

Journal of Pharmaceutical Research International

**33(60A): 501-511, 2021; Article no.JPRI.80480** ISSN: 2456-9119 (Past name: British Journal of Pharmaceutical Research, Past ISSN: 2231-2919, NLM ID: 101631759)

## Physicochemical Characteristics and Thermal Stability of Perilla Seed Oil of Indian Origin

Akriti Dhyani <sup>a≡</sup>, Priyanka Prajapati <sup>b</sup>, Rajni Chopra <sup>c\*#</sup>, Meenakshi Garg <sup>b#</sup> and Priyanka Singh <sup>aφ</sup>

<sup>a</sup> Department of Food and Nutrition, Institute of Home Economics, Delhi University, Delhi, India. <sup>b</sup> Department of Food Technology, Bhaskaracharya College of Applied Sciences, University of Delhi, Dwarka, New Delhi, 110075, India. <sup>c</sup> Department of Food Science and Technology, National Institute of Food Technology Entrepreneurship and Management, Sonipat, Haryana, India.

#### Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

#### Article Information

DOI: 10.9734/JPRI/2021/v33i60A34511

Open Peer Review History:

This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/80480

**Original Research Article** 

Received 09 October 2021 Accepted 18 December 2021 Published 20 December 2021

### ABSTRACT

There is an increasing interest of food scientists in finding new alternatives to PUFA rich edible oil. Perilla seed oil (CPSO), an underutilized oilseed, can be used as an edible oil source. Oil extracted by the cold-pressed method from perilla seeds gives a yield of 36.50%. This study reports the physicochemical properties, the oxidative and thermal stability of the cold-pressed perilla seed oil. The viscosity, specific gravity, refractive index, and smoke point of CPSO were 28 m.Pa.s, 0.92, 1.43, and 241 °C, respectively. The peroxide, acid, iodine, saponification value, and unsaponified matter of CPSO were 4.81 meq O2/kg oil, 1.61 g KOH/kg oil, 132 g KOH/kg oil, 180 g I2/kg oil, and 0.64%, respectively. It consists of high  $\alpha$  -linolenic acid (55.80% of total oil) followed by oleic acid (20.54%). The extracted oil is analyzed for its thermal stability (peroxide value, free fatty acids, p-anisidine value, totox value, and total polar compounds) and storage stability for 120 days in two different storage conditions (refrigerated and room temperature). Despite having high nutritional benefits, the oil stability index (0.50 h) of the perilla seed oil is low, limiting its utilization as a frying oil. Therefore, perilla seed oil requires process optimization to increase its stability during heating.

- #Associate Professor;
- <sup>®</sup>Research scholar

<sup>&</sup>lt;sup>■</sup>Senior Research Fellow;

<sup>\*</sup>Corresponding author: E-mail: rajnichopra145@gmail.com;

Keywords:  $\alpha$  – linolenic fatty acid; oxidative stability; perilla seed oil; thermal stability.

## **1. INTRODUCTION**

Nowadays, there is a significant focus on utilizing plant oils due to their healthy bioactive and polyunsaturated fatty acid constituents (PUFAs). The nutritional quality of fats and oils depends on the type of fatty acid, degree of unsaturation, and arrangement of fatty acid in triacylglycerol structure [1]. Several researchers proved a direct association of type and amount of oil consumption with cardiovascular diseases CVDs. Therefore, the focus of nutrition research has shifted to replacing saturated and trans fats with healthy PUFA rich oils [2]. Most commonly consumed oils are rich in saturated and monounsaturated fatty acids but deficient in PUFA. a -Linolenic acid and linoleic acids are important types of PUFA and are classified as essential fatty acids. Numerous studies have proven the role of these essential fatty acids in normal growth and development and in various diseases preventing such as cardiovascular, dyslipidemia, hypertension, diabetes mellitus, obesity, inflammatory diseases [3]. Fish oil is the richest animal source of essential fatty acid. However, it is not suitable for the vegetarian population. Thus, scientists and food manufacturers are interested in plant-based PUFA rich oils with the increasing demand for essential fatty acids.

Perilla plant (Perilla frutescence) is a member of the Lamiaceae family and is commonly called perilla. This annual crop is native to India, China, Korea, and Japan. The valuable parts of the perilla plant are its seeds and leaves. The major constituents of perilla seeds include 42-45% of oil and 25 % of protein [4]. Perilla seed oil is a golden yellow clear and transparent liquid with a pungent odor. The oil consists of 78% unsaturated fatty acids, 60% of which is  $\alpha$  - linolenic acid. Other unsaturated fatty acids are oleic acid and linoleic acid. This oil also contains about 6.7 -7% saturated fatty acids [5].

