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Research Article

Effect of some Cultivation Factors and Extraction Methods on *Terminalia Catappa* L. Seed Oil

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Terminalia catappa L. is a common tropic tree for shade and ornament in many countries. Recently, *Terminalia catappa* L. seed oil has been considered as a new oleaginous seed for dietary and biofuel production. In this study, ripe *Terminalia catappa* L. fruits originated in Vietnam were collected and seed oil extracted. In our experiment conditions, the effect of tree location, tree age, and annual harvest time on seed weight and seed oil content was investigated. As results, the seeds at the eastern site of the ground obtained not only the biggest size (3.607 g) but also contained the highest oil mass percentage (56.38%). The suitable annual harvesting time for the good seed quality was from March to April. In addition, the *Terminalia catappa* L. seeds for oil extraction began being harvested in the fourth year with 55.88% oil content compared to 55.99% of the five and six-year-old and trees. After seed drying, the seed oil was extracted by five different methods including cold screw pressing, hot screw pressing, hydraulic pressing, and solvent extracting, and combining method (cold screw pressing then solvent extraction of oil cake). Among the physical methods, cold screw pressing observed the highest oil yield of 77.32%, and the good oil quality was obtained with low free fatty acid (0.550% oleic acid), low acid value (1.080 mg KOH/g oil), and low peroxide value (1.240 meq O_2/kg oil). However, the combination of cold screw pressing and cake oil extracting by solvent increased the oil yield by 14.61%. The saponification values fluctuated between 196 to 197 mg KOH/g oil, while the iodine values were in the range of 77.00 and 79.89 g I_2/kg oil.

1. Introduction

Terminalia catappa L. (TC) is a common tropic tree planted for shade and ornament in various areas such as Southern Asia, Eastern Southern Asia, Africa, and Southern American. However, the fruit pulp and seeds from this kind of tree are wholesome and the children often eat TC seeds [1, 2]. In recent years, studies on TC tree have been done and the results showed that TC leaves contained high concentration of bioactivity compounds [3, 4]; the TC seeds contained high concentration of protein (over 20.14%) and various essential minerals. Besides, the researchers are interested in TC seed oil extraction. Depending on the origin, the TC seed oil content was in range of 30% to 60%. Therefore, TC seed oil could be a new resource for biofuel producing [5–7] or an edible lipid [8, 9]. The fatty acid profile of TC seed oil was indicated. The main saturated fatty acid is palmitic acid at the concentration over 40% of total fatty acid mass. This oil also contains monounsaturated fatty acid (20-30% oleic acid) and polyunsaturated fatty acid (20–30% linoleic acid). Therefore, it has been stated to be suitable for food production [8–15].

In some previous reports on TC seed oil extraction, some usual pretreatments were done such as drying, roasting, and crushing before hexane extraction using Soxhlet apparatus. The crude oil or refined oil (degummed and bleached) was then determined the oil indices including free fatty acid (%), acid value (mg KOH/g oil), iodine value (g I_2/100 g oil), saponification value (mg KOH/g oil), and fatty acid profile [5, 7, 8, 16–18].

However, extraction using Soxhlet apparatus is a laboratory method and unsuitable for scaling up. Hence, the research on TC seed oil extraction procedure is very essential for oil production in the future. In our country, Vietnam, the TC trees are familiar, especially in the South of the country. In this study, we grew the TC tree in Trang Bang, Tay Ninh, Vietnam for harvesting seeds. The tree age, the tree location site and annual harvest time were investigated to obtain suitable seeds for oil extraction. Then, the TC seed oil was extracted using physical methods (cold screw pressing, hot screw pressing, and hydraulic pressing), chemical method (hexane and ethanol solution as solvent) and combining method (cold screw pressing and then cake solvent extracting). The oil yield and oil quality parameters were evaluated for finding out the suitable extraction method.

2. Materials and Methods

2.1. Collection of Terminalia catappa L. Fruits. In our study, *Terminalia catappa* L. (TC) trees grew in Trang Bang, Tay Ninh Province, Vietnam. Then, TC fruits were harvested to collect seed oil. Some cultivation factors were investigated to find out the good material for oil extraction.

In the first experiment, the seed characteristics from trees at the different site of the growing ground were investigated. In a rectangular ground with 18,000 m^2 area (120 m* 150 m) (Figure 1). TC trees were grown at the four sides of the ground and the distance between trees was 3 m. The two width sides located at the north and the south sites and the length sides located at the east and the west sites. The seed weight and seed oil content of the trees at different location sites were compared. Next, the seed properties of two annual harvest times were evaluated. In addition, for indicating the suitable tree years for harvesting, the seeds of different tree ages were investigated.

2.2. Seed Pretreatments. Some pretreatments were done to obtain the material for oil extracting. The TC seeds were often removed from ripe fruit pulp, dried in oven, and ground before oil extraction [9, 15]. In our experiments, the fresh fruits were cleaned with tap water. Then, they were blanched at 100°C for 10 minutes with the solid–liquid ratio of 1 : 5 and immersed in 3% salt solution for 2 hours to deactivate enzymes and soften the fruit pulp. After that, the pulp was removed by a pulper (CYF–JH, Chin Ying Fa, Taiwan) and the hard shells of the seeds were also cracked to collect the TC seeds. The seed yield was 10.5 ± 0.5% TC fruits weight. Next, the fresh seeds were dried at 60 ± 5°C for 4 hours with an oven (Memmert UE 500 Lab Oven, Switzerland). The moisture content of the dried seeds was about 12% and they were stored in the polyethylene packs at 5 ± 1°C until oil extracting.

In order to test the TC seed properties, the fruits at the eastern site of the growing ground were collected in March when the trees were six years old. The hardness (H) of the seed was determined by using a piston to press the seeds until they were completely broken. The equipment utilized to indicate the seed hardness was EZ Test series C224-E027 (Shimadzu, Japan). The inside structure of the seeds was analyzed by Scanning electron microscope (Hitachi S-4800). Observations were carried out with voltage acceleration of 10 kV.

