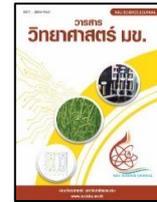




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การสกัดและการวิเคราะห์คุณลักษณะของน้ำมันเมล็ดมะม่วงจากสายพันธุ์ มันเดือนเก้าสำหรับการใช้ในเครื่องสำอาง

Extraction and Characterization of Mango Seed Oil from Mun Duean Kao Cultivar for Cosmetic Applications

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บทคัดย่อ

งานวิจัยนี้มุ่งเน้นการสกัดและวิเคราะห์คุณสมบัติของน้ำมันจากเนื้อในเมล็ดมะม่วงสายพันธุ์มันเดือนเก้าหรือ ทวายเดือนเก้า เพื่อเพิ่มมูลค่าและนำไปใช้ประโยชน์ในผลิตภัณฑ์เครื่องสำอาง ทำการวิเคราะห์คุณสมบัติทางเคมีกายภาพและ สารออกฤทธิ์ทางชีวภาพของน้ำมันเมล็ดมะม่วง และประยุกต์ใช้น้ำมันเมล็ดมะม่วงสกัดในผลิตภัณฑ์มอยเจอร์บาร์ จากนั้น ทดสอบฤทธิ์ต้านอนุมูลอิสระด้วยวิธี DPPH รวมถึงวิเคราะห์ฤทธิ์ในการยับยั้งเอนไซม์ไทโรซิเนสของผลิตภัณฑ์มอยเจอร์บาร์ และน้ำมันเมล็ดมะม่วงที่สกัดได้ ผลการศึกษาพบว่าวิธีสกัดแบบ Reflux extraction โดยใช้เฮกเซนเป็นตัวทำละลาย ในอัตราส่วน 1:3 ที่อุณหภูมิ 55 องศาเซลเซียส ให้ปริมาณน้ำมันเมล็ดมะม่วงสกัดสูงสุด (ร้อยละ 8.71) น้ำมันที่ได้มีคุณภาพดี ไม่เหม็นหืน มีค่าไอโอดีน $40.17 \pm 0.14 \text{ g I}_2/100\text{g oil}$ ค่าสaponification 200.46 $\pm 1.03 \text{ mg KOH/g oil}$ ค่าความเป็นกรด 2.81 $\pm 0.06 \text{ mg KOH/g oil}$ ค่าเพอร์ออกไซด์ 2.98 $\pm 0.03 \text{ meq peroxide/Kg oil}$ และมีค่าความหนืดใกล้เคียงกับ น้ำมันพืชชนิดอื่น (50.51 $\pm 0.01 \text{ cP}$) มีปริมาณสารสำคัญของปีตาแคโรทีน 3.48 mg/100g ฟีนอลิกทั้งหมด 4.57 mg GAE/g และ วิตามินอี 1.51 mg/100g น้ำมันเมล็ดมะม่วงสกัดและผลิตภัณฑ์มอยเจอร์บาร์ที่พัฒนาขึ้นมีฤทธิ์ต้านอนุมูลอิสระ สูง พบว่ามีค่า IC₅₀ 0.23 และ 0.19 มิลลิกรัม/มิลลิลิตร ตามลำดับ เมื่อเทียบกับกรดแอสคอร์บิก (0.18 มิลลิกรัม/มิลลิลิตร) อย่างไรก็ตามฤทธิ์ยับยั้งเอนไซม์ไทโรซิเนสของน้ำมันเมล็ดมะม่วงสกัดและผลิตภัณฑ์มอยเจอร์บาร์ยังอยู่ในระดับต่ำ มีค่า IC₅₀ 0.54 และ 0.47 มิลลิกรัม/มิลลิลิตร ตามลำดับ เมื่อเทียบกับกรดโคจิก (0.02 มิลลิกรัม/มิลลิลิตร) และกรดแอสคอร์บิก (0.09 มิลลิกรัม/มิลลิลิตร) ผลการศึกษานี้ชี้ให้เห็นว่าน้ำมันเมล็ดมะม่วงเป็นส่วนผสมจากธรรมชาติที่สามารถนำไปใช้พัฒนาผลิตภัณฑ์ เครื่องสำอาง โดยเฉพาะอย่างยิ่งเป็นสารต้านอนุมูลอิสระ