Numerous studies have shown that the consumption of perilla seed oil is associated with a lower level of blood lipids and serum cholesterol. Our previous review paper compiled the pharmacological properties of perilla oil such as anti-asthmatic, antidiabetic, anti-depressant, anti-cancer, anti-microbial, antioxidant, cardioprotective, and neuroprotective [6]. Perilla seed oil has been recently used as a functional food due to its good health effects in various countries such as China, Japan, Korea, and the

U.S.A. Japanese and Chinese manufacturers have added perilla seed oil to various products for the production of new health food [7]. There are numerous researches on the nutritional composition and physicochemical characteristics of perilla seed oil varieties from different countries, e.g., India [8], Iran [9], China [10], Japan [11], Korea [12], Bangladesh [13] considering its scope as an alternative PUFA n-3 rich vegetable oil source.

These studies only reported about the nutritional and physicochemical properties of perilla seed oil. No previous study has been conducted on the oxidative and thermal stability of perilla seed oil, which are essential parameters for the processing and utilization of oil. Therefore, this study aimed to investigate physicochemical characteristics, fatty acid profile, oxidative and thermal characteristics of perilla seed oil extracted by the cold-pressed method (CPSO).

## 2. MATERIALS AND METHODS

### 2.1 Materials

The perilla seeds were procured from the Forest Research Institute (F.R.I.), Dehradun, India. All reagents used were E. Merck or Sigma Aldrich.

### 2.2 Oil Extraction

Perilla oil from perilla seed was extracted by cold-pressed method (Screw Press Model 85 mm) in two batches (1 kg in one batch) at 10 MPa for 20 min from a commercial mill (Mohan oil mill) in New Delhi, India. The extracted oil was filtered through muslin cloth and Whatman no.2 filter paper to remove the impurities and stored in a sealed dark amber bottle until further use [14].

#### 2.3 Determination of Physical Properties

The color was measured using the Lovibond tintometer (Model F) method; the color intensity was measured in 1" cell in the transmittance mode of white glass filter, yellow glass filter, and red glass color and expressed as 5R+Y Lovibond units, the method given by AOCS Method no. Cc 13e-92 (AOCS, 2000)[15]. The moisture content of oil was a loss of weight from the initial weight on keeping the oil sample in a hot air oven for 1 hour at  $105 \pm 1^{\circ}$ C [16]. Viscosity was measured using Brookfield viscometer at constant

temperature (25°C) and shear rate. Specific gravity was measured using a 10 ml pycnometer at 20°C. The Refractive index was determined by an Abbe refractometer; the temperature of the refractometer was maintained at  $25 \pm 0.1^{\circ}$ C using a thermostatically controlled water bath. The smoke point of oil was measured according to the AOCS method [15], Cc 9a-48. First, 150 ml of oil was taken in a beaker and heated until it started producing smoke; a digital thermometer was used to measure this temperature.

#### 2.4 Determination of Chemical Properties

The peroxide value (PV), acid value (AV), iodine value (IV), saponification value (SV), and unsaponifiable matter (USM) were measured by the standard methods of the American Oil Chemist's Society (AOCS, 2000)[15].

#### 2.5 Determination of Oxidative Stability Index (Rancimat)

Oxidative stability of CPSO can be achieved by conducting an accelerated oxidation test and measured by Induction period (IP, hour) using the method given by Anwar *et al.*, 2007[17]. Oxidation induction times were measured by a Rancimat model 743 using 3 g of oil, heated at 120 °C with 20L/h airflow. At the end of the process, volatile and secondary products are formed, absorbed by measuring a vessel containing deionized water, and then measured electrical conductivity.

### 2.6 Fatty Acid Composition

Fatty acid composition by gas chromatography as per method given by Chaudhary et al., 2015 [18]. Fatty acids of the oil sample were converted to fatty acid methyl esters (FAMEs) using the IUPAC standard method (IUPAC 1987). FAMEs were analyzed with a gas chromatograph (Agilent 7890 B), equipped with a flame ionization detector (FID) and FP 2560 cephalic column (100 mm X 0.25  $\mu$ m X 0.2  $\mu$ m) coated with CP-SIL 88 as the stationary phase. The temperature of the oven was 200°C. The injector and FID temperature was 250°C. The FAMEs were expressed as relative area percentages.

The calculated oxidisability (COX) value of the perilla seed oil was calculated by the formula given by Fatemi and Hammond [19].

COX = [1(C18:1%) +10.3 (C18:2%) +21.6 (C18:3%)]/100

#### 2.7 Evaluation of Thermal Stability Test

#### 2.7.1 Heating procedure

The heating procedure was performed according to the method given by Anwar et al. [17] with slight modifications in time duration. In this method, fresh oils were heated at 180 °C in an electrical fryer with temperature control (Inalsa Professional 2 fryer, 18/8 steel, 2 L, digital timer). Successive heating was conducted for 4 days which took 6 hours of heating each day, giving total heating of 24 hours. 100 mL of oil was taken at the end of each day and stored in amber glass bottles at 4°C until used for analysis. The thermo-oxidative degradation level in oils was assessed by measuring changes in color, peroxide value, free fatty acid, p-anisidine value, totox value, oxidative stability (Rancimat), and total polar components (TPC) after each cycle. Methodology of color. peroxide value, free fatty acid was discussed in section 2.3.