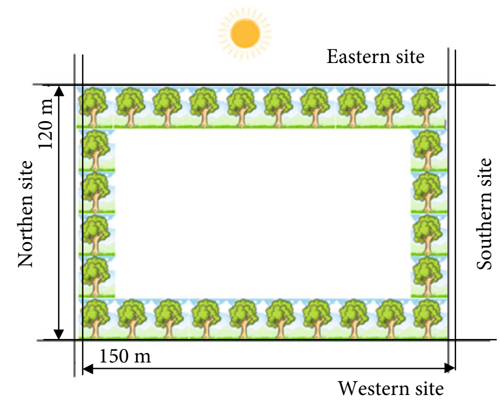

Figure 1: *Terminalia catappa* L. tree location map.

2.3. Seed Oil Extraction Methods

2.3.1. Seed Oil Extraction by Cold Screw Pressing (M1). Cold pressing is one of the common mechanical methods for oil extraction. The temperature of the outlet oil in this method was in ranges of room temperature to 50°C. The previous studies showed that the cold pressing could maintain the higher bioactivity compound concentration than the solvent extraction or the hot pressing in seed oils [19–21]. In our study, the dried seeds (1 kg) were frozen at -18°C for 24 hours before extracting oil with a screw presser (YF–J503, Zhengzhou Yuhuo, China). The outlet temperature of the crude oil was 40.9–41.5°C.

2.3.2. Seed Oil Extraction by Hot Screw Pressing (M2). The dried seeds (1 kg) were pressed for collecting crude oil by a screw presser (YF-J503, Zhengzhou Yuhuo, China). In order to increase the temperature during extraction process, a heater was plugged outside the barrier of the oil presser. The heater temperature was set at 230°C and the oil temperature of the outlet oil was 160 ± 2°C.

2.3.3. Seed Oil Extraction by Hydraulic Presser (M3). In this extraction method, the dried seeds (1 kg) were crushed into powder, the particle size of 1.5 mm, by a blender (RRH2000A, Ririhong, China). The seed powder was heated to 75°C by steam for 45 minutes. The powder with 20 ± 2% (*w*/*w*) moisture content was then pressed by a hydraulic presser (KPB10, Osaka, Japan) to obtain crude oil.

2.3.4. Seed Oil Extraction by Immersing in Organic Solvent (M4). In the chemical extraction of seed oil, the type of solvent is a very important factor, which influence on both oil yield and components in crude oil. Hexane is known as the traditional solvent for this process. In recent years, ethanol has begun to be used as oil extraction solvent in many recent researches [22–25].

In this study, the mixture of n-hexane and ethanol solution in water (70% *v*/*v*) at the ratio of 1 : 2 (*v*/*v*) was used as extraction solvent. In order to increase the surface area, the dried seeds (1 kg) were crushed into powder by a blender (RRH2000A, Ririhong, China). After that, the seed powder was immersed in the solvent for 2 hours. The ratio of solid: solvent was kept constant at 1 : 5 (*w*/*v*). In addition, a continuous stirring was done during extraction period with an agitator at the speed of 800 rpm. This solvent extracting process was done at room temperature.

2.3.5. Seed Oil Extraction by Cold Screw Pressing and Solvent Extraction of Oil Cake (M5). In this process, firstly, the oil seed was extracted by cold screw pressing as in Part 2.2.1 to obtain the oil. Next, the residual oil in the cake was isolated by solvent extraction. The parameters of the solvent extraction were exactly as same as seed powder extraction (Part 2.2.4). The oil from pressing and solvent extracting above were then mixed to obtain the crude oil of this method.

2.4. Oil Cake Removing and Crude Oil Degumming. In order to degum, the TC crude oil obtained by mechanical pressing and the solvent extraction (mixture of crude oil and solvent) were cooled down to 2–4°C for 24 hours in a refrigerator for precipitating phosphatide gum and protein then centrifuged at the speed of 1,016×g and the temperature of 4°C by high speed centrifuge (CNC-106 H1650 Tabletop).

Next, the degummed oil was heated to 60 ± 5°C and removed the remained gum by the vacuum filter (Rocker 300C-VF12, Taiwan) with membrane pores' size of 10–15 μm. After that, the transparent purified oil (mechanical extracting) or degummed filtered micella (solvent extracting) was obtained. Purified oil was stored at 4 ± 1°C for analyzing some parameters including oil extraction yield (%), free fatty acid (% as oleic acid), acid value (mg KOH/g oil), saponification value (mg KOH/g oil), peroxide value (meqO_2/kg oil), and iodine value (g I_2/100 g oil).

In case of solvent extracting, the degummed-filtered micella was evaporated at 75°C by Sohxlet apparatus to remove solvent from oil before keeping at cool temperature (4 ± 1°C).

2.5. Determination of Terminalia catappa L. Seed Oil Properties

2.5.1. Determination of Extraction Oil Yield and Residual Oil Content in the Cake. The extraction oil yield was indicated by the following formula.

$$\mathrm{EOY}(\%) = \frac{\mathrm{We}}{\mathrm{OC} * m} * 100\%. \quad (1)$$

The residual oil content of the cake was indicated by the following formula.

$$\mathrm{ROC}(\%) = \frac{\mathrm{Wro}}{\mathrm{OC} * m} * 100\%, \quad (2)$$

where We (g) is the weight of extracted oil. Wro (g) is the weight of residual oil in the cake which determined by hexane extraction with Soxhlet apparatus for 6 hours. OC (%) is the total oil content in 100 g seeds which determined by hexane extraction with Soxhlet apparatus for 6 hours. *m* (g) is the weight of the extraction seeds (on dry matter).

