm0.09^{\rm b}$	$11.34\pm0.12^{\rm b}$	$11.27\pm0.12^{\rm b}$	$11.23\pm0.09^{\rm b}$	$11.23\pm0.12^{\rm b}$	$11.34\pm0.20^{\rm b}$	$11.29\pm0.13^{\rm b}$	$12.59\pm0.10^{a}$
MUFA	$12.53\pm0.10^{\rm d}$	$14.02\pm0.08^{b}$	$14.87\pm0.13^{\text{a}}$	$14.09\pm0.30^{ab}$	$14.38\pm0.22^{\text{a}}$	$14.34\pm0.18^{a}$	$14.63\pm0.09^{a}$	$13.73\pm0.15^{\rm c}$
PUFA	$76.19 \pm 0.19^{a}$	$74.64 \pm 0.10^{\mathrm{b}}$	$73.86\pm0.14^{c}$	$74.68 \pm 0.16^{\mathrm{b}}$	$74.39\pm0.32^{\rm b}$	$73.96\pm0.25^{\rm bc}$	$74.08 \pm 0.12^{\mathrm{b}}$	$73.68\pm0.14^{c}$
Triacylglycerol (%)								
LnLnLn	$41.21\pm0.20^{c}$	$41.75\pm0.32^{\rm bc}$	$42.34\pm0.10^{\rm b}$	$42.02\pm0.25^{\rm b}$	$43.42\pm0.13^{a}$	$41.04\pm0.12^{c}$	$40.88\pm0.14^{\rm d}$	$39.32\pm0.35^{\rm d}$
LnLnL	$22.08\pm0.05^{\rm b}$	$22.07\pm0.19^{\rm b}$	$23.09 \pm \mathbf{0.20^a}$	$22.99\pm0.10^a$	$23.43\pm0.21^{a}$	$20.32\pm0.10^{\rm b}$	$20.58\pm0.18^{\rm b}$	$20.19\pm0.14^{\rm b}$
LnLnO	$7.12\pm0.05^{\rm c}$	$6.48\pm0.10^{\rm d}$	$6.38\pm0.12^{\rm d}$	$5.49\pm0.10^{\rm e}$	$5.17\pm0.09^{\rm e}$	$8.66\pm0.20^{\rm b}$	$9.17\pm0.15^{\text{a}}$	$6.39\pm0.08^{\rm d}$
LnLL	$4.28\pm0.06^{\rm b}$	$3.87\pm0.03^{\rm c}$	$4.09\pm0.10^{bc}$	$3.23\pm0.02^{\rm d}$	$2.95\pm0.10^{\rm d}$	$4.60\pm0.12^{a}$	$4.73\pm0.06^{a}$	$\textbf{4.11} \pm \textbf{0.03}^{b}$
LnLnP	$8.87 \pm \mathbf{0.05^{b}}$	$9.12\pm0.05^{a}$	$9.34\pm0.10^{a}$	$\textbf{8.48} \pm \textbf{0.06}^{c}$	$9.31\pm0.09^a$	$8.57 \pm 0.04^{c}$	$8.87 \pm \mathbf{0.06^{b}}$	$8.96\pm0.10^{ab}$
OLLn	$1.37\pm0.07^{\rm e}$	$1.68\pm0.06^{\rm d}$	$\textbf{2.43} \pm \textbf{0.04}^{\rm b}$	$3.13\pm0.11^{\rm a}$	$1.15\pm0.05^{\rm e}$	$2.92\pm0.06^a$	$2.02\pm0.10^{\rm c}$	$1.97\pm0.02^{\rm c}$
LnLP	$3.71\pm0.06^{\rm c}$	$3.63\pm0.12$ <sup>cd</sup>	$3.51\pm0.05^{\rm d}$	$3.64\pm0.05~^{\rm cd}$	$\textbf{3.47} \pm \textbf{0.12d}$	$4.33\pm0.10^{\rm b}$	$3.85\pm0.05^{\rm c}$	$5.29\pm0.06^{a}$
OOLn + OLL	$4.68\pm0.10^{a}$	$4.00\pm0.07^{c}$	$2.66\pm0.20^{\rm d}$	$3.87\pm0.07^{\rm c}$	$4.52\pm0.02^{\rm b}$	$2.89\pm0.03^{\rm d}$	$3.71\pm0.01^{\rm c}$	$\textbf{4.49} \pm \textbf{0.05}^{\rm b}$
LnOP	$3.67\pm0.03^{\rm b}$	$3.79\pm0.06^{\rm b}$	$\textbf{3.48} \pm \textbf{0.05^c}$	$3.33\pm0.08^{\rm c}$	$3.67\pm0.05^{\rm b}$	$3.60\pm0.05^{\rm b}$	$3.32\pm0.02^{\rm c}$	$5.83\pm0.04^{a}$
OOL	$0.53\pm0.03^{\rm b}$	$0.89\pm0.00^{a}$	$0.20\pm0.00^{c}$	$0.99\pm0.04^{a}$	$0.50\pm0.02^{\rm b}$	$0.55\pm0.05^{\rm b}$	$0.43\pm0.01^{\rm b}$	$0.82\pm0.02^{\text{a}}$
PPLn	$0.57\pm0.00^a$	$0.60\pm0.02^{a}$	$0.59\pm0.02^{a}$	$0.59\pm0.01^a$	$0.62\pm0.02^{a}$	$0.58\pm0.01^{a}$	$0.58\pm0.04^a$	$0.61\pm0.01^{a}$
PPO	$0.56\pm0.05^a$	$0.63\pm0.04^{a}$	$\textbf{0.67} \pm \textbf{0.10}^{a}$	$0.63\pm0.04^{a}$	$0.56\pm0.06^a$	$0.57\pm0.05^a$	$0.61\pm0.03^a$	$0.58\pm0.02^{a}$
OOS	$1.35\pm0.10^{c}$	$1.49\pm0.03^{b}$	$1.22\pm0.03^{\rm d}$	$1.61\pm0.04^a$	$1.23\pm0.05^{\rm d}$	$1.37\pm0.03^{c}$	$1.25\pm0.10~^{cd}$	$1.44\pm0.04^{bc}$

Values represent the mean  $\pm$  SD of triplicated measurements.