#### 2.7.2 p-anisidine value (p-AV)

The p-AV measures secondary oxidation products (aldehyde content) in the oil; it was determined according to the method given by the AOCS method number Cd 18-90.

#### 2.7.3 Totox value

The TOTOX (i.e., total oxidation products) value was introduced to evaluate the stability of oils. The p-AV is often combined with PV to calculate the TOTOX value given formula as TV = 2PV + p-AV. The reason for the multiplication of PV by a factor 2 is that the PV has an additional obvious consequence on the stability of oil than the p-AV [20].

#### 2.7.4 Total polar compounds

The TPC of frying oils was determined by the Column Chromatography technique, as per the Official International Union of Pure and Applied Chemistry (IUPAC) method. It is based on separating polar compounds from non-polar components and is given by Arslan et al. [21]. The percentage of TPCs was calculated using the equation:

Total Polar compounds (%) = (weight of oil sample-weight of non-polar fraction / weight of oil samples) x 100

#### 2.7.5 Evaluation of oxidative stability

The cold-pressed perilla seed oil was stored in amber bottles in two storage conditions, at room temperature  $25 \pm 2$  °C and 4 °C for 120 days. The oxidative stability of oil was evaluated by measuring peroxide and acid value after every 30 days interval.

#### 3. RESULTS AND DISCUSSION

This study summarises the physicochemical properties of perilla seed oil of Indian origin obtained by the cold-pressed method. The results are compared with other varieties of perilla seed oil from different countries and the other PUFA rich oils such as chia seed oil and linseed oil. This paper also reports the oxidative and thermal stability of cold-pressed perilla seed oil (CPSO) and compares it with commercial refined palm olein (RPO). Palm olein is a very popular vegetable oil in India, used in many cooking applications; hence, the cold-pressed perilla seed oil (CPSO) was compared with commercially available RPO for thermal and oxidative study.

#### 3.1 Sample Preparation

Perilla seeds were procured from the forest research institute (Dehradun. Uttarakhand, India). Seeds were cleaned to remove impurities and stored in the dark at room temperature until further use.

### 3.2 Oil Extraction and Yield

Perilla seed contains a substantial amount of oil, about 38-59%. Temperature and pressure employed during the extraction process govern the oil extraction yield. In the present study, perilla oil was extracted using the cold-pressed method and contributed a yield of 36.50%. In comparison, a recent study reported a similar value (34.40%) by the cold pressing method [10]. Oil yield is a significant characteristic of oilseeds and pivots on different varieties and regions in which they are grown. Various studies have reported the different oil yield of perilla seed by employing different extraction techniques such as solvent extraction (39.61 %), mechanical expeller (38.4%), aqueous enzymatic extraction (31.28%) [10,22,23]. Several factors responsible for the variation in the oil yield during extraction are moisture content of seeds, solvent type, time, temperature, and type of extraction [22]. Furthermore, various other pretreatments of perilla seeds before extraction were tested to improve the oil yield, such as roasting of seeds, use of superheated steam treatment, use of freeze-thaw pretreatment, use of compressed fluids ( $CO_2$  and L.P.G) [24, 25, 26].

#### **3.3 Physical Parameters**

Physical parameters of Perilla oil are listed in Table 1. Color is an important physical parameter for consumer acceptance; perilla seed oil has a golden yellow appearance, and its darkness is attributed to the presence of high polyphenols content. The Lovibond Tintometer reading in the transmittance mode for perilla seed oil was 35 Lovibond units. The value is 25 units lesser than the value reported by Mojumdar and colleagues in perilla seed oil by solvent extraction, i.e., 60 Lovibond units. The lesser color value in coldpressed oil is attributed to the fact that some of the pigments are extracted along with the oil [13]. Moisture content is an important quality parameter for fats and oils; it is one of the deciding factors in oil storage conditions and processing. It was reported that 0.09% in perilla seed oil is within the acceptable range, i.e., 0.05-0.30 [27]. The high moisture content in oils increases the rate of hydrolytic breakdown, leading to the formation of free fatty acids acid and rancidity. The viscosity of the oil indicated its stability; CPSO showed a viscosity of 23 m.Pa.s. Specific gravity and refractive index are important factors for determining the quality of any vegetable oil. They both parameters are essential for the optimization of any processing technique. The specific gravity of CPSO was reported to be 0.92 at 25°C; the same value was reported by Mojumdar and collegues [13]. Specific gravity is directly proportional to chain length but inversely related to the degree of unsaturation. The specific gravity of CPSO was found to be 0.92 at 25 °C: The refractive index is a suitable and low-cost method to measure the authenticity of oil; it measures the degree of unsaturation and presence of uncommon components in the oil. It increases with the increase in chain length and double bonds. The RI of perilla oil was found to be 1.48; The value is similar to the value reported by Scapin et al. (2017) [26], and the value is within the accepted range of vegetable oils. However, chia seeds oil and Flaxseed oil also reported the same value [28]. The smoke point is the temperature at which oil starts producing continuous smoke during heating [29]. In the present study, the smoke point of CPSO is 241°C, and the value falls in the range of the literature value reported by Xu et al. (2013) [30]; the CPSO is reported to fulfill the recommended value of frying oils, i.e., above 200°C.