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ABSTRACT

This research investigates the extraction and characterization of mango seed oil from the Mun Duean Kao (also known as Tawai Duean Kao) cultivar to enhance its value and utilize this oil in cosmetic products. The physicochemical properties and bioactive compounds of mango seed oil were analyzed and used as an active ingredient in a moisturizer bar product. Mango seed oil and the moisturizer bar were evaluated for antioxidant activity by the DPPH method and tyrosinase inhibitory activity. Reflux extraction using hexane as a solvent at a ratio of 1:3 at 55°C yielded the highest amount of extracted mango seed oil (8.71%). The oil exhibited good quality and not rancid, with an iodine value of 40.17 ± 0.14 g I₂/100 g oil, a saponification value of 200.46 ± 1.03 mg KOH/g oil, an acid value of 2.81 ± 0.06 mg KOH/g oil, a peroxide value of 2.98 ± 0.03 meq peroxide/kg oil, and a viscosity comparable to other vegetable oils (50.51 ± 0.01 cP). The oil contains bioactive compounds, including β -carotene at 3.48 mg/100 g, total phenolic contents at 4.57 mg GAE/g, and vitamin E at 1.51 mg/100 g. Both the oil and the moisturiser bar demonstrated strong antioxidant activity, with IC₅₀ of 0.23 and 0.19 mg/mL, respectively, compared with ascorbic acid (0.18 mg/mL). However, the anti-tyrosinase activity of both remained relatively low, with IC₅₀ of 0.54 and 0.47 mg/mL, respectively, compared with kojic acid (0.02 mg/mL) and ascorbic acid (0.09 mg/mL). The results indicated that mango seed oil can be used in cosmetic products, especially as a natural antioxidant.

คำสำคัญ: มันเดือนเก้า น้ำมันเมล็ดมะม่วง สารต้านอนุมูลอิสระ การยับยั้งเอนไซม์ไทโรซิเนส มอยเจอร์บาร์

Keywords: Mun Duean Kao, Mango Seed Oil, Antioxidant, Anti-tyrosinase, Moisture Bar

INTRODUCTION

Mango (*Mangifera indica* L.) is one of Thailand's most economically significant fruits, with an annual production of 1 - 1.4 million tons, particularly abundant in Chachoengsao Province. Consumption and processing generate substantial amounts of waste, especially mango seeds, which are typically discarded despite their potential as a valuable resource. Mango seed kernels contain oil rich in unsaturated fatty acids and bioactive compounds, including phenolic compounds and tocopherols, which are recognized for their antioxidants, antimicrobial, and skin-protective properties (Puravankara *et al.*, 2000; Ribeiro and Schieber, 2010).

Previous studies have reported that mango seed oil exhibits promising characteristics, such as antioxidant activity, antimicrobial potential (Abdalla *et al.*, 2007), and tyrosinase inhibition (Schieber *et al.*, 2003; Mahato, 2019; Quintana, 2021), that support its potential for cosmetic applications. Research on mango seed oil from different cultivars has demonstrated variable yields and physicochemical properties depending on extraction conditions. For example, Nzikou *et al.* (2010) extracted mango seed oil from Congo cultivars using Soxhlet, reporting a 14% yield and favorable quality indices for cosmetic use. Similarly, Olajumoke (2013) showed compositional variability and confirmed its suitability for cosmetic applications. However, comprehensive studies on Thai cultivars remain scarce, particularly for *Mango indica* L. 'Mun Duean Kao' (also known as Thawai Duean Kao), a variety widely grown in Chachoengsao Province. In addition, there is currently no optimized extraction protocol tailored to this cultivar, nor

sufficient data on its physicochemical properties and bioactivity in cosmetic formulations. This knowledge gap hinders the valorization of mango seed oil as a sustainable and cost-effective alternative to conventional plant oils used in cosmetic products.