Different letters indicate significant difference (P < 0.05).

Abbreviations are: C16:0, P, Palmitic acid; C18:0, S, Stearic acid; C20:0, Arachidic acid; C21:0, Heneicosylic acid; C16:1, Palmitoleic acid; C20:1, Gadoleic; C18:1, O, Oleic acid; C18:2, Linoleic acid, L; C18:3, Ln, α-linolenic acid; SFA, Saturated fatty acid; MUFA, Monounsaturated fatty acid; PUFA, Polyunsaturated fatty acid.

published by Uoonlue and Muangrat (2018). They observed that the percentages of UFAs (including C18:3, C18:2 and C18:1) in *Camellia sinensis* seed oil were treated with different solvents varied markedly with the polarity. These results demonstrated that the solvents obtained UFAs with some selectivity. Notably, the solvents used in our study did not significantly affect saturated fatty acids (SFA) content (P > 0.05). In another study on krill oil extraction, Xie et al. (2017) also confirmed no significant differences in SFA levels, despite the different types of solvents administered for extraction.

As shown in Table 1, fourteen triacylglycerol components were also identified in Bischofia polycarpa seed oil, mainly including LnLnLn (39.32-43.42 %), LnLnL (20.19-23.43 %), LnLnP (8.48-9.34 %), followed by LnLnO (5.17-9.17 %), LnLL (2.95-4.73 %), LnLP (3.47-5.29 %), LnOP (3.32-5.83 %), OOLn + OLL (2.66-4.68 %), and OLLn (1.15–3.13 %). We also observed that the triglyceride types in Bischofia polycarpa seed oils extracted with seven solvents were identical, but the contents differed (P < 0.05). LnLnLn levels were highest and lowest in Bischofia polycarpa seed oil extracted by Folch and isopropanol, respectively, while OOLn + OLL levels were highest in *n*-hexane extraction. Both isopropanol and acetone had a higher level of LnLnO and LnLL. Further, Folch and ethyl acetate obtained oil samples containing more LnLnL. However, there was no significant difference in the contents of PPLn and PPO in different types of Bischofia polycarpa seed oil extracted by solvent (P > 0.05), which was about 0.60 %. According to these findings, Bischofia polycarpa seed oil's triacylglycerol composition is closely associated with its fatty acid composition. Similarly, Gao et al. (2019) used five different extraction solvents to obtain the triglycerides contents in walnut oil and noted that the triglycerides contents were influenced remarkably by the fatty acid composition.

# UFA from oxidizing. As shown in Table 2a and Fig. 2, total tocopherols (1698.40 to 2108.99 mg/kg) were determined in the studied oils (P < 0.05). The *Bischofia polycarpa* oil sample collected using Folch exhibited the highest total tocopherols, followed by those extracted with Hx: Iso, ethyl acetate and *n*-hexane, then cold pressing, petroleum ether, and finally, acetone and isopropanol. The outcomes showed that mixed solvents might be more favorable for extracting tocopherol compared with single solvents. Further, four tocopherol homologs ( $\alpha$ -, $\beta$ -, $\gamma$ -, and $\delta$ -tocopherol) were also detected in this study (Table.2a), with the predominance of $\gamma$ -tocopherol. It's worth noting that $\gamma$ -tocopherol levels in this study were almost 3–4 times greater than flaxseed, peony, and perilla seed oil reported by Yang et al. (2018), indicating that *Bischofia polycarpa* seed oil with high $\gamma$ -tocopherol content is a high-performance nutrient source.

Furthermore, *Bischofia polycarpa* seed oil extracted by *n*-hexane presented the highest content in  $\alpha$ - and  $\beta$ -tocopherol, whilst Hx: Iso and Folch were observed to be more suitable for obtaining  $\gamma$ -tocopherol. The variation of solvent polarity may cause the difference in tocopherol homologues content. According to Huang et al. (2021) and Mendonca et al. (2005), the polarity of tocopherol decreases in the order of  $\delta$ -,  $\gamma$ -,  $\beta$ -, and  $\alpha$ - tocopherol due to the increase of methyl group in the chromanol ring. Therefore, the extraction solvents with greater polarity tend to yield higher levels of  $\gamma$ -tocopherol. Even though the compositions of tocopherols in *Bischofia polycarpa* seed oil extracted with different solvents were the same, the proportions were not. Likewise, Bhatnagar and Krishna (2013) reported similar observations, who investigated commercial Niger seed oil obtained using six solvents (petroleum ether, hexane, acetone, ethanol, methanol, and chloroform) with distinctly different levels in tocopherol homologue.

### 3.4. Tocopherol content

3.5. Phytosterol content

Tocopherol is an effective indicator of plant oil quality and prevents

Phytosterols are important bioactive compounds that contribute to

### Table 2a

Effect of extraction solvent on the phytochemicals content (mg/kg) of Bischofia polycarpa seed oils.