Physical parameters	Results
Color (5R+Y Lovibond units)	34.83±0.76
Viscosity (m.Pa.s)	28±0.16
Specific gravity	0.92±0.01
Refractive index	1.47±0.00
Smoke point (□)	241±3.20

# Table 1. Physical parameters of cold-pressed perilla seed oil (CPSO)

Results are expressed as mean ± standard deviation of three measurements

#### **3.4 Chemical Properties**

properties provide Chemical information regarding the stability and freshness of fat and oils. Peroxide and acid values measure the hydrolytic and oxidative rancidity of vegetable oils. The peroxide value of Indian origin CPSO was found to be  $4.81 \pm 0.40$ , which was higher than the perilla oil of Iranian origin (Table-2). The peroxide value of CPSO falls within the acceptable limit (15 meg O2/kg)) given by FSSAI. As per the Codex standards, the acid value of cold-pressed oils should not be more than 4.0 mg KOH/g fat or oil, and the acid value of CPSO is within the standard limit; it suggests that cold-pressed CPSO is suitable for edible purposes.

lodine value is a measure of the degree of unsaturation. Chia seeds oil and linseed oil were reported to have 54% and 64.3% more degree of unsaturation, respectively, than CPSO oil [28,31]. Iranian Perilla oil has been reported to have a higher iodine value than Indian CPSO; This might be due to the higher PUFA content found in the Iranian perilla seed oil [9]. Joshi and colleagues reported the iodine and saponification of Perilla oil in their study. It was 2.93% and 2.56% lower than the result obtained in this study [8]; this variation may be due to the different methods employed for oil extraction. The saponification value of perilla seed oil reported in this study is lesser than the commonly consumed oils such as soybean oil (189-195), sunflower oil (190-196), and virgin olive oil (190-195); thus, it indicates the presence of high molecular weight fatty acids present in the oil. The unsaponified content (%) in CPSO was in agreement with the previous findings [11]. The high unsaponified value indicates the presence of lignans, crude fiber, protein, and minerals.

Refining edible oils is important to remove any undesirable elements (trace metals, pigments, waxes, and gums) present in crude oil. This is because they might cause damage to human health and also act as a prooxidant during the storage of edible oil. For example, refining perilla oil had reported a 72.5% decrease in peroxide and 82 % in acid values while a significant increase in saponification value [32].

## 3.5 Oil Stability Index (OSI)

Oxidation stability is one of the crucial parameters for measuring the deterioration in edible oils due to oxidation. Oxidative stability index (OSI) is defined as the time required for decomposition of primary oxidation products produced by oxidation in the oil; it also indicates the oil's shelf life and is measured through rancimat test. The rancimat test is a technique in which oil is exposed to a higher temperature in the presence of excess air and temperature. Oils containing a high amount of unsaturated fatty acids are more prone to oxidation. The OSI of Perilla seed oil (0.50 hours) were less than those published for other commonly consumed vegetable oils [33], probably due to its high PUFA content. Various literature has reported a low oxidation stability index of perilla seed oil ( $\leq 1$ h) [11]; however, Ghaleshahi and colleagues reported high OSI of perilla seed oil (1.42 h) than basil seed oil and flaxseed oil [9]. Despite having high PUFA content, Perilla seed oil has more OSI than chia seed oil (0.43 h) and flaxseed oil (0.37 h) [34]. This contradiction could be associated with the higher amounts of tocols and antioxidants in perilla seed oil. This study suggests that perilla seed oil is prone to oxidative rancidity and requires innovative strategies for improving its stability.

## 3.6 Fatty Acid Composition

Table -3 summarizes the fatty acid profile of Perilla seed oil, and it was observed that the amount of  $\alpha$ -Linolenic acid (C18:3) is more in CPSO. Thus,  $\alpha$  -Linolenic acid is the dominant fatty acid, followed by oleic acid and linoleic acid in CPSO. Kim and colleagues reported a similar composition of fatty acid of perilla seed [12]. The oil extracted from the Indian Perilla variety has the lowest PUFA content among two other varieties (Iranian & South Korean). Pan and colleagues showed the fatty acid composition of perilla seed oil after every refining step and reported no significant difference among and concluded that the refining process did not alter the fatty acid composition of CPSO them [32].