2.5.2. Determination of Acid Value and Free Fatty Acid. The acid value (AV) and free fatty acid (FFA) of the degummed TC seed oil was analyzed according to ISO 660–2009 [26]. In practice, the oil sample (10 g) in a conical flask was dissolved in 50 mL of solvent mixture including diethyl ether and ethanol (1 : 1 *v*/*v*). A few drops of phenolphtalein solution were added and the mixture was titrated with 0.1 M ethanolic potassium hydroxide until a pink color was observed. The AV and FFA were calculated by the following equations.

$$\mathrm{AV}(\mathrm{mg\,KOH/g}) = \frac{56.1 * c * V}{m}, \qquad \mathrm{FFA}(\%) = \frac{V * c * M * 100}{1000 * m}, \quad (3)$$

where *c* is the exact concentration (*M*) of the standard volumetric potassium hydroxide solution used. *V* is the volume of standard volumetric potassium hydroxide solution used (mL). *m* is the mass of the test portion (g). *M* is the molar mass of the oleic acid (282.47 g/mol).

2.5.3. Determination of Saponification Value. The saponification value (SV) of TC seed oil was indicated based on ISO 3657–1988 [27]. The oil sample (2 g) was weighed into a conical flask; a volume of 25 mL ethanolic potassium hydroxide was added. The mixture was boiled and stirred for 60 minutes. A few drops of phenolphtalein indicator was added and titrated the mixture with 0.5 M HCl until a pink color disappeared. The same procedure was done with the blank sample without oil. And the SV was indicated as following:

$$\mathrm{SV}(\mathrm{mg\,KOH/g}) = \frac{56.1 * (V_o - V_i) * c}{m}, \quad (4)$$

where *c* is the exact concentration (M) of the standard volumetric potassium hydroxide solution used. V_o is the volume of standard volumetric potassium hydroxide solution used for the blank samples (mL). V_i is the volume of standard volumetric potassium hydroxide solution used for the oil samples (mL). *m* is the mass of the test portion (g).

2.5.4. Determination of Iodine Value. The method by ISO 3961–1989 [28] determined iodine value (IV) was used for testing TC seed oil. The oil sample (0.2 g) was dissolved with 20 mL carbon tetrachloride in a conical flask. An exactly 25 mL of the Wijs reagent was added and the stopper was inserted. The flask was placed in the dark for 1 hour. At the end of the time, 20 mL of the potassium iodine solution (100 g/L) and 150 mL of water were poured into the flask. The mixture was then titrated with the standard volumetric sodium thiosulfate solution (0.1 M) until the yellow colour due to iodine has almost disappeared. At that time, a few drops of the starch solution were added, and the titration was continued until the blue colour disappeared. The IV was calculated as follows:

$$\mathrm{IV}(\mathrm{g}\,I/100\mathrm{g\,oil}) = \frac{12.69 * (Vo - Vi) * c}{m}, \quad (5)$$

where c is the exact concentration (M) of the standard volumetric sodium thiosulfate solution used. V_o is the volume of sodium thiosulfate solution used for the blank samples (mL). V_i is the volume of sodium thiosulfate solution used for the oil samples (mL). m is the mass of the test portion (g).

2.5.5. Determination of Peroxide Value. The oil sample (1.0 g) was dissolved in 20 mL of the glacial acetic acid/isooctane solution by gently swirling the flask. Saturated potassium iodide solution (1.0 mL) was added, and the solution was kept in dark for 30 min. Then 100 mL of distilled water was added. After that, the liberated iodine in sample was titrated with 0.01 N sodium thiosulfate standard solution, to a yellow-orange and then light-yellow color, and after adding 0.5 mL of the starch solution. The titration was stopped when the color changed from purple to colorless (within 30 seconds) [29].

Formula for calculating peroxide value (meq O_2/kg)

$$PV = \frac{(V - V_0) * c \times 1000}{m}, \quad (6)$$

where V (mL) is the volume of the 0.01 N sodium thiosulfate standard solution used for the determination; V_0 (mL) is the volume (mL) of the 0.01 N sodium thiosulfate standard solutions used in the blank test. c (0.01 N) is the approximate concentration of sodium thiosulfate standard solution. m (g) is the mass of oil sample.

2.5.6. Determination of Seed Oil Content Using Soxhlet Apparatus. The volume of 300 mL n-hexane was poured into round bottom flask of the Soxhlet apparatus. The mass of 10 g of TC seed powder was covered by filter paper. The Soxhlet was heated. When the solvent was boiled, the vapor rises through the vertical tube into the condenser at the top. The liquid condensate dripped into seed powder. The extract filled the siphon tube, where it flowed back down into the round bottom flask. This was allowed to continue for 6 hours. The seed powder was then removed from the tube, dried in the oven, cooled in the desiccators, and weighed again to determine the amount of extracted oil [30].

2.6. Statistical Analysis. The experiments were performed in triplicate. The analysis of variance method at 95% confidence was used to analyze the data with Stagraphic Centurion XV software.

3. Results and Discussion

3.1. Effect of Tree Locations and Harvest Time on Seed Weight and Seed Oil Content. The seed weight and the seed oil content from the tree in four location sites (east, west, south, and north) were evaluated. As shown in Figure 2, both of seed weight and seed oil content were affected by the tree locations (P value <0.0001). Particularly, the seeds of the trees in the eastern site were the most valuable material for oil extraction with the highest seed weight (3.607 ± 0.086 g) and seed oil content (56.38 ± 0.282%). It is explained that the eastern site could receive more sunlight intensity than others. In many previous studies, the light condition was mentioned as an important factor influenced photosynthesis and accumulating the metabolites in tree growth and crops [31–33]. For example, even in one apple tree, the fruits at the different places on the trees (on top, inside the canopy, east side, or west side) observed the different size, color, and other characteristics due to the change of light condition [33]. The same result was reported by Moon et al. [32] for mandarin, the fruits at the upper of the tree with more intensive light got better quality, high values of fruit weight, pulp weight, and pulp thickness.