Compounds	$\begin{array}{l} \text{Mean}\pm\text{SD} \text{ (n} \\ = 3 \text{)} \end{array}$							
	<i>n</i> -Hexane	Petroleum ether	Ethyl acetate	Hx: Iso	Folch	Acetone	Isopropanol	Cold pressing
Tocopherols (mg/kg)								
α-Tocopherol	$196.77\pm2.31^{a}$	$182.99\pm2.09^b$	$170.20\pm3.00^{c}$	$159.86\pm2.04^{e}$	${\begin{array}{c} 168.41 \pm \\ 3.30^{d} \end{array}}$	$162.93\pm2.07^d$	$163.26\pm1.18^{\text{d}}$	$156.68\pm1.34^{\text{e}}$
β-Tocopherol	$207.26 \pm 3.40^{a}$	$203.84\pm1.06^a$	$206.28 \pm 0.82^{a}$	$195.14\pm1.40^{b}$	$182.12\pm0.79^{c}$	$183.76 \pm 3.20^{c}$	$181.49\pm2.84^c$	$184.66\pm2.01^{c}$
γ-Tocopherol	$\begin{array}{c} 1316.50 \pm \\ 12.00^{\rm d} \end{array}$	$\begin{array}{c} 1258.01 \ \pm \\ 9.90^{\rm f} \end{array}$	$1430.23 \pm 12.04^{\rm c}$	$1602.79 \pm 12.53^{ m b}$	$1680.23 \pm 15.32^{\rm a}$	$\frac{1290.57}{18.08^{de}} \pm$	$1280.17 \pm 12.11^{\rm e}$	$\begin{array}{c} 1344.09 \pm \\ 13.05^{d} \end{array}$
δ-Tocopherol	$71.46 \pm 1.00^{bc}$	$\textbf{70.08} \pm \textbf{0.42}^c$	$72.65 \pm 0.30^{b}$	$\textbf{78.71} \pm \textbf{0.62}^{a}$	$\textbf{78.23} \pm \textbf{0.41}^{a}$	$75.55\pm1.03^{ab}$	$\textbf{73.48} \pm \textbf{1.63}^{b}$	$\textbf{72.47} \pm \textbf{0.90}^{b}$
Sum	$\begin{array}{c} 1791.99 \pm \\ 21.10^{\rm d} \end{array}$	$\begin{array}{c} 1714.92 \pm \\ 19.40^{e} \end{array}$	$\begin{array}{l} 1879.36 \ \pm \\ 20.40^{c} \end{array}$	$\begin{array}{c} 2036.50 \ \pm \\ 23.12^{b} \end{array}$	$\begin{array}{l} 2108.99 \pm \\ 25.05^{a} \end{array}$	$\begin{array}{l} 1712.81 \ \pm \\ 21.01^{e} \end{array}$	$\begin{array}{c} 1698.40 \pm \\ 15.90^{\rm e} \end{array}$	$\begin{array}{c} 1757.90 \pm \\ 18.02^{de} \end{array}$
Phytosterols (mg/kg)								
Campesterol	$200.23 \pm 1.89^{bc}$	$206.64\pm0.60^b$	$187.63\pm3.40^{e}$	$232.04\pm6.02^a$	$172.30\pm4.02^{\rm f}$	$\underset{cd}{195.48}\pm2.05$	$199.27 \pm 3.01^{\circ}$	$193.21 \pm 1.02^{d}$
Stigmasterol	$242.30\pm2.32^{a}$	$\textbf{221.88} \pm \textbf{1.82}^{b}$	$170.81\pm2.80^{c}$	$228.33\pm3.02^{b}$	${\begin{array}{c} 227.12 \pm \\ 1.02^{b} \end{array}}$	$138.80\pm0.50^d$	$122.52\pm0.61^{e}$	$226.71\pm0.30^{b}$
β-Sitosterol	$2705.32 \pm 23.57^{c}$	$3343.85 \pm 34.27^{a}$	$\begin{array}{c} 2520.00 \ \pm \\ 30.21^{d} \end{array}$	${2848.23} \pm \\ {19.95}^{\rm b}$	$2680.49 \pm 26.57^{c}$	$\begin{array}{c} 2560.80 \pm \\ 18.74^{d} \end{array}$	$\begin{array}{c} 2201.89 \pm \\ 20.06^{e} \end{array}$	$2765.01 \pm 25.63^{c}$
$\Delta$ -7-Avenasterol	$13.40\pm0.14^{e}$	$19.02\pm0.20^{d}$	$18.15\pm0.28^{\text{d}}$	$31.33 \pm 0.06^a$	$\textbf{32.00} \pm \textbf{0.18}^{a}$	$20.79\pm0.20^{c}$	$21.25\pm0.35^{bc}$	$22.56\pm0.50^b$
$\Delta$ -5-Avenasterol	$23.90\pm0.06^{\rm b}$	$20.70\pm0.30^{c}$	$18.49 \pm 0.28^{d}$	$30.61\pm0.13^a$	$33.02 \pm 0.50^{a}$	$25.82\pm0.32^{\rm b}$	$16.67\pm0.10^{\rm e}$	$25.06\pm0.07^{\rm b}$
Fucosterol	$48.05 \pm 0.62^{bc}$	$40.88\pm0.20^{\rm d}$	$45.91 \pm 0.07^{c}$	$60.61 \pm 1.00^{\mathrm{a}}$	$63.02\pm0.53^{\rm a}$	$35.82\pm0.27^{\rm f}$	$41.74 \pm 0.70^{e}$	$52.04\pm0.10^{\text{b}}$
Sum	$3233.20 \pm 30.01^{\circ}$	$3852.97 \pm 45.33^{a}$	$2960.99 \pm 38.31^{d}$	$3431.15 \pm 28.49^{b}$	$3207.95 \pm 30.42^{c}$	$\begin{array}{r} 2977.51 \pm \\ 28.77^{\rm d} \end{array}$	$2603.34 \pm 34.06^{\rm e}$	$3284.59 \pm 26.78^{c}$
Squalene (mg/kg)	$55.13\pm0.52^{a}$	$55.21\pm0.35^a$	$49.82\pm0.20^{c}$	$52.32\pm0.34^{b}$	$52.41\pm0.08^{ab}$	$41.65\pm0.73^{d}$	$41.15\pm0.48^{d}$	$37.72 \pm 0.30^{e}$
Polyphenols (mg GAE/kg)	$150.25\pm6.38^{f}$	$163.92\pm3.85^e$	$203.65\pm5.43^{\text{d}}$	$224.51\pm1.00^{\text{c}}$	$\begin{array}{c} 240.97 \pm \\ 0.84^b \end{array}$	$264.20\pm2.94^a$	$271.34\pm6.70^a$	$242.87\pm2.90^b$

All data represents the mean of three replications (Mean  $\pm$  SD).