Table 2. Chemical	parameters of	cold-pressed	perilla seed oil	(CPSO)
	puluini 0 0 0			

Chemical parameters	Perilla Seed oil (Present Study)	Iran origin [9]	Japan Origin [11]	Bangladesh origin [13]	China origin [10]
Peroxide Value (meq O <sub>2</sub> /kg)	4.81 ± 0.40	0.35	5.4	9.09	1.60
Acid Value (mg KOH/g)	1.61±0.02	2.12	0.74	-	0.57
Iodine Value (g/100g)	132.00±1.25	195	-	187	197.6
Saponification value (mg KOH/g)	180.00±1.25	182	-	175	189.6
Unsaponified matter (%)	0.64±0.25	1.49	0.60	0.20	0.57

Results are expressed as mean ± standard deviation of three measurements

#### Table 3. Fatty acid composition of cold-pressed perilla seed oil (CPSO)

Fatty acid composition	Perilla Seed Oil (Present study)	Iran origin [9]	Korea origin [12]	Japan origin [11]	China origin [10]	Bangladesh origin [13]
Palmitic acid (C16:0)	7.44±0.06	6.35	8.30	6.33	5.60	6.78
Palmitoleic acid (C16:1)	0.23±0.02	0.03	-	0.12	-	0.08
Stearic acid (C18:0)	2.24±0.19	1.95	1.40	1.70	1.50	1.30
Oleic acid (C18:1)	20.54±0.20	13.2	9.30	12.61	11.30	18.47
Linoleic acid (C18:2)	13.75±0.15	12.5	19.80	18.30	15.30	22.71
α-Linolenic acid (C18:3)	55.80±0.53	65.6	61.10	59.87	66.40	50.52
SFA	9.68±0.25	8.60	7.58	8.17	7.00	8.18
MUFA	20.77±0.19	13.6	16.57	12.86	11.30	18.47
PUFA	69.55± 0.42	78.3	75.85	78.17	81.70	73.23
SFA: MUFA: PUFA Ratio	1:2.1:7.1	1:1.6:7.1	1:2.1:10	1:1.6:9.5	1:1.6:11.6	1:2.25:9
Cox value	13.66±0.02	13.47	15.33	14.94	16.03	13.43

Results are expressed as mean ± standard deviation of three measurements

Table 4. Thermal stability	v of cold-pressed	perilla seed oil (	CPSO)	and commercial	refined p	alm oil (	RPO)
	<i>y</i> or oora procooa		0.00,				

Duration of thermal treatment (h)	Oil color (Lovibond units)	Peroxide value	Free fatty acids	p-Anisidine value	Totox value	Total polar compounds
0	34.83±0.76 <sup>E</sup>	4.81±0.40 <sup>E</sup>	0.93±0.05 <sup>E</sup>	2.56±0.39 <sup>E</sup>	12.18±1.01 <sup>±</sup>	8.25±0.25 <sup>±</sup>
6	65.83±1.60 <sup>D</sup>	8.05±0.35 <sup>D</sup>	2.12±0.02 <sup>D</sup>	7.12±0.07 <sup>D</sup>	23.23±0.64 <sup>D</sup>	17.37±0.80 <sup>D</sup>
12	70.83±2.36 <sup>C</sup>	10.98±0.35 <sup>C</sup>	2.34±0.05 <sup>°</sup>	15.89±0.80 <sup>C</sup>	37.86±1.42 <sup>C</sup>	20.71±0.84 <sup>C</sup>
18	97.83±1.75 <sup>B</sup>	14.04±0.93 <sup>B</sup>	251±0.02 <sup>B</sup>	20.54±0.72 <sup>B</sup>	48.62±0.92 <sup>B</sup>	25.87±1.36 <sup>B</sup>
24 CPSC	134.20±2.02 <sup>A</sup>	15.16±0.23 <sup>A</sup>	2.78±0.07 <sup>A</sup>	25.64±0.41 <sup>A</sup>	55.96±0.88 <sup>A</sup>	34.63±0.78 <sup>A</sup>
Before heating	34.83±0.76	4.81±0.40	0.93±0.05	2.56±0.39	12.18±1.01	8.25±0.25
After heating RPO	134.20±2.02	15.16±0.23	2.78±0.07	25.64±0.41	55.96±0.88	34.63±0.78
Before heating	11.43±0.40	0.95±0.15	0.15±0.0	0.41±0.15	2.31±0.39	2.44±0.20
After heating	34.80±0.81	4.92±0.27	0.56±0.05	15.05±0.60	24.86±1.15	10.87±0.79

A-E Means within each column with different superscripts are significantly (P ≤0.05) different; CPSO cold-pressed perilla seed oil; RPO commercial refined palm oil