This study aimed to: (i) optimize an extraction method for Mun Duean Kao mango seed oil; (ii) evaluate its physicochemical properties, including iodine, saponification, acidity, peroxide, and viscosity values, as well as the content of bioactive compounds such as β -carotene, total phenolic, and vitamin E; and (iii) assess its antioxidant and tyrosinase inhibitory activities in both its crude oil form and when incorporated into a cosmetic moisture bar. This study is expected to provide new insights into the potential of mango seed oil from an underexplored Thai cultivar grown in Chachoengsao Province as a functional and sustainable ingredient for cosmetic applications, in line with the principles of the circular economy.

MATERIALS AND METHODS

1. Materials

All reagents were of analytical grade and obtained from commercial suppliers. α -Tocopherol, 2,2-diphenyl-1-picrylhydrazyl (DPPH), gallic acid monohydrate, kojic acid, tyrosinase, and L-3,4-dihydroxyphenylalanine (L-DOPA) were purchased from Sigma-Aldrich. Ascorbic acid was obtained from Fluka, and Folin-Ciocalteu's phenol reagent from EMD Millipore.

2. Preparation of sample materials

Samples of discarded mango seeds from the Mun Duean Kao cultivar were collected from various sources in Chachoengsao Province and fruit vendors at Rajabhat Rajanagarindra University in Bang Khla District, Chachoengsao, between October 2022 and January 2023. To prepare the seeds, the hard outer shells were manually removed to access the white kernels. The kernels were then thinly sliced and oven-dried at 60°C for 18 hours to reduce the moisture content to below 10%. A total of approximately 6 kg of kernels were processed, as the drying process is known to influence oil yield (Nader *et al.*, 2016). The moisture content was determined according to the AOAC 2000 method, which involves oven-drying at 105°C until a constant weight is achieved. The dried kernels were ground, sealed in zip-lock bags, and stored at temperatures below 8°C until the mango seed oil was extracted.

3. Optimal Conditions for Mango Seed Oil Extraction

Mango seed oil was extracted from 50 g of dried mango kernel powder using Soxhlet extraction, reflux extraction, and overhead stirring, with hexane and ethanol as solvents at sample-to-solvent ratios of 1:2, 1:3, and 1:4 (w/v), corresponding to 100, 150, and 200 mL of solvent, respectively. Soxhlet extraction was performed with a standard Soxhlet apparatus (250 mL capacity) at 60°C for 8 h, and the solvent was removed using a rotary evaporator (Buchi B-171, Switzerland) at 40°C under reduced pressure, followed by a high-vacuum pump (Edwards, UK). Reflux extractions were conducted using a reflux condenser system (general laboratory glassware, Thailand) with the optimal solvent and Soxhlet-derived conditions, while overhead stirring was carried out using an overhead stirrer (RW 20, IKA, China) under the same conditions.

Each extraction was performed in triplicate, and oil yields (% w/w) were calculated based on the initial dry weight. To determine the optimal extraction temperature, experiments were conducted at 50°C, 55°C, and 60°C using the most suitable extraction method. The optimal condition was applied for scale-up, using 4.5 kg of dried mango kernel powder for oil extraction.

4. Analysis of the physicochemical properties of extracted mango seed oil

The physicochemical properties of the extracted mango seed oil were investigated by analyzing various parameters, including iodine value, saponification value, peroxide value, and acid value, following the AOAC (2000) methods. The viscosity value was determined by heating the extracted mango seed oil at 45°C for 15 min., and then the viscosity was measured using a Viscometer (model DV2T, Brookfield Viscometer).

5. Determination of the β -carotene content

The β -carotene content was determined following the method of Nagata and Yamashita (1992), as applied in previous studies (Chaichumpoo *et al.*, 2019; Langkapin *et al.*, 2023). An acetone–hexane mixture was prepared in a 2:3 ratio. Approximately 1.00 g of mango seed oil was weighed and transferred into a conical screw-cap test tube, and 20 mL of the prepared solvent mixture was added. The sample was homogenized using a homogenizer (Joanlab, 220V, MHZ-01, China) for 1 - 2 min, sealed, and then allowed to stand at 25°C for 24 h. After settling, the clear upper layer was analyzed for absorbance using a UV–VIS spectrophotometer at wavelengths of 663, 645, 505, and 453 nm. All measurements were performed in triplicate, and the β -carotene content was calculated using the following equation.