TC fruits were harvested in many countries at various period of a year depending on the local climate. In southern India, there are two annual crops: spring (April, May) and fall (October and November) [1]. In Cote d'Ivore, the TC fruits were collected from September to March [12]. In another study at Nigeria, the TC fruit crop was between November and March [8]. In our study, the TC fruits originate in Vietnam and there are two crops: the first is March-April; the second is July–August. The seed weight and seed oil content at various harvest times are presented in Figure 3.

The TC seeds collected at the March–April not only weighed heavier but also contained more oil than the other. The second harvest time (July–August) is the rainy season in Vietnam (from April to October); therefore, the rainy and cloudy weather could affect the TC fruit crop. Marjenah and Putri [31] reported that cloudy weather could cause the decrease of photosynthesis reactions in the tree then lead to the falling down of the young TC fruits. In our result, the appropriate period to collect the TC seeds is from March to April with the seed weight and seed oil content of 3.658 ± 0.089 g and 56.62 ± 0.125%, respectively.

The seed weight and seed oil content of TC obtained in some previous studies were fluctuated in a large range because of the different origins. In a study of TC diversity in Nigeria, the seed weight was about 0.93–3.81 g [34]. The TC seed weight from Indonesia was in range of 5.06–7.33 g [31]. The seed oil content of TC was also showed in the previous research and the value was in range of 30–60% [2, 6, 11, 12]. In general, Vietnamese TC seeds observed medium weight and oil content compared to TC of other countries.

3.2. Effect of the Tree Age on Seed Weight and Seed Oil Content. The TC trees were distributed throughout the tropics and TC fruits are often picked from the third year of planting [15]. In our experiments, the TC seeds from the trees at the fourth, fifth, and sixth year were collected. The location of the tree and the annual harvest time were fixed at the eastern side of the ground and the first harvest time (March–April).

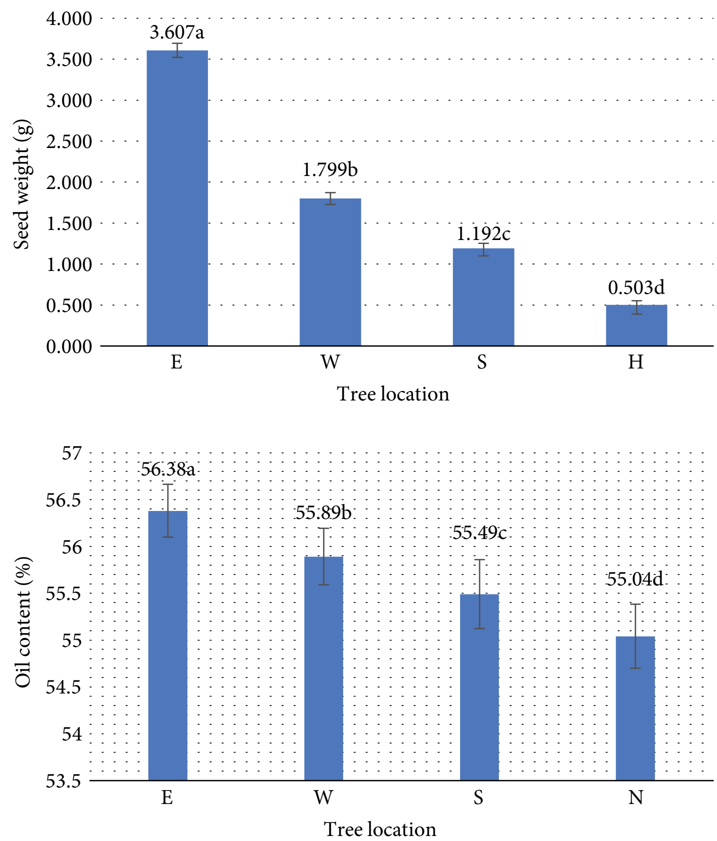

Figure 2: Effect of tree locations on seed weight (g) and seed oil content (% *w*/*w*). Note: *P* value <0.0001. Different letters (a, b, c, …) showed the significant difference at 0.05 level. E, W, S, and N are presented for the eastern, western, southern, and northern sites of the ground on which the *Terminalia catappa* L. were grown.

As in Table 1, the age of the tree significantly affected seed weight and seed oil content. On the fourth year, the seeds were the biggest (3.683 ± 0.086 g), but the seed oil content was the lowest (55.88 ± 0.067%). Particularly, the seed weight of the five-year-old trees decreased about 3.42% compared to the four-year-old trees but equaled to the six-year-old trees. In addition, the oil content increased to 55.99 ± 0.068% on the fifth year and the sixth year. It is meant that the development stages of TC tree could affect the seed weight and seed oil content. This result is agreement with some previous studies about the influence of the tree age on fruit properties (fruit weight, fruit components content, and exocarp thickness). For example, apples of the 2-year-old trees were bigger and contained higher total soluble solid than those of the 3-year-old [35]. However, apples of too young apple trees (<6 ages) were lower resistant ability to bruising and *Pezicula malicorticis* decay than the old trees [36]. In addition, the tree age also influenced the other fruits such as mandarins [37], guavas [38], and pomelos [39].

In general, the TC seeds could be harvested from the fourth year and the seed oil content was over 55%. In comparison to the oil content of other oleaginous seeds such as *Denoix regia* seed: 29% [40]; castor seed: 33.2% [41]; pumpkin seed: 46% [42]; citrus seed 22: 33% [43], or *Thevetia peruviana* seed 45: 64% [44], Vietnamese TC seeds contain quite high oil mass percentage, and it seems to be a new source for oil extraction.

In order to construct the procedure for TC seed oil extraction, the structure of the dried seed must be tested. In our study, the hardness and the inside microstructure of the TC seeds were tested.