Values in the list with different letters are significant difference at P < 0.05.



Fig. 2. Circos plot visualizing the content of phytochemicals of studied samples.

the prevention of coronary heart disease. As shown in Table 2a, Bischofia polycarpa seed oils contain different levels of phytosterols depending on the solvent used. Phytosterol individuals were identified and quantified, including  $\beta$ -sitosterol, stigmasterol, campesterol, fucosterol,  $\Delta$ -5-avenasterol, and  $\Delta$ -7-avenasterol. Among these homologs,  $\beta$ -sitosterol content was the highest, accounting for about 80 % of phytosterols. It is worth noting that, compared with other C18:3-rich oils, the content of  $\beta$ -sitosterol in *Bischofia polycarpa* seed oil was significantly higher than that in flaxseed or camelina seed oil (Piravi-vanak et al., 2022; Zeng et al., 2022), but similar to that in peony seed oil (Chang et al., 2020). The highest phytosterol content was obtained by petroleum ether extraction (3852.97 mg/kg), whereas the lowest was obtained by isopropanol extraction (2603.34 mg/kg) (Table 2a and Fig. 2). Xie et al. (2017) observed that the solubilization ability of phytosterols in polar solvents was lower than that in non-polar solvents. Polar solvents cannot penetrate the lipid chain region because the hydrophobic nature of this structure repels it (Tir et al., 2012). For the same reason, phytosterols are weakly soluble in polar solvents. Nevertheless, the results of *n*-hexane and Hx: Iso treatments were just the opposite, that is, Hx: Iso showed a better ability to extract phytosterols than *n*-hexane. This result may be because isopropanol in the binary mixture allows the desorption and dissolution of the lipid bilayer, which facilitates the solubilization of phytosterols in the solvent mixture (Tir et al., 2012).

In addition, the extraction efficiency of phytosterols may not only be affected by polarity because, in this study, phytosterol content in ethyl acetate (intermediate properties, but closer to nonpolar) and acetone (polar) did not differ significantly (P > 0.05). Indeed, Dabrowski et al. (2020) reported that extracting desired targeted compounds by organic solvents might also be related to other parameters, including molecular volume, molecular weight, and topological polar surface area. All these factors influence the mass flow resistance of solutes through plant cell membranes and cell walls. On the other hand, we observed significant differences in the content of phytosterol homologs (P < 0.05). *Bischofia polycarpa* seed oil obtained with *n*-hexane had the highest stigmasterol levels, but the  $\Delta$ -7-avenasterol was the lowest. The efficiency of extracting  $\Delta$ -5-Avenasterol and fucosterol in a mixed solvent (Hx: Iso and Folch) was higher than that in a single solvent system.

### 3.6. Squalene content

Squalene is an unsaturated hydrocarbon with six isoprene units and has been proven effective in reducing serum cholesterol levels. As described in Table 2a and Fig. 2, squalene content ranged from 37.72 to 55.21 mg/kg, indicating that there was a significant difference between the extracted oil samples (P < 0.05). In descending order, squalene content was petroleum ether > n-hexane > Folch > Hx: Iso > ethyl acetate > acetone > isopropanol. Dbrowski et al. (2020) also reported that squalene has non-polar properties and shows a stronger affinity for non-polar solvents than polar solvents. Our findings further confirmed that squalene extraction efficiency was higher with non-polar solvents. Furthermore, it was found that regardless of the solvent type, the content of squalene obtained using solvents was significantly (P < 0.05) higher than that of cold-pressed oil. Likewise, Shi et al. (2018) compared the squalene contents of Torreya grandis seed oil obtained using different extraction methods and observed that the cold-pressed oil contained fewer squalene. These findings confirmed that solvent extraction had more advantages than mechanical pressing for squalene extraction.

### 3.7. Polyphenols content

Polyphenols are well known for their powerful antioxidant properties and ability to inhibit lipid peroxidation. As shown in Table 2a and Fig. 2, the levels of polyphenols varied markedly among *Bischofia polycarpa* seed oils (from 150.25 to 271.34 mg GAE/kg), demonstrating that polyphenols were affected by solvent types (P < 0.05). A higher level of polyphenol was extracted by isopropanol and acetone, while the lowest level was extracted by *n*-hexane. Meanwhile, compared with petroleum ether, the content of polyphenols obtained by mixed solvent treatment was higher. These results showed that the increased solvent polarity might improve the possibility of extracting more polyphenols. A similar phenomenon was also reported by Zhang et al. (2017). They found that ketones and alcohol solvents had obvious advantages in extracting polyphenols from *Phyllanthus emblic* L. seed. Polar solvents are capable of hydrogen bonding, which breaks weaker bonds between plant matrix and polyphenols, thereby enhancing the release of more polyphenols from the seed matrix components.

Moreover, cold-pressed extraction of polyphenols from *Bischofia polycarpa* seed oil was competitive with most solvents, except isopropanol and acetone. According to this study, *Bischofia polycarpa* seed oil can be obtained with moderate levels of polyphenols via cold pressing. On the other hand, although the polyphenol level in this study was lower than that of Perilla seed oil in China, it was significantly higher than that of common edible vegetable oils such as flaxseed oil, corn oil, rice bran oil, and palm oil (Li et al., 2018). Thus, the interest in extracting polyphenol from *Bischofia polycarpa* seed oil to meet future needs for nutrition labeling is reasonable.