$$\beta\text{-carotene content (mg/100 g)} = 0.216 A_{663} - 1.22 A_{645} - 0.304 A_{505} + 0.452 A_{453}$$

6. Determination of total phenolic content

Total phenolic content was determined using a modified method from Tsai *et al.* (2002). First, 1.00 g of the sample was dissolved in 10 mL of methanol and subjected to ultrasonic bath (120 W, 40 kHz, UC-5120, SZLANGEE, China) treatment for 30 min. A 5 mL aliquot of the supernatant was mixed with 5 mL of 10% Folin–Ciocalteu reagent. After 3 min, 2 mL of 7.5% Na_2CO_3 was added, and the mixture was incubated in the dark at room temperature for 1 h. Absorbance was measured at 765 nm by a UV-VIS spectrophotometer. Total phenolic content was calculated from a gallic acid standard curve and expressed as mg GAE/g extract.

7. Determination of vitamin E content

The content of Vitamin E was analyzed using a modified method from Mitsikaris *et al.* (2022). Vitamin E was extracted by mixing 5.00 g of mango seed oil with 20 mL of methanol, followed by sonication (30 min) and centrifugation (3,000 rpm, 30 min). The supernatant was analyzed by HPLC (ODS-3 GL C18, 4.6 × 150 mm) using 100% methanol as the mobile phase at a flow rate of 2.0 mL/min, detection at 295 nm, and a 20 μL injection volume.

8. Application of mango seed oil extract in cosmetic products

A prototype moisture bar was formulated using mango seed oil as the active ingredient, with shea butter and beeswax as excipients. The formulation consisted of 50.00 g of mango seed oil, 50.00 g of shea butter, 50.00 g of beeswax, and 0.50 g of mango fragrance. All ingredients were weighed and transferred into a clean beaker. The mixture was heated in a temperature-controlled water bath at 60°C until it completely melted and homogenized. Shea butter possesses emollient and moisturizing properties, while beeswax contributes to a smooth texture and helps the product maintain its form. Mango fragrance was incorporated to impart a characteristic fruity aroma. The molten mixture was poured into pre-cleaned aluminum containers, each with a net weight of approximately 30 g. After cooling at ambient temperature, the moisture bars were sealed. The resulting moisture bars were light yellow in color and exhibited a smooth surface with consistent texture.

9. Antioxidant activity of mango seed oil extract and cosmetic product

Mango seed oil and the moisturizer bar were evaluated for antioxidant activity using the DPPH method according to the method of Masuda *et al.* (1999). Ascorbic acid, mango seed oil extract, and moisture bar samples were prepared at concentrations of 0.05, 0.10, 0.15, 0.20, 0.25, and 0.30 mg/mL using 10% DMSO in ethanol as solvent. For each concentration, 100 µL of sample solution was transferred into a test tube, followed by the addition of 300 µL of 0.2 mmol/L DPPH solution and 3 mL of ethanol. The mixtures were gently shaken and incubated in the dark for 30 min. Absorbance was measured at 515 nm using a UV spectrophotometer. All concentrations were analyzed in triplicate. Antioxidant activity (% inhibition) was calculated, and IC_{50} values were obtained from the standard curve of concentration versus % inhibition. The equation used for the calculation was as follows:

$$\% \text{ Inhibition} = \frac{(\text{abs Control} - \text{abs Example})}{\text{abs Control}} \times 100$$

Where: abs Example = the absorbance measured from the sample extract mixed with the ethanolic DPPH

abs Control = absorbance measured from ethanolic DPPH mixed with the solvent used