Figure 4 showed the compressive force load on TC seed until it was broken. The mean hardness of the seed was 425 N per seed. This parameter helps to calculate the force for crushing or pressing this kind of seed. The difference of the seed hardness depended on the size, the inside structure of the seeds and the seed pretreatment such as drying, steaming, and microwave treating. In many projects on seed oil extracting, the hardness of the seed was also tested.

In a study by Haque et al. [45], the hardness of jatropha, castor, Karanja, and rubber seeds was indicated before oil extraction and the hardness gained the values of 2.7, 1.9, 1.7, and 8.6 kg, respectively. The hardness of perilla seed was determined at 157–1166 gf depended on the cultivar [46]. In another research, the hardness of different genotype pomegranate seeds presented in range of below 150 N to over 400 N [47].

Beside the hardness, inside structure of oleaginous seeds was also tested by scanning electronic microscope in some previous works. The existence of pores and cavities or the dense level inside the seed would influence on the

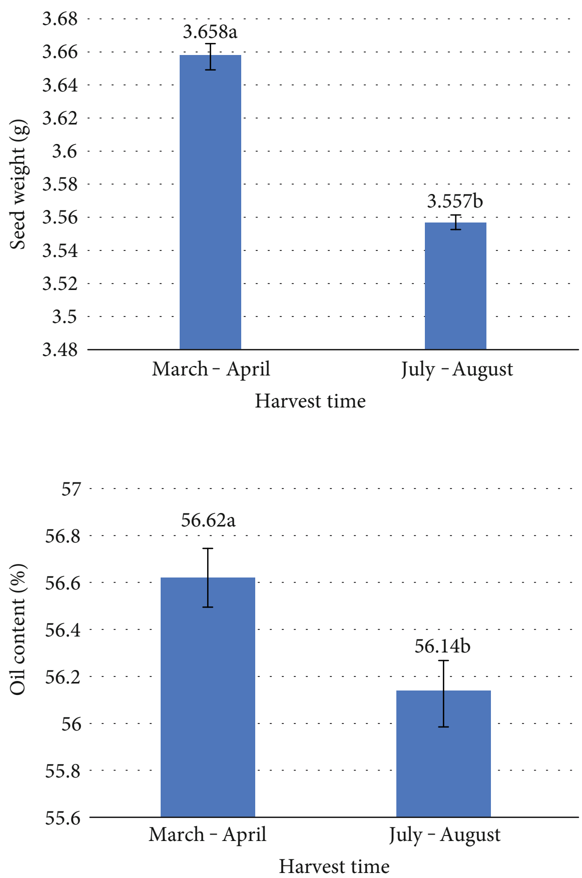

Figure 3: Effect of annual harvest time on seed weight (g) and seed oil content (% w/w). Note: P value of seed weight = 0.0002; P value of seed oil content <0.0001. Different letters (a, b) showed the significant difference at 0.05 level.

Table 1: Effect of the tree age on seed weight (g) and seed oil content (% w/w).

Harvesting year	Seed oil content (%)	Seed weight (g)
Fourth	$55.88^b \pm 0.057$	$3.683^a \pm 0.086$
Fifth	$55.99^a \pm 0.068$	$3.557^b \pm 0.014$
Sixth	$55.99^a \pm 0.075$	$3.582^b \pm 0.082$
P value	<0.0001	0.0004

Note: different letters (a, b) showed the significant difference in the same column at 0.05 level.

effectiveness of the pretreatment and extracting steps such as crushing, pressing, and solvent extraction. Figure 5 shows images of the longitudinal section. The inside structure of the TC seed was quite dense but there were many pores with various sizes. Additionally, the images of transverse section (Figure 6) present the large hole in the center of the seed. The whole shape looks like an ellipse with the long diameter about 700 μm and the short diameter about 300 μm. The other areas of the transverse section were rather rough.

Due to the porosities inside, the TC seeds were not only easily broken by pressing force but also easily absorbed solvent for oil extraction. In many previous studies, the oleaginous seeds were often dried or heated for creating more porosities inside before oil extraction, and the scanning electronic microscope were used to evaluate the inside microstructure of the materials [48–50].

3.3. Effect of Extraction Methods on Seed Oil Yield. In our project, TC seeds was pretreated by drying and/or crushing then oil extracted by different methods including cold screw pressing (M1), hot screw pressing (M2), hydraulic pressing (M3), solvent extracting (M4), and cold screw pressing combining with oil cake solvent extracting (M5). The oil extraction yields (OEY) and residual oil (RO) or cake oil contents of these five methods were indicated. As in Table 2, the physical methods (M1, M2, and M3) presented significantly lower oil yield (from 40.59% to 77.32%) and higher residual oil (RO) mass percentage (>10%) than the solvent extraction method (M4) and the combined method (cold screw pressing and cake solvent extraction–M5).

Among the physical extraction methods, cold screw pressing observed the highest EOY (77.32%). In this process, the seeds were pretreated by freezing—thawing. In the frozen—thawed samples, the seed cell wall is often destroyed by ice crystal formation and then creates more pores/holes for oil moving out of the material. Our result agreed with some recent studies. In detail, this pretreatment step was also used by Lee et al. and the perilla seed oil yield was 2.5 times greater than the control sample [51]. In another report, Gurdil et al. was investigated the effect of freezing temperature on sunflower seed hardness, the results showed that the range temperature from -2°C to (-12°C) reduced the deformation energy while the temperature of -36°C was not influenced on the seed property [52].

About the hot screw pressing method (M2), this process obtained the lowest extraction oil yield (EOY) of 40.59% due to the extremely high heating temperature (230°C) which caused protein denaturation and reduced the EOY. In some previous studies, the suitably high temperature could rupture the seed cell wall, decrease the viscosity of the seed oil, in this case, the oil is easily obtained. The heating temperature of presser often is about 80-100°C which were positive influence on oil pressing [52–55].