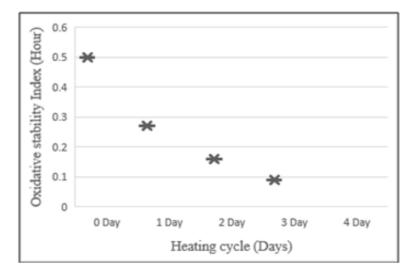


Fig. 1. Changes in oxidative stability index of perilla seed oil during heating

Dhyani et al.; JPRI, 33(60A): 501-511, 2021; Article no.JPRI.80480

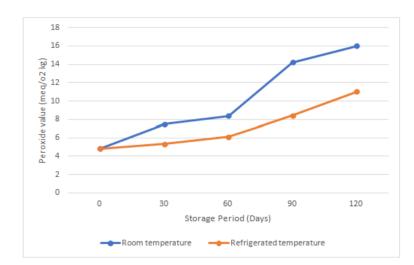


Fig. 2. Peroxide value of Cold-pressed perilla seed oil at different storage condition (Room temperature 25±2 °C; Refrigerated temp 4 °C)

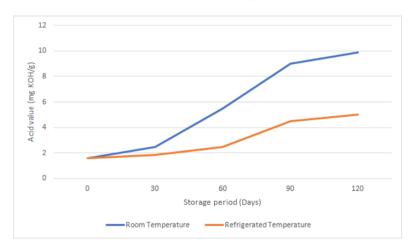


Fig. 3. Acid value of Cold-pressed perilla seed oil at different storage condition (Room temperature 25±2 °C; Refrigerated temp 4 °C)

#### 3.7 Thermal Stability of Cold Pressed Perilla Seed Oil

Thermal stability is an essential measure in quantifying the heat-induced changes in fats and oils. Measuring the quality parameters (color, peroxide value, fatty acid %, p-anisidine value, totox value, and OSI) are the essential criteria to judge the thermal stability of the oil. The changes in color, peroxide value, free fatty acid, panisidine value, totox value, and total polar compounds of CPSO in response to four heating cycles (180°C) for a total of 24 hours are reported in the Table 4. Color is the initial parameter for frying oil quality; however, color darkness increases with heating time and temperature. For example, CPSO showed a color value of 34.83, and it gained 99.37 units to a value of 134.20 after the 4th day of heating, showing more than 4 fold increase while RPO showed 3 fold increase in color intensity; This may be due to the prolonged accumulation of non-volatile compounds in the heated oil. The generation of free fatty acid is the key indicator in assessing oil stability, as free fatty acids are more prone to oxidation than neutral fatty acids present in the oil. At the end of the heating cycle, it was observed that the amount of free fatty acid in CPSO is increased by 200%, while RPO showed an FFA increase of 273% after heating (Table 4).

Peroxide value and p-anisidine value helps in the determination of primary and secondary oxidation products of fats and oils. As the duration of heating increased, a corresponding increase was observed in CPSO and RPO. However, the value did not show a significant increase at a certain point due to the formation of

secondary oxidation products. The p-anisidine value of fresh CPSO was 2.56 units higher than the Iranian variety of Perilla oil. This difference might be due to the presence of the high amount of antioxidants in Iranian perilla oil, which help in preventing oxidation [9]. However, the panisidine value of CPSO was slightly higher than the accepted p-anisidine value for good quality oil, i.e., < 2 [35]. This study reported that the panisidine, peroxide, and free fatty acids show increment with increasing heating time. For example, after heating for 24 hours at 180°C, the p-anisidine value of CPSO and RPO shows a spontaneous increase from 2.56 to 25.64 and from 0.41 to 11.22, respectively. Estimation of Totox value helps quantify the combined effect of primary and secondary oxidation products generated during oil heating. It was observed that the totox value increases significantly during the entire heating process of CPSO and RPO. The heating caused a significant and rapid increase in the TPC value of oil (Table 4). At the end of the heating cycle (4<sup>th</sup> day), the TPC level of CPSO and RPO was 34.63% and 10.87%, respectively. The recommended minimum standard limit for TPC varies from country to country and mainly ranges from 23-29% [36]; the TPC value of CPSO was within the acceptable range till 12 hours of heating.

The OSI of the oil sample is inversely proportional to the degree of unsaturation. Therefore, the OSI of oil decreases with the heating due to a higher rate of deterioration during heating; in CPSO, OSI decreases at 93%, while the decrease in RPO was 47% (Fig. 1). This higher rate of decrement of OSI in CPSO is due to the high content of unsaturated fatty acids in perilla seed oil.

#### 3.8 Oxidative Stability of Cold-Pressed Perilla Seed Oil

The storage stability of cold-pressed perilla seed oil stored at different conditions (Room temperature  $25\pm2^{\circ}$ C, Refrigerated temperature 4°C) was assessed by measuring PV and AV for 120 days at a frequent interval of 30 days and presented in Figs. 2 and 3. A high rate of increment in PV (233%) and AV (513%) was reported in oil stored at room temperature. However, in the refrigerated condition, the PV (128 %) and AV (210%) was maintained at low levels compared to room temperature. In refrigerated conditions, the percent increase of PV and AV is 2 times lower than the room temperature. Thus, it indicates refrigerated condition provide more shelf life to CPSO than room temperature.