10. Tyrosinase inhibitory activity of mango seed oil extracts and cosmetic products

The anti-tyrosinase activity was determined using the dopachrome method, adapted from Srisook *et al.* (2010). A 0.02 M sodium phosphate buffer (pH 6.8) was prepared by dissolving 1.76 g $Na_2HPO_4 \cdot 2H_2O$ and 0.69 g $Na_2HPO_4 \cdot H_2O$ in distilled water, adjusting the volume to 1,000 mL and the pH to 6.8. Tyrosinase solution (314.8 U/mL) was prepared by dissolving 10.00 mg of enzyme in 250 mL of buffer. L-DOPA solution (0.34 mM) was prepared by dissolving 16.76 mg in 250 mL of buffer. Sample solutions (mango seed oil extract, moisture bar, ascorbic acid, kojic acid) were prepared in ethanol at concentrations of 0.01, 0.05, 0.10, 0.50, and 1.00 mg/mL. Tyrosinase inhibitory activity was assessed using the Dopachrome method. Each sample concentration was tested in triplicate. Ascorbic acid and kojic acid were used as standards. Solutions A–D were added to the test tubes according to Table 1.

Table 1 Conditions for the tyrosinase inhibitory activity test

Tube	Tyrosinase 314.8 unit/mL	0.02 M Sodium Phosphate Buffer (pH 6.8)	Ethanol	Standard or Sample	L-DOPA 0.34 mM
A	0.50 mL	1.00 mL	0.50 mL	-	0.50 mL
B	-	1.00 mL	0.50 mL	-	0.50 mL
C	0.50 mL	1.00 mL	-	0.50 mL	0.50 mL
D	-	1.00 mL	-	0.50 mL	0.50 mL

Sample and standard solutions at various concentrations were pipetted and mixed with 0.1 M phosphate buffer (pH 6.8) and tyrosinase enzyme (314.8 U/mL) in the volumes indicated in Table 1. The mixtures were shaken to ensure thorough mixing and incubated at 25°C for 10 min. Subsequently, 0.50 mL of 0.34 mM L-DOPA solution was added to each test tube, followed by incubation for 20 min. Absorbance was measured at 492 nm, and the tyrosinase inhibitory activity was calculated as a percentage using ascorbic acid and kojic acid as standards according to the formula:

$$\% \text{ Tyrosinase inhibition} = \left[\frac{(A-B)-(C-D)}{(A-B)} \right] \times 100$$

Where: A = Absorbance at 492 nm at 20 min in the absence of test substance but with tyrosinase

B = Absorbance at 492 nm at 20 min in the absence of both test substance and tyrosinase

C = Absorbance at 492 nm at 20 min in the presence of both test substance and tyrosinase

D = Absorbance at 492 nm at 20 min in the presence of test substance but without tyrosinase

11. Statistical analyses

Statistical data were analyzed and reported as mean \pm standard deviation (S.D.). The SPSS version 27 program was used to analyze the experimental results at the 95% statistical difference level (one-way analysis of variance (ANOVA)), and the mean comparisons were performed using the Duncan New Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

1. Extracted mango seed oil yield

The optimum conditions for mango seed oil extraction are summarized in Table 2. Soxhlet extraction demonstrated that hexane was more effective than ethanol as a solvent, with an optimal seed-to-solvent ratio of 1:3. This ratio provided sufficient solvent volume to maximize solute-solvent interactions and ensure efficient lipid transfer without excessive dilution, while lower (1:2) or higher (1:4) ratios gave no significant improvement, consistent with previous reports that oil recovery efficiency increases only up to a saturation point (Mas'ud *et al.*, 2018). Among the evaluated extraction methods, reflux extraction at 55°C produced the highest oil yield. Although extraction at 60°C gave a slightly higher yield, it also caused brown coloration and sediment formation, likely due to thermal degradation and polymerization of unsaturated fatty acids and phenolics, as well as oxidation and Maillard-type reactions, which reduced storage stability

(Sultana and Ashraf, 2019; Mwaurah *et al.*, 2020). Statistical analysis confirmed that the extraction method has a significant influence on oil recovery ($p \leq 0.05$). Reflux extraction at 55 - 60°C yielded the highest oil contents (8.70 - 8.91% w/w, group a), followed by reflux at 50°C and overhead stirring (8.34 - 8.62% w/w, group b), which were both higher than Soxhlet extraction with hexane (7.34% w/w, group c). Ethanol extraction produced the lowest yield (5.14% w/w, group e). These results indicate that hexane and optimal reflux temperatures (55°C) are the optimal conditions for maximizing oil extraction efficiency. For large-scale extraction, 4.5 kg of dried mango kernel powder yielded 391.87 g of oil (8.71% w/w). The extracted oil was stored in amber bottles under refrigeration to preserve quality.