### 3.8. Antioxidant activity

Antioxidant activity is an important and necessary criterion to evaluate the quality of vegetable oils. As presented in Table 2b, four assays, such as DPPH, FRAP, ABTS, and ORAC, were employed in this survey to measure the antioxidant activity of Bischofia polycarpa seed oil. The polar extracts achieved with isopropanol exhibited the strongest DPPH scavenging capacity, followed by acetone, but the DPPH values of *n*-hexane were the lowest (P < 0.05). For FRAP values, all oil samples were varied in the range of 580.70–770.75  $\mu mol~TE/100$  g. In addition, in line with FRAP and DPPH, isopropanol extract resulted in the highest ABTS value. The trends were consistent despite the different 3 antioxidant activity methods' absolute values. ORAC is another assay widely adopted to evaluate the antioxidant activity of vegetable oil. The ORAC values were in the following order of petroleum ether < n-hexane <ethyl acetate < Hx: Iso < acetone < Folch < isopropanol. This observation differed from the other three methods (ABTS, FRAP, and DPPH), possibly attributed to the different mechanisms used by these assays. The principles of ABTS, FRAP, and DPPH are based upon single-electron transfer non-reactive oxygen. In contrast, the ORAC assay is based upon a classical hydrogen atom transfer, thus leading to the difference in ORAC results (Gao et al., 2019). Overall, Bischofia polycarpa seed oil obtained by isopropanol manifested stronger antioxidant activity than other solvents, consistent with the previous study (Tir et al., 2012). Moreover, Table 2b shows the DPPH scavenging capacity of the whole oil and non-polar extracts. Bischofia polycarpa seed oil obtained using Folch and isopropanol solvent had the highest DPPH (nonpolar extracts) and DPPH (whole oil), respectively. On the contrary, n-hexane extracted oil presented a lower DPPH value for whole oil and non-polar extracts. In conclusion, both strong polar solvents and mixed solvents are beneficial in enhancing the antioxidant activity of Bischofia polycarpa seed oils.

Additionally, oxidation stability induction (OSI) time was used to examine the oxidation stability of seed oil obtained from *Bischofia polycarpa*. As shown in Table 2b, the OSI values of the oil samples were significantly different (5.20 to 8.31 h, P < 0.05). It is clear from this study that isopropanol-obtained oil exhibited the highest OSI value, indicating that the oxidation occurred weakly during storage. While the OSI of petroleum-ether extracted oil was the shortest, that of cold pressed oil was the medium. Overall, the free radical scavenging ability was consistent with OSI, and both of them reflected the antioxidant ability of *Bischofia polycarpa* seed oil.

### 3.9. Correlations between phytochemicals and antioxidant activity

In vegetable oils, active phytochemicals are responsible for their

### Table 2b

Effect of extraction solvent on oxidative-stability index (h) and antioxidant activity (µmol TE/100 g) of Bischofia polycarpa seed oils.

	n-Hexane	Petroleum ether	Ethyl acetate	Hx: Iso	Folch	Acetone	Isopropanol	Cold pressing
Oxidation stability (h) Antioxidant activity (umoL TE/100 g)	$5.65\pm0.05^e$	$5.20\pm0.11^{\rm f}$	$5.80\pm0.12^{e}$	$6.15\pm0.15^{de}$	$7.53\pm0.10^{\text{c}}$	$8.12\pm0.05^{b}$	$8.31\pm0.6^a$	$6.37\pm0.10^{d}$
Polar extract								
DPPH	$\textbf{425.19} \pm \textbf{5.09}^{f}$	$\begin{array}{c} 439.02 \ \pm \\ 10.23^{\rm f} \end{array}$	$475.35 \pm 3.69^{e}$	$500.88\pm2.02^d$	$540.42\pm3.80^c$	$608.39\pm7.22^b$	$664.37\pm4.68^a$	$509.75 \pm 8.02^{d}$
FRAP	$586.64 \pm 5.31^{\rm f}$	$580.70 \pm 9.20^{\rm f}$	$632.28\pm8.21^{e}$	$709.93 \pm 0.90^{d}$	$736.40\pm2.22^{c}$	$750.31 \pm 2.68^{b}$	$770.75 \pm 3.90^{a}$	$\underset{cd}{716.05}\pm10.60$
ABTS ORAC	$\begin{array}{l} 528.65 \pm 3.00^{\rm f} \\ 1022.42 \pm \\ 19.00^{\rm e} \end{array}$	$\begin{array}{l} 516.21 \pm 5.10^{\rm f} \\ 955.18 \pm \\ 12.03^{\rm f} \end{array}$	$\begin{array}{l} 543.52 \pm 2.89^{\rm e} \\ 1054.56 \pm \\ 26.53^{\rm e} \end{array}$	$\begin{array}{c} 617.83 \pm 7.02^{d} \\ 1128.41 \pm \\ 32.04^{d} \end{array}$	$\begin{array}{c} 635.50 \pm 2.80^{c} \\ 1224.63 \pm \\ 22.00^{c} \end{array}$	$\begin{array}{l} 690.44 \pm 5.30^{b} \\ 1207.39 \pm \\ 16.30^{b} \end{array}$	$\begin{array}{l} 720.05 \pm 4.05^a \\ 1270.23 \pm \\ 12.56^a \end{array}$	$\begin{array}{l} 602.51 \pm 4.31^{d} \\ 1105.74 \pm \\ 20.13^{d} \end{array}$
Nonpolar extract								
DPPH	$204.05\pm2.03^e$	$262.38 \pm 2.59^{ m b}$	$230.64\pm0.95^c$	$265.27 \pm 3.03^{b}$	$295.40\pm2.80^a$	$210.01\pm1.86^d$	$205.32\pm4.30^{de}$	$220.25\pm1.60^{c}$
Whole oil								
DPPH	$658.90\pm8.84^e$	$665.56 \pm 6.80^{ m de}$	$712.18\pm8.53^{c}$	$\begin{array}{c} 750.54 \ \pm \\ 10.00^c \end{array}$	$\begin{array}{l} 838.90 \pm \\ 8.75^{ab} \end{array}$	$\begin{array}{c} 820.35 \pm \\ 12.80^{b} \end{array}$	$856.10 \pm 10.05^{a}$	$670.32 \pm 7.20^{d}$

All data represents the mean of three replications (Mean  $\pm$  SD).