#### 4. CONCLUSION

Perilla frutescence oilseeds is one of the novel seed traditionally used in India's Himalayan region and north-eastern area. This seed has started gaining recognition by scientists and the food industry due to its high oil yield and PUFA content. The physicochemical analysis reported in this study reports that this oil could be an excellent alternative to other PUFA rich oil. However, the oil stability index (OSI) value depicts that prolonged storage and heating or frying decreases stability. Therefore, further detailed studies need to be carried out to optimize the extraction procedure and storage conditions to increase its stability to mark profitability and use for industrial purposes.

#### DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

#### CONSENT

It is not applicable.

#### ETHICAL APPROVAL

It is not applicable.

### COMPETING INTERESTS

Authors have declared that no competing interests exist.

#### REFERENCES

- Dorni C, Sharma P, Saikia G, Longvah T. Fatty acid profile of edible oils and fats consumed in India. Food Chemistry. 2018;238:9-15.Available:https://doi.org/10.1016/j.foodc hem.2017.05.072
- 2. Ayerza R. Effects of seed color and growing locations on fatty acid content and composition of two chia (*Salvia hispanica*)

L.) genotypes. Journal of the American Oil Chemists' Society. 2010;87(10):1161-1165.

- 3. Heshmati J. Effect of omega-3 fatty acid supplementation on gene expression of inflammation, oxidative stress and cardiometabolic parameters: Systematic review and meta-analysis. Journal of Functional Foods. 2021;85:104619.
- Sargi SC, Silva BC, Santos HMC, 4. Montanher PF, Boeing JS, Santos Júnior Visentainer 00. Souza NE, JV. Antioxidant capacity and chemical composition in seeds rich in omega-3: chia, flax, and Perilla, Food Sci, Technol, 2013:33:541-548. Available:https://doi.org/10.1590/S0101-

20612013005000057.

- Ciftci ON, Przybylski R, Rudzińska M. Lipid components of flax, perilla, and chia seeds. European Journal of Lipid Science and Technology. 2012;114(7):794-800.
- Dhyani A, Chopra R, Garg M. A Review on Nutritional Value, Functional Properties and Pharmacological Application of Perilla (*Perilla Frutescens* L.). Biomedical and Pharmacology Journal. 2019;12(2):649-660.

Available:https://dx.doi.org/10.13005/bpj/1 685

- Zhu Q, He L. Study on High Value Application of Perilla Seed Oil and Meal. J food Biotechnol Res. 2018;2(1):1.
- Joshi A, Sharma A, Pandey DP, Bachheti RK. Physico-chemical properties of Perilla frutescens seeds. Der Pharma Chemica. 2015;7(5):35-41.
- 9. Ghaleshahi AZ, Ezzatpanah H, Rajabzadeh G, Ghavami M. Comparison and analysis characteristics of flax, perilla basil seed oils cultivated and in Iran. Journal Food of Science and Technology. 2020;57(4):1258-1268.
- 10. Zhao B, Fu S, Li H, Chen Z. Chemical Characterization of Chinese Perilla Seed Oil. Journal of Oleo Science. 2021;70(11): 1575-1583.
- 11. Bondioli P, Folegatti L, Rovellini P. Oils rich in alpha linolenic acid: chemical composition of perilla (*Perilla Frutescens*) seed oil. OCL. 2020;27:67.
- 12. Kim HU, Lee KR, Jeon I, et al. Fatty acid composition and oil content of seeds from perilla (*Perilla frutescens* (L.) var. *frutescens*) germplasm of Republic of

Korea. Genet Resour Crop Evol. 2019;66:1615–1624. Available:https://doi.org/10.1007/s10722-019-00803-8

- Mojumdar MAK, Hossain HMM, Uddin AFM, Meherunnessa. Characterization of fatty acids and nutrient composition of SAU Perilla-1 [(*Perilla frutescens* (L.) Britton] seeds grown with agro climatic conditions in Bangladesh. Journal of Bioscience and Agricultural Research. 2021;27(2):2307-2314.
- 14. Mazaheri Y, Torbati M, Azadmard- Damirch S, Savage GP. Oil extraction from blends of sunflower and black cumin seeds by cold press and evaluation of its physicochemical properties. Journal of Food processing and preservation. 2019;43(10):e14154. Acailable:https://doi.org/10.1111/jfpp.1415 4
- 15. AOCS (2000). Official Methods and Recommended Practices of the American Oil Chemists' Society. AOCS Press, Champaign, Illinois.
- Soltani M, Takavar A, Alimardani R. Moisture content determination of oilseeds based on dielectric measurement. Agricultural Engineering International: CIGR Journal. 2014;16(1):313-318.
- Anwar F, Hussain AI, iqbal S, Bhanger MI. Enhancement of the oxidative stability of some vegetable oils by blending with Moringa oleifera oil. Food chemistry. 2007;103:1181-1191. Available:http://dx.doi.org/10.1016/j.foodch em.2006.10.023
- Choudhary M, Grover K, Kaur G. Development of rice bran oil blends for quality improvement. Food Chemistry. 2015;770-777. Available:https://doi.org/10.1016/j.foodche m.2014.10.051
- Fatemi SH, Hammond EG. Analysis of oleate, linoleate and linolenate hydroperoxides in oxidized ester mixtures. Lipids. 1980;15:379–385.
- Nayak PK, Dash UMA, Rayaguru K, Krishnan KR. Physio- chemical changes during repeated frying of cooked oil: A Review. Journal of Food Biochemistry. 2016;40(3):371-390. Available:http://dx.doi.org/10.1111/jfbc.122 15
- 21. Arslan FN, Şapçı AN, Duru F, Kara H. A study on monitoring of frying performance