Table 2 Extracted mango seed oil content (%w/w)

Oil extraction method	Extracted mango seed oil content (%w/w)			Average \pm S.D.
	1	2	3	
Soxhlet extraction ^{A1}	7.39	7.35	7.29	7.34 \pm 0.05 ^c
Soxhlet extraction ^B	5.16	5.13	5.14	5.14 \pm 0.02 ^d
Soxhlet extraction ^{A2}	7.39	7.35	7.29	7.34 \pm 0.05 ^c
Soxhlet extraction ^{A3}	7.39	7.35	7.29	7.34 \pm 0.05 ^c
Overhead stirrer ^{C1}	7.72	7.69	7.72	8.62 \pm 0.02 ^b
Reflux extraction ^{C1}	8.35	8.33	8.35	8.34 \pm 0.01 ^b
Reflux extraction^{C2}	8.70	8.69	8.70	8.70 \pm 0.01^a
Reflux extraction ^{C3}	8.89	8.92	8.92	8.91 \pm 0.02 ^a

^{A1} = use hexane as solvent (1:3)

^{A2} = use hexane as solvent (1:2) at 50°C

^{A3} = use hexane as solvent (1:4) at 50°C

^B = use ethanol as solvent

^{C1} = use hexane as solvent (1:3) at 50°C

^{C2} = use hexane as solvent (1:3) at 50°C

^{C3} = use hexane as solvent (1:3) at 60°C. Each experiment was performed in triplicate.

^{a-d} = the meaning of data in the same row with different letters indicating statistical difference ($p \leq 0.05$).

2. Physicochemical properties of mango seed oils

The physicochemical properties of the extracted mango seed oil were evaluated, including iodine value, saponification value, peroxide value, acid value (AOAC, 2000), and viscosity values. The results are presented in Table 3.

Table 3 Physicochemical properties of mango seed oils

Physicochemical properties	Analytical results \pm S.D.
1. Iodine value (g I ₂ /100g oil)	40.17 \pm 0.14
2. Saponification value (mg KOH/g oil)	200.46 \pm 1.03
3. Acid value (mg KOH/g oil)	2.81 \pm 0.06
4. Peroxide value (meq peroxide/Kg oil)	2.98 \pm 0.03
5. Viscosity value (cP)	50.51 \pm 0.01

Means of triplicate determinations \pm standard deviation (S.D.).

The iodine value, saponification value, acidity value, and peroxide value are key indicators of oil quality. The iodine value of the mango seed oil extract was 40.17 ± 0.14 g I₂/100 g oil. This parameter reflects the degree of unsaturation in the fatty acid composition, with higher values indicating greater proportions of unsaturated fatty acids and a higher number of double bonds. The measured value suggests a moderate level of unsaturation in mango seed oil, which may influence its oxidative stability and potential applications in cosmetic formulations. The saponification value was 200.46 ± 1.03 mg KOH/g oil, which indicates relatively short fatty acid chain lengths. This result is consistent with previous reports (Nwaokobia *et al.*, 2018; Kittiphoom and Substasinee, 2013) and confirms the suitability of the oil for soap production. The acid value was 2.81 ± 0.06 mg KOH/g oil, which is below the Codex Alimentarius limit of 4 mg KOH/g for cold-pressed oils. Since acid values above 20 mg KOH/g typically indicate rancidity (Let *et al.*, 2005), the low value confirms good stability of the oil. Therefore, it is confirmed that the extracted mango seed oil does not become rancid after extraction. The peroxide value of the mango seed oil was 2.98 ± 0.03 meq peroxide/kg oil, indicating the absence of rancidity. Peroxide value is commonly used to assess the extent of lipid oxidation, as it reflects the concentration of primary oxidation products present in oils. In general, acceptable peroxide values for vegetable oils are below 20 meq/kg (Let *et al.*, 2005), while Codex Alimentarius (2019) specifies that refined oils should have values below 15 meq/kg. The viscosity of mango seed oil was determined by heating at 45°C for 15 minutes and measuring the viscosity using a viscometer. The viscosity was 50.51 cP. This is comparable to other vegetable oils, which typically exhibit viscosities in the range of 45 - 60 cP. Viscosity generally increases with longer fatty acid chain lengths and decreases with higher degrees of unsaturation. Therefore, the viscosity value indirectly supports the oil's structural integrity and resistance to oxidative degradation under thermal conditions. These findings, particularly the moderate iodine value combined with the low peroxide and acid values, further confirm the good oxidative stability of mango seed oil.

