LWT - Food Science and Technology 60 (2015) 1226–1234

Contents lists available at ScienceDirect

LWT - Food Science and Technology

journal homepage: www.elsevier.com/locate/lwt

# Supercritical carbon dioxide extraction of oils from two *Torreya grandis* varieties seeds and their physicochemical and antioxidant properties

Qinxue Ni [a], Qianxin Gao [a], Weiwu Yu [b], Xingquan Liu [a], Guangzhi Xu [a], Youzuo Zhang [a, *]

[a] The Key Laboratory for Quality Improvement of Agricultural Products of Zhejiang Province, Zhejiang Agriculture and Forestry University, Lin'an, Zhejiang Province 311300, China

[b] Nurturing Station for the State Key Laboratory of Subtropical Silviculture, Zhejiang Agriculture and Forestry University, Lin'an, Zhejiang Province 311300, China

A R T I C L E I N F O

*Article history:*
Received 2 May 2013
Received in revised form
11 June 2014
Accepted 1 September 2014
Available online 10 September 2014

*Keywords:*
*Torreya grandis* seed oil
Supercritical carbon dioxide extraction
Physicochemical property
Fatty acid
Antioxidant activity

A B S T R A C T

Supercritical carbon dioxide extraction of oil from *Torreya grandis* seeds was optimized using response surface methodology. Its physicochemical properties, fatty acid composition, total phenolic content and antioxidant activities from two varieties namely *T. grandis* var. merrillii and *T. grandis* var. *jiulongshanensis* were investigated. As results, the maximum oil yield was predicted to be 35.43 g/100 g dry seed using an extraction pressure of 24.98 MPa, temperature of 51.39 °C, extraction duration of 4 h and $CO_2$ flow rate of 15.42 L/h. *T. grandis* var. merrillii seed oil has better overall quality. Its acid, peroxide, iodine and saponification value were 0.63 mg KOH/g oil, 3.37 m $EqO_2$/Kg, 124.33 mg/100 g oil, 122.86 mg KOH/g oil, respectively. Large proportions of PUFA (59.99%) were found in the oil with linoleic acid (37.81 g/100 g oil) as the main fatty acid. The oil was also found present high content of phenolic compounds (16.9 mg GAE/g) and antioxidant activity in DPPH radical scavenging assay and β-carotene bleaching test ($IC_{50}$ values of 123.76 ± 0.13 μg/mL and 194.65 ± 0.09 μg/mL, respectively). Our results indicate that *T. grandis* var. merrillii seeds oil is a good candidate for use as nutrient rich edible oil or ingredients for functional foods.



## 1. Introduction

*Torreya grandis*, which belongs to the family Taxaceae and category *Torreya*, is a large, evergreen and ornamental coniferous tree of great economic value that mainly grows in China and Japan. The seed is one of the rarest dry fruits in the world with their high nutritional and medicinal value. In China, it was initially used as a Traditional Medicine for more than 1500 years for the purposes of expelling parasites, preventing malnutrition, moistening lung tissues, loosing bowel, preventing phlegm and stopping cough.

Recently, much attention were attracted on the study of the potential nutritional and health benefits of *T. grandis* seed and other organs (Endo, Osada, Kimura, & Fujimoto, 2006; Saeed et al., 2007, 2010). The seed has multiple biological properties, including anti-inflammatory, antiviral, anti-atherosclerosis, antihelmintic, antitussive, carminative, laxative, antifungal, antibacterial and antitumor activities due to its rich content of B vitamins (*i.e.* nicotinic acid, folic acid), mineral elements and phenolic compounds that may possess anti-oxidant activity (Chen et al., 2006). Therefore, *T. grandis* seed can be considered as an important source for the food and nutraceutical supplement industries due to their functional properties.

*Torreya* seed contained oil 42.6–61.5 g/100 g dry seeds, and its fatty acid composition and content were varied with different species of *Torreya* (Chen, Zheng, Fu, Zhou, & Weng, 1998). There are 6 species, 2 varieties and 11 cultivars of *Torreya* genus around the world, of which *T. grandis* is the superior specie and wildly used in China. *T. grandis* var. merrillii and *T. grandis* var. *jiulongshanensis* are its two varieties.

However, to the best of our knowledge, no data on the extraction of *T. grandis* seed oil have been reported and the seed oil and its physicochemical properties and potential functions were also rarely reported.