In particular, in our experimental conditions, the most effective method for isolating TC seed oil was the combined method (M5) in which the oil was extracted twice. Firstly, the cold screw presser was utilized as in M1 process and the EOY after pressing was 77.32%. After that, the oil cake was extracted by solvent with the same conditions of M4 process and increases the final EOY of 14.61%. Therefore, the M5 method obtained not only the highest EOY (91.93%) but also reduced the residual oil mass percentage to the lowest value of 3.22%.

In a study by Akubude et al. [53], TC seed oil was also collected by physical method, mechanical expression, and the expression efficiency observed 76.35%. That value was higher than the hot screw pressing method (M2) and hydraulic pressing method (M3), but lower than the cold

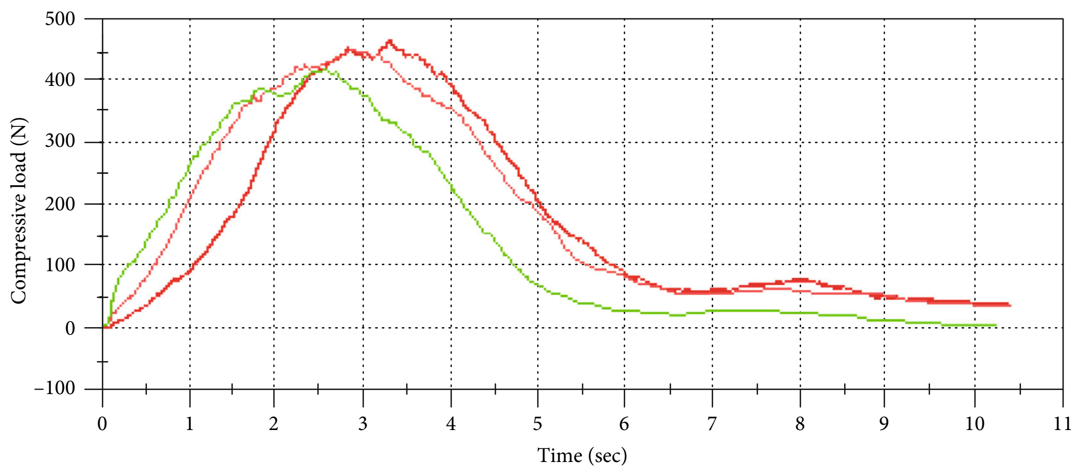

FIGURE 4: *Terminalia catappa* L. seed hardness. Note: the three curves present three times of hardness determination.

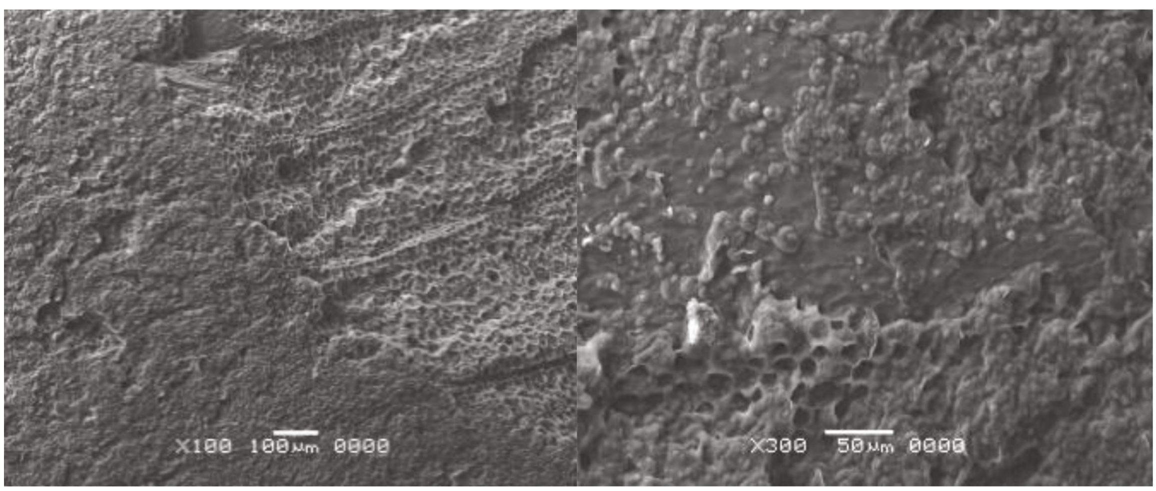

FIGURE 5: Scanning electron micrograph of longitudinal section of *Terminalia catappa* L. seed.

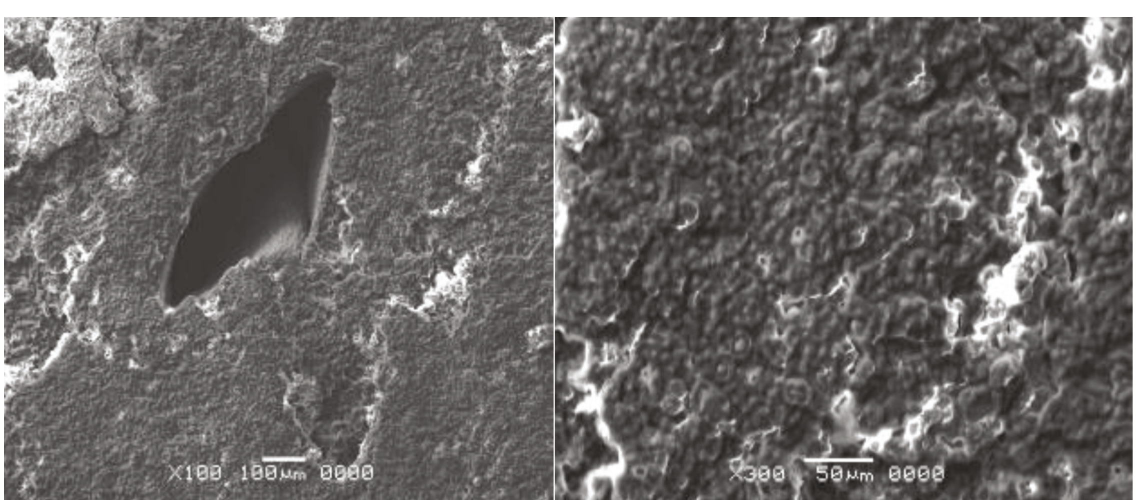

FIGURE 6: Scanning electron micrograph of transverse section of *Terminalia catappa* L. seed.