Although the current study did not include long-term stability testing, future work will focus on evaluating the accelerated storage stability of mango seed oil under various temperature conditions and through freeze-thaw cycling, and comparing its performance with standard oils commonly used in cosmetic formulations.

3. Analysis results for β -carotene, total phenolic compounds, and vitamin E content

The content of β -carotene, total phenolic compounds, and vitamin E was summarized in Table 4. The β -carotene content in mango seed oil was 3.48 ± 0.0200 mg/100 g, consistent with its pale yellow color. β -Carotene is a naturally occurring yellow to orange pigment found in fruits and vegetables, acting as a potent antioxidant and a precursor of vitamin A. Its highly unsaturated structure makes it susceptible to degradation by heat, light, and oxidation (Britton *et al.*, 1995; Rao and Rao, 2007). In mango seed oil, β -carotene contributes to color and enhances the oil's nutritional value. The total phenolic content was determined using the Folin-Ciocalteu method, yielding a value of 4.57 ± 0.0004 mg GAE/g oil. This indicated that the oil has a moderate antioxidant potential, contributing to oxidative stability and functional properties (Shahidi and Ambigaipalan, 2015). Additionally, the Vitamin E content was quantified using α -tocopherol as a standard, which was found to be 1.51 ± 0.0048 mg/100 g oil. Vitamin E further enhances antioxidant capacity and skin protection potential, making the oil suitable for cosmetic applications (Traber and Atkinson, 2007). Importantly, the gentle extraction method used preserves the integrity of β -carotene, phenolics, and vitamin E, maintaining the oil's stability during storage.

Table 4 β -carotene, total phenolic compounds, and vitamin E content

Determined quantity	Analysis results \pm S.D.
1. β -carotene (mg /100 g oil)	3.48 ± 0.0200
2. Total phenolic compounds (mg GEA/g oil)	4.57 ± 0.0004
3. Vitamin E (mg/100 g oil)	1.51 ± 0.0048

Means of triplicate determinations \pm S.D.

4. Antioxidant activity of extracted mango seed oil and moisture bar products

The antioxidant capacity of mango seed oil and a formulated moisture bar was assessed using the DPPH assay, with ascorbic acid as the reference standard. Absorbance was measured at 515 nm using a UV spectrophotometer, and all concentrations were tested in triplicate. The % inhibition was calculated, and IC_{50} values were obtained from standard curves generated by plotting concentration against % inhibition. Statistical analysis using one-way ANOVA followed by Duncan's Multiple Range Test (DMRT) revealed significant differences among the samples ($p < 0.05$). Ascorbic acid exhibited the strongest antioxidant activity ($IC_{50} = 0.18$ mg/mL), followed by the moisture bar (0.19 mg/mL), and mango seed oil alone (0.23 mg/mL). Superscript letters in Table 5 indicate statistically distinct groups. The slightly enhanced antioxidant activity of the moisture bar suggests synergistic effects from additional ingredients such as shea butter, which contains tocopherols and phenolic compounds (Honfo *et al.*, 2013). The antioxidant activity of mango seed oil can be attributed to its phenolics, vitamin E, and carotenoids, compounds known for their antioxidant properties (Sogi *et al.*, 2013). Therefore, mango seed oil can be used as an antioxidant and is more effective when used in cosmetic formulations such as moisture bars.