Values in the list with different letters are significant difference at P < 0.05.

antioxidant properties. As presented in Fig. 3 and Table 2S, bivariate correlations analysis was conducted to reveal the correlation between the antioxidant activity of Bischofia polycarpa seed oil and its phytochemicals. The OSI was highly positively correlated with polyphenol concentration (r = 0.880, P < 0.01), suggesting that there might be a significant dose-effect relationship between polyphenol contents and antioxidant activity. Likewise, free radical scavenging ability (ABTS, DPPH) or FRAP were positively correlated with polyphenols (r = 0.913~ 0.977, P < 0.01), while ORAC was slightly less correlated with polyphenols (r = 0.811, P < 0.05). This difference attributed to the fact that ORAC may be affected by other coexisting antioxidant components. Several studies have reported the superior antioxidant activity of polyphenols (Gao et al., 2019; Shi et al., 2018), and our study confirmed the important role of polyphenols in Bischofia polycarpa seed oil. In addition,  $\gamma$ -tocopherol (r = 0.760, P < 0.05) and  $\beta$ -sitosterol (r = 0.718, P < 0.05) were associated with non-polar DPPH extracts. β-Sitosterol is thought to be an antioxidant that scavenges free radicals via activating antioxidant enzymes. From the above results, we concluded that  $\beta$ -sitosterol and  $\gamma$ -tocopherol could be the major antioxidants in the non-polar extracts of Bischofia polycarpa seed oil. On the other hand, negative correlations



**Fig. 3.** Correlations between phytochemicals, oxidative stability, and antioxidant activity of studied samples; \*\*. Correlation is significant at the 0.01 level (2-tailed); \*. Correlation is significant at the 0.05 level (2-tailed); DPPH. wo (DPPH, whole oil); DPPH. ne (DPPH, nonpolar extract).

between  $\alpha$ -tocopherol,  $\beta$ -tocopherol and FRAP were also observed in this study (r = -0.750  $\sim$  -0.711, P < 0.05).

### 3.10. Multiple linear regression

A stepwise method was accomplished for MLR analysis among antioxidant activity and minor phytochemicals of Bischofia polycarpa seed oil (Table 3). Bischofia polycarpa seed oil had high regression coefficients (R  $= 0.913 \sim 2.070)$  for all antioxidant indexes (except DPPH non-polar extract), indicating that that polyphenols were the dominant antioxidant component. The model constructed by the DPPH polar extract reached the maximum adjusted  $R^2$  (0.970), suggesting that polyphenol levels in Bischofia polycarpa seed oil can be used to predict the DPPH scavenging ability of polar extract accurately. Two variables appeared in the prediction equation for DPPH non-polar extract, with partial correlation coefficients of 0.803(y-tocopherol), and 0.253 (β-sitosterol), respectively. In addition, the ORAC model was mainly associated with polyphenols,  $\alpha$ -tocopherol, and  $\beta$ -Sitosterol, and the predictive equation was  $Y = -4.889 \times 10^{-7} + 1.462$  (polyphenols) + 0.807  $(\alpha$ -tocopherol) + 0.264( $\beta$ -sitosterol). Consistent with the results of correlation analysis, the ORAC model differs from ABTS, FRAP, and DPPH models, requiring multiple factors for prediction. Overall, the polyphenols of Bischofia polycarpa seed oil played the most important role in predicting its antioxidant activity.

### 4. Conclusions

The current investigation revealed that the Bischofia polycarpa seed oil obtained from different solvents (isopropanol, acetone, ethyl acetate, petroleum ether, n-hexane, Folch, and Hx: Iso) presented diversified lipid yield, chemical composition, and antioxidant activity. High lipid yields were obtained by Hx: Iso and Folch, while those of strongly polar solvents such as isopropanol and acetone were characterized by lower lipid yields. The C18:3 content of *Bischofia polycarpa* oil obtained by Folch was significantly higher than that of other methods, but the C18:2 was lower (P < 0.05). Further, Folch solvents presented the best extraction efficiency of total tocopherols, and the extraction contents of petroleum ether for squalene and phytosterol were the highest. Although phytosterol and squalene extracted with acetone and isopropanol were lower than other solvents, they had significant advantages in extracting polyphenols. Furthermore, it was also found that the oil samples extracted by acetone and isopropanol had higher oxidation stability and antioxidant activity. The correlation analysis showed that

### Table 3

		1				1	· · · ·		• .				•
Lanotion	TTOMIONIO	and sooso	non anott	101001 100	tho	nrodiation	a + +	ho optionidopt	000001111	brr mo11	Itimio	110000 000	2200001020
FULLATION	varianie	AUG LEVIES	STOLL COPE	1012111 111					Canacity	11V 1111	1111111	IIIIPAL IPO	PRESENTIT
Luuuuuu	variable.	und reste		ICICIIC III	i uic	DICUICIOI	<b>UI</b> U	ne uniteritation	Cubucity	<i>D</i> <b>1 1 1 1 1 1 1 1 1 1</b>	TUDIC	mou io	
1	,					F				J -	· · ·		<b>,</b>