Therefore, in the present study, we investigated supercritical carbon dioxide extraction (SC-$CO_2$ extraction) of oil from *T. grandis* seeds. Response Surface Methodology (RSM) was used to optimize the main operating parameters, namely, pressure, temperature, extraction time, and $CO_2$ flow rate. The physicochemical properties and fatty acid composition of seed oil from two varieties, namely, *T.*

* Corresponding author. Tel./fax: +86 571 63741689.
*E-mail address:* yyzzhang2002@hotmail.com (Y. Zhang).

http://dx.doi.org/10.1016/j.lwt.2014.09.007

*grandis* var. *jiulongshanensis* and *T. grandis* var. merrillii, were analyzed and compared using GC–MS. Furthermore, the total phenolic contents and antioxidant activity of both seed oils under optimal conditions was determined by means of Folin–Ciocalteu method and the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical-scavenging assay and the β-carotene bleaching test. We suppose the study would provide practical guidance to further exploiting and application of the natural seed oil resource and useful information for food industry.

## 2. Method

### 2.1. Materials

Seeds of *T. grandis* var. merrillii and *T. grandis* var. *jiulongshanensis* were collected during autumn 2011 from Shaoxing, Zhejiang province, China. The strains were identified by Prof. Luhuan Lou, School of Forestry and Biotechnology, Zhejiang Agriculture and Forestry University, Lin'an, China. Hulled seeds were dried at 60 °C for 24 h in an oven. The moisture content of the seeds (8.83 g/100 g dry seeds) was determined with an infrared moisture analyzer (OHAUS, Pine, USA). Seeds were then ground in a blender (Yili Industry & Trade CO. Ltd., Wuyi, China). All solvents used were of analytical grade.

### 2.2. Supercritical carbon dioxide extraction (SC-$CO_2$ extraction)

SC-$CO_2$ extraction of seed oil was performed on an HA221-50-06 SFE device (Hua'an Supercritical Fluid Extraction Corp., Nan-tong, China). Samples (100 g) of *T. grandis* var. merrillii seeds were placed into the extraction vessel. The amount of extracted oil was determined gravimetrically after collection, and the extraction yield is expressed as the percent ratio of the mass of extracted oil to the mass of *T. grandis* seeds loaded in the extraction vessel, as follows:

$$\text{Yield of seed oil } (\text{g}/100\text{ g dry seed}) = (\text{mass of extracted oil}/\text{mass of dried seed}) \times 100 \quad (1)$$

### 2.3. Experimental design

A four-variable, three-level Box–Behnken design (BBD) was employed in this optimization study based on the results of preliminary experiments. Pressure (MPa, $X_1$), temperature (°C, $X_2$), extraction time (h, $X_3$) and $CO_2$ flow rate (L/h, $X_4$) were the independent variables selected to optimize the extraction of *T. grandis* seed oil. Extraction yield ($Y$) was taken as the response in the design experiments. Based on the single-factor experimental analysis of setting pressure 9, 12, 15, 18, 21, 24, 27, 30, 33 Mpa, temperature 30, 40, 50, 60, 70 °C, extraction duration of 1, 2, 3, 4, 5, 6, 7 h and $CO_2$ flow rate 10, 13, 16, 19, 22 L/h, respectively, each variable was set at three levels in the BBD (Table 1). Twenty-nine experiments were repeated three times and carried out at the center points to evaluate the pure error.

A second-order polynomial regression model was used to express the yield as a function of the independent variables as follows:

$$Y = \beta_0 + \sum_{i=1}^{4} \beta_i X_i + \sum_{i=1}^{4} \beta_{ii} X_i^2 + \sum_{i=1}^{3} \sum_{j=i+1}^{4} \beta_{ij} X_i X_j \quad (2)$$

where $Y$ presents the response variables, $\beta_0$ is a constant and $\beta_i$, $\beta_{ii}$ and $\beta_{ij}$ are the linear, quadratic and interactive coefficients, respectively. $X_i$ and $X_j$ are the levels of the independent variables.