Table 5 The antioxidant activity of extracted mango seed oil and moisture bar product

Samples	IC ₅₀ (mg/mL) ± S.D.
1. Ascorbic acid	0.18 ± 0.0210 ^a
2. Extracted mango seed oil	0.23 ± 0.0005 ^b
3. Moisture bar product	0.19 ± 0.0003 ^a

Values are means ± S.D. (n = 3). Ascorbic acid is used as the reference standard.

^{a-b} = the meaning of data in the same row with different letters indicating statistical difference ($p \leq 0.05$).

5. Tyrosinase inhibitory activity of extracted mango seed oil and moisture bar product

The tyrosinase inhibitory activity of mango seed oil and its formulated moisture bar was evaluated using the dopachrome method at 492 nm, with kojic acid and ascorbic acid serving as reference standards. As shown in Table 6 and Figure 1, kojic acid exhibited the strongest inhibition (IC₅₀ = 0.02 mg/mL), followed by ascorbic acid (0.09 mg/mL), consistent with their well-established roles as potent skin-lightening agents (Maisuthisakul and Gordon, 2009). Mango seed oil demonstrated only mild activity (IC₅₀ = 0.54 mg/mL), in agreement with previous reports attributing its moderate inhibition to polyphenols and tannins (Namngam *et al.*, 2018). Interestingly, the moisture bar formulation showed slightly stronger inhibition (IC₅₀ = 0.48 mg/mL), suggesting a synergistic effect from added ingredients such as shea butter, which may contribute additional phenolic antioxidants. Statistical analysis using one-way ANOVA followed by DMRT confirmed significant differences among all samples ($p < 0.05$), with each forming distinct groups. These findings indicate that while mango seed oil alone exhibits modest tyrosinase inhibitory activity, its efficacy can be enhanced through formulation, supporting its potential application in natural cosmetics.

Table 6 Tyrosinase inhibitory activity of mango seed oil and moisture bar

Samples	IC ₅₀ (mg/mL) ± S.D.
1. Ascorbic acid	0.09 ± 0.0011 ^b
2. Kojic acid	0.02 ± 0.0008 ^a
3. mango seed oil	0.54 ± 0.0014 ^d
4. Moisture bar	0.47 ± 0.0009 ^c

Means of triplicate determinations ± S.D. Ascorbic acid and kojic acid are used as the reference standard.

^{a-d} = the meaning of data in the same row with different letters indicating statistical difference ($p \leq 0.05$).

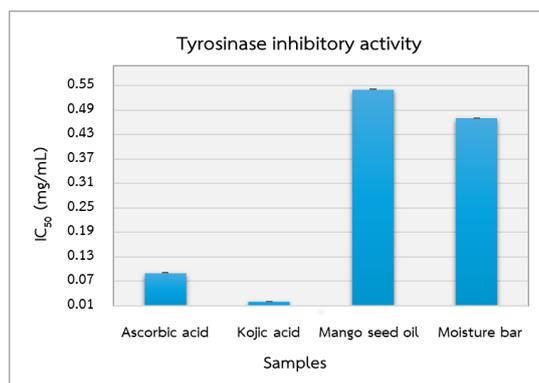


Figure 1 Tyrosinase inhibitory activity of mango seed oil, moisture bar, ascorbic acid, and kojic acid.

CONCLUSIONS

Mango seed oil can be efficiently extracted under optimized conditions using hexane at a kernel-to-solvent ratio of 1:3 and 55°C, yielding a stable oil with favorable physicochemical properties and notable levels of bioactive compounds, including β -carotene, phenolics, and vitamin E. The oil exhibited antioxidant activity comparable to standard references, and its incorporation into a moisture bar formulation further enhanced this effect, supporting its potential as a functional cosmetic ingredient. Although its tyrosinase inhibitory activity was relatively mild, the oil shows promise for skin care applications when combined with more potent active agents, underscoring its value for the development of natural, antioxidant-rich formulations. Future research will not only evaluate the antioxidant efficacy of mango seed oil *in vitro* and *in vivo* but also investigate its additional biological properties, such as anti-inflammatory, moisturizing, and photoprotective effects, to further support its application as a multifunctional ingredient in food and cosmetic products for sustainable value creation.

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