Antioxidant assay	(R <sup>2</sup> )	(R <sup>2</sup> Adjusted)	Variable	R	Standard error	t	Significance (two-trails p)	Equation
Polar extract								
DPPH	0.983	0.970	(Constant)	-1.141E- 06	0.156	0.000	1.000	$Y = -1.141*10^{-6} + 0.913$ (polyphenols)
			Polyphenols	0.913	0.167	5.476	0.002	
FRAP	0.955	0.947	(Constant)	-1.221E- 06	0.081	0.000	1.000	$Y = -1.221*10^{-6} + 0.977$ (polyphenols)
			Polyphenols	0.977	0.087	11.253	0.000	
ABTS	0.864	0.841	(Constant)	8.815E-08	0.141	0.000	1.000	$Y = 8.815 * 10^{-8} + 0.929$ (polyphenols)
			Polyphenols	0.929	0.151	6.172	0.001	
ORAC	0.939	0.915	(Constant)	-4.889E- 07	0.063	0.000	1.000	$Y = -4.889 \times 10^{-7} + 1.462$ (polyphenols) + 0.807 (α-tocopherol) + 0.264 (β-sitosterol)
			Polyphenols	1.462	0.127	11.465	0.001	
			α-Tocopherol	0.807	0.141	5.731	0.005	
			β-Sitosterol	0.264	0.087	3.041	0.038	
DPPH nonpolar extract	0.881	0.834	(Constant)	6.911E-07	0.144	0.000	1.000	$Y = 6.911*10^{-7} + 0.803 \text{ (}\gamma\text{-tocopherol)} + 0.253 \text{ (}\beta\text{-sitosterol)}$
			γ-Tocopherol	0.803	0.155	5.193	0.003	
			β-Sitosterol	0.253	0.085	3.574	0.016	
DPPH whole oil	0.995	0.989	(Constant)	-7.173E- 07	0.037	0.000	1.000	$\begin{split} Y &= -7.173*10^{-7} + 2.070 \text{ (polyphenol)} + 1.037 \text{ (squalene)} \\ &+ 0.496 \text{ (}\alpha\text{-tocopherol)} - 0.255 \text{ (}\gamma\text{-tocopherol)} \end{split}$
			Polyphenols	2.070	0.097	21.244	0.000	
			Squalene	1.037	0.106	9.787	0.002	
			α-Tocopherol	0.496	0.083	5.990	0.009	
			γ-Tocopherol	-0.255	0.069	-3.717	0.034	

polyphenols were the main variable affecting *Bischofia polycarpa* seed oil's antioxidant activity. MLR models were constructed to predict the results of antioxidant activity (ORAC, FRAP, ABTS, and DPPH). To summarize, this comparative study showed that different extraction methods affected the overall quality of *Bischofia polycarpa* seed oil. *Bischofia polycarpa* seed oil can be used as alternative vegetable oil and an important source of lipid nutrients.

### CRediT authorship contribution statement

Yongjin Wang: Formal analysis, Methodology, Visualization, Writing – original draft. Yijia Su: Formal analysis, Software, Writing – review & editing. Qayyum Shehzad: Investigation, Writing – review & editing. Le Yu: Conceptualization, Data curation. Ailing Tian: Resources, Software. Shihao Wang: Visualization, Validation. Lukai Ma: Methodology, Visualization. Lili Zheng: Investigation, Validation. Lirong Xu: Project administration, Conceptualization, Writing – review & editing, Supervision.

### **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

Data will be made available on request.

### Acknowledgements

This publication was supported by the National Natural Science Foundation of China (32202083).

### Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.fochx.2023.100588.

### References

- Abdelghany, A. M., Zhang, S. R., Azam, M., Shaibu, A. S., Feng, Y., Li, Y. F., ... Sun, J. M. (2020). Profiling of seed fatty acid composition in 1025 Chinese soybean accessions from diverse ecoregions. *Crop Journal*, 8(4), 635–644.
- AOAC. (2000). AOAC Official methods of analysis of AOAC international (19th Ed.). Washington, DC: Association of Official Analytical Chemists.
- Bhatnagar, A. S., & Krishna, A. G. G. (2013). Effect of extraction solvent on oil and bioactives composition of commercial Indian niger (Guizotia abyssinica (L.f.) Cass.) seed. Journal of the American Oil Chemists Society, 90(8), 1203–1212.
- Berezin, O. Y., Tur'yan, Y. I., Kuselman, I., & Shenhar, A. (1996). Rapid and complete extraction of free fatty acids from oilseeds for acid value determination. *Journal of* the American Oil Chemists' Society, 73(12), 1707–1711.
- Chang, M., Wang, Z. T., Zhang, T., Wang, T., Liu, R. J., Wang, Y., ... Wang, X. G. (2020). Characterization of fatty acids, triacylglycerols, phytosterols and tocopherols in peony seed oil from five different major areas in China. *Food Research International*, 137, Article 109416.
- Chew, S. C. (2020). Cold-pressed rapeseed (Brassica napus) oil: Chemistry and functionality. *Food Research International*, 131, Article 108997.
- Dabrowski, G., Czaplicki, S., & Konopka, I. (2020). Composition and quality of poppy (Papaver somniferum L.) seed oil depending on the extraction method. *LWT-Food Science and Technology*, 134(2), Article 110167.
- Gao, P., Liu, R. J., Jin, Q. Z., & Wang, X. G. (2019). Comparison of solvents for extraction of walnut oils: Lipid yield, lipid compositions, minor-component content, and antioxidant capacity. *LWT-Food Science and Technology*, 110, 346–352.
- Huang, L. M., Li, J., Bi, Y. L., Xu, Y. H., Wang, Y. M., Wang, J., & Peng, D. (2021). Simultaneous determination of α-tocopherol, β-tocopherol, γ-tocopherol, δ-tocopherol, sesamin, sesamolin, sesamol, and asarinin in sesame oil by normalphase high performance liquid chromatography. *Journal of Food Composition and Analysis*, 104, Article 104132.
- Lee, M. H., Kim, S. S., Cho, C. W., Choi, S. Y., In, G., & Kim, K. T. (2013). Quality and characteristics of ginseng seed oil treated using different extraction methods. *Journal* of Ginseng Research, 37(4), 468–474.
- Li, H., Yin, X. Z., Jing, J. Z., Wang, M. L., Shen, R. L., & Li, N. (2019). Physicochemical properties and fatty acid composition of Bischofia polycarpa seed oil. *China oils and fats*, 44(6), 99–101.
- Li, X. J., Shen, Y. B., Wu, G. C., Qi, X. G., Zhang, H., Wang, L., & Qian, H. F. (2018). Determination of key active components in different edible oils affecting lipid accumulation and reactive oxygen species production in HepG2 cells. *Journal of Agricultural and Food Chemistry*, 66(45), 11943–11956.
- Mendonca, C. R., Bica, C. I., Piatnicki, C. M., Simo-Alfonso, E. F., & Ramis-Ramos, G. (2005). Characterization of hydroxyaromatic compounds in vegetable oils by capillary electrophoresis with direct injection in an oil-miscible KOH/propanol/ methanol medium. *Electrophoresis*, 26(17), 3307–3314.
- Piravi-vanak, Z., Azadmard-Damirchi, S., Kahrizi, D., Mooraki, N., Ercisli, S., Savage, G. P., ... Martinez, F. (2022). Physicochemical properties of oil extracted from camelina (Camelina sativa) seeds as a new source of vegetable oil in different regions of Iran. *Journal of Molecular Liquids*, 345, Article 117043.