**Table 1**
Box–Behnken design for optimization of the SC-$CO_2$ extraction of *Torreya grandis* seed oil and values of observed responses.

| Run | Independent variable | | | | Response (seed oil extraction yield (g/100 g seeds)) | |
|---|---|---|---|---|---|---|
| | $X_1$ Pressure (MPa) | $X_2$ Temperature (°C) | $X_3$ Time (h) | $X_4$ $CO_2$ flow rate (L/h) | Experimental | Predicted |
| 1 | −1 (21) | −1 (40) | 0 (3) | 0 (16) | 30.43 | 30.40 |
| 2 | 1 (27) | −1 (40) | 0 (3) | 0 (16) | 32.88 | 33.14 |
| 3 | −1 (21) | 1 (60) | 0 (3) | 0 (16) | 32.31 | 32.14 |
| 4 | 1 (27) | 1 (60) | 0 (3) | 0 (16) | 32.79 | 32.90 |
| 5 | 0 (24) | 0 (50) | −1 (2) | −1 (13) | 32.07 | 32.48 |
| 6 | 0 (24) | 0 (50) | 1 (4) | −1 (13) | 34.83 | 34.62 |
| 7 | 0 (24) | 0 (50) | −1 (2) | 1 (19) | 34.43 | 34.72 |
| 8 | 0 (24) | 0 (50) | 1 (4) | 1 (19) | 33.94 | 33.62 |
| 9 | −1 (21) | 0 (50) | 0 (3) | −1 (13) | 31.19 | 31.48 |
| 10 | 1 (27) | 0 (50) | 0 (3) | −1 (13) | 33.4 | 33.35 |
| 11 | −1 (21) | 0 (50) | 0 (3) | 1 (19) | 32.03 | 32.22 |
| 12 | 1 (27) | 0 (50) | 0 (3) | 1 (19) | 34.01 | 33.86 |
| 13 | 0 (24) | −1 (40) | −1 (2) | 0 (16) | 32.98 | 33.04 |
| 14 | 0 (24) | 1 (60) | −1 (2) | 0 (16) | 33.2 | 32.99 |
| 15 | 0 (24) | −1 (40) | 1 (4) | 0 (16) | 32.41 | 32.76 |
| 16 | 0 (24) | 1 (60) | 1 (4) | 0 (16) | 34.23 | 34.31 |
| 17 | −1 (21) | 0 (50) | −1 (2) | 0 (16) | 32.78 | 32.48 |
| 18 | 1 (27) | 0 (50) | −1 (2) | 0 (16) | 34.32 | 34.08 |
| 19 | −1 (21) | 0 (50) | 1 (4) | 0 (16) | 32.83 | 32.85 |
| 20 | 1 (27) | 0 (50) | 1 (4) | 0 (16) | 34.68 | 34.76 |
| 21 | 0 (24) | −1 (40) | 0 (3) | −1 (13) | 33.03 | 32.60 |
| 22 | 0 (24) | 1 (60) | 0 (3) | −1 (13) | 31.72 | 31.71 |
| 23 | 0 (24) | −1 (40) | 0 (3) | 1 (19) | 31.79 | 31.58 |
| 24 | 0 (24) | 1 (60) | 0 (3) | 1 (19) | 33.76 | 33.97 |
| 25 | 0 (24) | 0 (50) | 0 (3) | 0 (16) | 34.96 | 35.14 |
| 26 | 0 (24) | 0 (50) | 0 (3) | 0 (16) | 35.02 | 35.14 |
| 27 | 0 (24) | 0 (50) | 0 (3) | 0 (16) | 35.33 | 35.14 |
| 28 | 0 (24) | 0 (50) | 0 (3) | 0 (16) | 35.31 | 35.14 |
| 29 | 0 (24) | 0 (50) | 0 (3) | 0 (16) | 35.07 | 35.14 |

### 2.4. Soxhlet extraction

A conventional method of Soxhlet extraction (SE) was performed to compare the extraction performances with SC-$CO_2$ extraction. The powder of *T. grandis* var. merrillii seeds (50 g) was continuously extracted with a Soxhlet extractor for 10 h using *n*-hexane as the solvent (80 °C). After extraction, *n*-hexane was evaporated at 40 °C by a rotary vacuum evaporator and subsequently, the extracts were treated with nitrogen gas to remove residual solvent. The oils obtained were weighed and the yields were calculated.

### 2.5. Physicochemical properties of the seeds and seed oil from *T. grandis*

Total protein was determined by the Kjeldahl method. Protein was calculated using the general factor (6.25). The data are expressed as the percentage of dry weight. Moisture was determined according to the AOAC Official Method 930.15 (AOAC, 1990). The results are expressed as percentages. Oil was extracted from seeds using petroleum ether. The weight of the oil extracted from 10 g of the seed powder was determined to calculate the oil percentage relative to the seed powder dry matter.

Standards ISO (International Organization for standardization) were used for the determination of the peroxide value (ISO 3960, 2001), acid value (ISO 660, 1996), iodine value (ISO 3961, 1996) and saponification value (ISO 3657, 2002) of oil.