Table 2: Extraction oil yield and residual oil content of different extraction method.

Extraction methods	Oil extraction yield (%)	Residual oil content (%)
M1	$77.32^c \pm 0.260$	$12.29^b \pm 0.130$
M2	$40.59^e \pm 3.343$	$18.64^d \pm 0.028$
M3	$68.75^d \pm 0.602$	$15.04^c \pm 0.369$
M4	$89.42^b \pm 0.088$	$3.34^a \pm 0.024$
M5	$91.93^a \pm 0.042$	$3.22^a \pm 0.045$
P value	<0.0001	<0.0001

Note: different letters (a, b) showed the significant difference in the same column at 0.05 level. The oil extraction methods: cold screw pressing (M1), hot screw pressing (M2), hydraulic pressing (M3), solvent extracting (M4), and cold screw pressing combined with cake solvent extraction (M5).

screw pressing method (M1) in our project. In another work, the oil yield about 70% was also obtained when extracted oil from jatropha seeds by twin-screw presser [54].

In some previous projects, the same discussions to ours were reported. The physical and chemical extraction methods were also applied in a study by Bhuiya et al. [56]. In that work, Australian native beauty leaf seed was oil extracted by screw pressing and solvent extracting (using hexane as the solvent). Those results also showed the advantages of solvent extraction, particularly, the mechanical pressing gained lower oil yield (<40%) than the chemical extraction (>50%). Can Cauich et al. (2019, 2021) investigate the effect of cold pressing and hexane extraction on oil yield and oil quality of pumpkin seeds [44, 57]. Those authors reported that the cold pressing gained lower oil yield (<35%) than solvent extraction (>45%). However, the seed oil isolated by solvent extraction contained lower concentration of bioactive compounds such as polyphenol, carotenoid, chlorophyll, and squalene so that led to lower antioxidant capacity. The bioactive compounds (polyphenol, hydroxy benzoic acid, benzoic acid, cinnamic acid, and thymoquinone) of black seed oil extracted by different methods (cold pressing, Soxhlet extraction, and microwave-assisted extraction) were determined by Kiralan et al. and the results showed that the oil of cold pressing got the highest contents of these substances [58].

In conclusion, the mechanical methods often observe the lower oil yield or extraction efficiency than solvent extraction method but maintain high bioactive compounds content [44, 56–59]. In our study, the best method to extract TC seed oil was the combined method (M5), in this process, the majority oil content (over 70%) was collected by cold screw pressing at low temperature (<45°C) and the residual oil was extracted by solvent extracting. Besides the high EOY observation, this combined method could maintain the bioactive components due to the minority content of oil (about 20%) directly mixed with solvent.

3.4. Effect of Extraction Methods on Terminalia catappa Seed Oil Quality. Besides the extraction efficiency, most of the projects on this field investigated the oil quality by testing some common parameters including acid value (AV), free fatty acid (FFA), saponification value (SV), iodine value (IV), and peroxide value (PV) [43, 46, 54, 58, 60]. Some factors could influence the oil quality such as high temperature of pretreatment, extraction, oil refining, or reacting with solvent extraction. For example, when collected oil from *Thevetia peruviana* seed [46] and black cumin seed [58] by two various methods: cold pressing and solvent extraction, the FFAs or AVs of the latter method were higher than those of the other. In the study by Evon et al., a twin-screw presser was used to extract jatropha seed oil, and the alteration of the operation conditions (e,g., temperature, screw speed, and the screw module) led to the changes of oil quality index including AV, FAA, and even IV [54].

Therefore, in our study, the extraction oil of the five methods (M1, M2, M3, M4, and M5) was indicated these parameters for finding out the suitable process for TC oil producing.

As presented in Table 3, the FFA, AV, and PV of TC oil of five methods were significantly different (P values <0.0001). In many reports, the high value of these parameters is due to the oxidation and destruction of the crude oil during the extraction and refining steps [8, 53].

In detail, the M2 (hot screw pressing) obtained the highest FFA, AV, and PV and M3 (hydraulic pressing) showed lower of those than M2. In the group of physical methods, the M1 (cold screw pressing) gained the best oil quality with low FFA, AV, and PV of 0.550%, 1.080 mg KOH/g and 1.240 meq O_2/kg, respectively. The solvent extraction method (M4) and the combined method (M5) showed similar FFA and AV but lower than those of the physical methods. However, the PV of M4 oil was the lowest, 1.187 meq O_2/kg.

Among the oil indices, the IV and SV especially depend on the type of lipid source, but these parameters might be changed more or less because of the oil production procedure. As in Table 4, the IVs and SVs were significantly difference among the five extraction methods. Maximum changes were 3.45 g I_2/100g oil for IV and 0.8 mg KOH/g oil for SV. The change of IV and SV might cause by chemical damage and lead to the change of saturated and unsaturated fatty acid ratio. The increase in SV was discussed in some reports. Those researchers pointed out that the chemical reactions shortened the fatty acid chains and made the SV rise [6, 46, 53]. Besides, the extraction methods affected the plant cell destruction degree and the extraction efficiency, so fatty acid profile and various oil indices, including IV and SV in the extracted oil were changed [61–63]. For example, by applying four extraction methods for muskmelon seed oil production, the results presented not only different oil extraction efficiency but also different oil indices between these four methods (pressing, organic extracting, Soxhlet extracting and aqueous extracting) [63].