### Y. Wang et al.

- Russin, T. A., Boye, J. I., Arcand, Y., & Rajamohamed, S. H. (2010). Alternative techniques for defatting soy: A practical review. *Food Bioprocess Technology*, 4, 2200–2223.
- Shi, L. K., Mao, J. H., Zheng, L., Zhao, C. W., Jin, Q. Z., & Wang, X. G. (2018). Chemical characterization and free radical scavenging capacity of oils obtained from Torreya grandis Fort. ex. Lindl. and Torreya grandis Fort. var. Merrillii: A comparative study using chemometrics. *Industrial Crops and Products*, 115, 250–260.
- Tir, R., Dutta, P. C., & Ahmed, Y. B. H. A. (2012). Effect of the extraction solvent polarity on the sesame seeds oil composition. *European Journal of Lipid Science and Technology*, 114(12), 1427–1438.
- Uoonlue, N., & Muangrat, R. (2018). Effect of different solvents on subcritical solvent extraction of oil from Assam tea seeds (Camellia sinensis var. assamica): Optimization of oil extraction and physicochemical analysis. *Journal of Food Process Engineering*, 42(2), Article 12960.
- Wang, C. Z., Xu, L., Wu, Q., Zhou, Z. K., Ren, X. C., & Yang, R. (2015). The importance of ultrahigh pressure processing over the quality of the extracted oil from peony seeds (Paeonia suffruticosa Andr.). *Industrial Crops and Products*, *76*, 1142–1147.
- Wang, Y., Yu, L., Zhao, A., Karrar, E., Zhang, H., Jin, W., ... X. (2021). Quality characteristics and antioxidant activity during fruit ripening of three monovarietal olive oils cultivated in China. *Journal of the American Oil Chemists' Society*, Article 12449.
- Xie, D., Jin, J., Sun, J., Liang, L., Wang, X., Zhang, W., ... Jin, Q. (2017). Comparison of solvents for extraction of krill oil from krill meal: Lipid yield, phospholipids content, fatty acids composition and minor components. *Food Chemistry*, 233, 434–441.
- Xu, L., Zhu, C., Liu, T., Karrar, E., Ouyang, Y., & Li, D. (2022). Effect of microwave heating on lipid composition, chemical properties and antioxidant activity of oils from Trichosanthes kirilowii seed. *Food Research International*, 159, Article 111643.
- Xu, L., Liu, T. R., Cao, H. Q., Zheng, L. L., Zhu, C. F., Karrar, E., ... Shen, X. C. (2022). Influence of different extraction methods on the chemical composition, antioxidant

- activity, and overall quality attributes of oils from Trichosanthes kirilowii Maxim seed. Food Control, 142, Article 109201.
- Xu, L., Wang, S., Tian, A., Liu, T., Benjakul, S., Xiao, G., ... Ma, L. (2022). Characteristic volatile compounds, fatty acids and minor bioactive components in oils from green plum seed by HS-GC-IMS, GC-MS and HPLC. Food Chemistry, X, Article 100530.
- Yang, J., Wen, C. T., Duan, Y. Q., Deng, Q. C., Peng, D. F., Zhang, H. H., & Ma, H. L. (2021). The composition, extraction, analysis, bioactivities, bioavailability and applications in food system of flaxseed (Linum usitatissimum L.) oil: A review. *Trends* in Food Science & Technology, 118, 252–260.
- Yang, R. N., Zhang, L. X., Li, P. W., Yu, L., Mao, J., Wang, X. P., & Zhang, Q. (2018). A review of chemical composition and nutritional properties of minor vegetable oils in China. Trends in Food Science & Technology, 74, 26–32.
- Yue, H., Qiu, B., Jia, M., Liu, W., Guo, X. F., Li, N., ... Li, D. (2021). Effects of alphalinolenic acid intake on blood lipid profiles systematic review and meta-analysis of randomized controlled trials. *Critical Reviews in Food Science and Nutrition*, 61(17), 2894–2910.
- Zeng, J. P., Xiao, T., Ni, X. G., Wei, T., Liu, X. R., Deng, Z. Y., & Li, J. (2022). The comparative analysis of different oil extraction methods based on the quality of flaxseed oil. *Journal of Food Composition and Analysis*, 107, Article 104373.
- Zhang, B. P., Yan, Z. Y., & Wu, S. L. (2016). Study on the characteristics and cultivation techniques of Bischofia polycarpa. *Contemporary Horticulture*, *3*, 32–34.
- Zhang, W., Li, K., Zheng, H., Liu, L., Ge, S., & Zhang, H. (2017). Quality analysis of Phyllanthus emblic L. seed oil extracted by different solvents. *Science and Technology* of Food Industry, 38(2), 261–265.
- Zhu, Y. K., Zhang, Z. S., Xie, Q. F., & Liu, Y. L. (2016). Effects of solvents on extraction rate and quality of Eucommia ulmoides olive seed oil. *Journal of Henan University of Technology(Natural Science Edition)*, 37(5), 10–14.