In addition, in our study, the oil indices of TC seed were acceptable to food usage: low AV (<6.6 mg KOH/g oil), low PV (<10 meq O_2/kg oil) and SV and IV in ranges of 184–196 mg KOH/g and 75–97 g I_2/100 g oil, respectively [64].

In the research of Akubude et al. [53], the TC seed oil was extracted by mechanical expression at the temperature

Table 3: Effect of extraction methods on FFA, AV, and PV of seed oil.

Extraction methods	FFA (%)	AV (mg KOH/g)	PV (meq O_2/kg oil)
M1	$0.550^c \pm 0.010$	$1.080^c \pm 0.130$	$1.240^c \pm 0.010$
M2	$0.700^a \pm 0.010$	$1.393^a \pm 0.028$	$1.383^a \pm 0.012$
M3	$0.620^b \pm 0.010$	$1.197^b \pm 0.369$	$1.327^b \pm 0.006$
M4	$0.503^d \pm 0.012$	$0.810^d \pm 0.024$	$1.187^e \pm 0.006$
M5	$0.503^d \pm 0.012$	$0.813^d \pm 0.045$	$1.203^d \pm 0.006$
P value	<0.0001	<0.0001	<0.0001

Note: different letters (a, b, c, …) showed the significant difference in the same column at 0.05 level. The oil extraction methods: cold screw pressing (M1), hot screw pressing (M2), hydraulic pressing (M3), solvent extracting (M4), and cold screw pressing combined with cake solvent extraction (M5). FFA: free fatty acid; AV: acid value; PV: peroxide value.

Table 4: Effect of extraction methods on IV and SV of seed oil.

Extraction methods	IV (g I_2/100 g oil)	SV (mg KOH/g)
M1	$79.89^c \pm 0.006$	$196.63^c \pm 0.006$
M2	$76.63^d \pm 0.006$	$196.17^e \pm 0.006$
M3	$76.44^e \pm 0.006$	$196.18^d \pm 0.012$
M4	$77.02^a \pm 0.006$	$196.97^a \pm 0.087$
M5	$77.00^b \pm 0.006$	$196.79^b \pm 0.006$
P value	<0.0001	<0.0001

Note: different letters (a, b, c, …) showed the significant difference in the same column at 0.05 level. The oil extraction methods: cold screw pressing (M1), hot screw pressing (M2), hydraulic pressing (M3), solvent extracting (M4), and cold screw pressing combined with cake solvent extraction (M5). IV: iodine value; SV: saponification value.

from 80 to 100°C, the AV and PV (in range of 1.009–2.693 mg NaOH/g and 2.700–2.812 meq O_2/kg oil, respectively) were quite higher than those of our product. The high values of AV and PV are caused by the heating influence on the TC seed oil quality. Additionally, the SV (125.6–128.6 mg NaOH/g oil) was remarkably lower than that of our TC seed oil. That low SV might be due to the heating destruction of the oil in the pressing step or the origin of the material.

In Nigeria, TC seed was also investigated as an oleaginous material, Adu et al. [8] studied the TC seed oil producing procedure and recognized that the seed roasting before extraction really increased the PV but decreased the AV, IV, and SV of the crude oil. In addition, the refining steps such as degumming and bleaching could affect the oil quality and alternate the AV, PV, IV, and SV. The IV of Nigerian TC seed oil after refining was about 76 g I_2/100 g, as same as our oil but the SV was rather higher, over 300 mg KOH/g while SV of Vietnamese TC seed oil was below 200 mg KOH/g. In another study which considered TC seed oil as a dietary lipid, oil properties seemed as quite same as our product, the seed oil content of 57.5%; FFA of 1.17%, and SV of 178 mg KOH/g [9]. TC seed oil was also investigated in Benin and TC seed oils from other origins (Nigeria and Congo) were compared. The results showed that, the seed oil content was between 52% and 62%; the SV and IV were in a large range of 128–207 mg KOH/g and 65–83 g I_2/100 g, respectively. However, the AV and PV of our product were a little lower than those of the TC seed oil from these African countries [15].

In general, the five extraction methods were really affected the TC seed oil quality. In our study, the cold screw pressing method (M1) and the combine method (M5) not only obtained the high extraction oil yield but also maintained the good quality of the oil. However, the cold screw pressing method (M1) was also a suitable solution for TC seed oil extraction in industrial scale due to the cheaper production cost without solvent usage.

4. Conclusion

In conclusion, Vietnamese *Terminalia catappa* seed containing over 50% oil is suitable for oil extraction. The results showed that the tree location site, the tree age, and the annual harvest time really affected the seed weight and the seed oil content. Therefore, the tree at the eastern side of the ground obtained the highest weight of 3.067 g and the highest seed oil content of 56.38%. Besides, the TC seeds could harvest for oil extraction when the trees are over four years old. Among the physical methods, the cold screw pressing gained the quite high oil yield of 77.32% with the good oil quality. In addition, the combination of cold screw pressing, and solvent extraction of oil cake obtained the highest oil yield (91.93%) of all. However, in industrial scale, the cold screw pressing was more suitable due to the cheaper production cost without usage of solvent and evaporation solvent after extraction.

Data Availability

Data are available on request from the authors.

Additional Points

Highlight. (i) *Terminalia catappa* L. seeds originated in Vietnam contain high oil concentration (>55%). (ii) The cold screw pressing method not only observed a high oil yield with good quality but also was easily scaled up with low operating cost. (iii) The *Terminalia catappa* L. seed oil quality gained CODEX Standard for edible oil.

Conflicts of Interest

The authors declare no conflict of interest.

Authors' Contributions

Le Hong Nguy was assigned in data curation (equal), investigation (equal), writing—original draft (equal), and software (equal). Dong Thi Anh Dao was tasked in conceptualization (equal), investigation (equal), methodology (equal), supervision (equal), writing review, and editing (equal). Ly Thi Minh Hien was assigned in investigation, writing—original draft (equal), and software (equal).

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