and oxidative stability of cottonseed and palm oil blends in comparison with original oils. International Journal of Food Properties. 2017;20(3):704-717.

- 22. Li H, Zhijun Z, Hou T, Xiaojun L, Tie C. Optimization of ultrasound- assisted hexane extraction of perilla oil using response surface methodology. Industrial Crops and Products. 2015;76:18-24.
- Li Y, Zhang Y, Sui X, Zhang Y, Feng H, Jiang L. Ultrasound-assisted aqueous enzymatic extraction of oil from perilla (*Perilla frutescens* L.) seeds. CETA-Journal of Food, 2014;12(1):16-21
- 24. Zhao T, Hong SI, Lee J, Lee JS, Kim IH. Impact of roasting on the chemical composition and oxidative stability of perilla oil. Journal of Food Science. 2012;77(12):C1273-C1278.
- 25. Lee KY, Rahman MS, Kim AN, Jeong EJ, Kim BG, Lee MH, Kim HJ, Choi SG. Effect of superheated steam treatment on yield, physicochemical properties and volatile profiles of perilla seed oil. LWT, 2021; 135:110240.
- Scapin G, Abaide ER, Martinsb FR, Vendruscolo RG, Mazutti MA, Wagner R, Rosa CS. Quality of perilla oil (*Perilla frutescens*) extracted with compressed CO2 and LPG. The Journal of Supercritical Fluids. 2017;130:176-182. Available:https://doi.org/10.1016/j.supflu.20 17.08.007
- 27. Choo WS, Birch J, Dufour JP. Physicochemical and quality characteristics of cold-pressed flaxseed oils. Journal of Food composition and Analysis. 2007;20(3-4):202-211.
- Timilsena, Yakindra, Vongsvivut, Jitraporn, Adhikari, Raju, Adhikari, Benu. Physicochemical and thermal characteristics of Australian chia seed oil. Food Chemistry. 2017;228. DOI:10.1016/j.foodchem.2017.02.021.
- 29. Hashempour-Baltork F, Torbati M, Azadmard-Damirchi S. Vegetable oil blending: A review of physicochemical, nutritional and health effects. Trends in

Foods Science & Technology. 2016;57:52-58.

Available:https://doi.org/10.1016/j.tifs.2016. 09.007

- 30. Xu W, Li H, Zhang Z, Li X. Determination of physicochemical properties and analysis on fatty acid composition of different perilla seed oil. Journal of the Chinese Cereals and Oils Association. 2013;28(12):106-109.
- Badhe Y. Effect of mechanical press oil extraction processing on the quality of linseed oil. Industrial Crops and Products. 2013;42:10-13.
- 32. Pan, Fengguang, Baoli Wen, Xiaoqing Wang, Xiaoxuan Ma, Jie Zhao, Chujie Liu, Yufei Xu, and Wenjun Dang. "Effect of the chemical refining process on perilla seed oil composition and oxidative stability." Journal of Food Processing and Preservation. 2019;43(9):e14094.
- Torri L, Bondioli P, Folegatti L, Rovellini P, Piochi M, Morini G. Development of Perilla seed oil and extra virgin olive oil blends for nutritional, oxidative stability and consumer acceptance improvements. Food Chemistry. 2019;286:584-591. Available:https://doi.org/10.1016/j.foodche m.2019.02.063
- 34. Jung H, Kim I, Jung S, et al. Oxidative stability of chia seed oil and flaxseed oil and impact of rosemary (*Rosmarinus* officinalis L.) and garlic (*Allium cepa* L.) extracts on the prevention of lipid oxidation. Appl Biol Chem. 2021;64:6. Available:https://doi.org/10.1186/s13765-020-00571-5
- 35. Subramanian R, Nandini KE, Sheila PM, Gopalakrishna AG, Raghavarao KSMS, Nakajima M, Kimura T, Maekawa T. Membrane processing of used frying oils. J Am Oil Chem Soc. 2000;77:323– 328
- Stier RF. 'Ensuring the health and safety of fried foods. European Journal of Lipid Science and Technology. 2013; 115(8):956–964. DOI: 10.1002/ejlt.201300180.

© 2021 Dhyani et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history: The peer review history for this paper can be accessed here: https://www.sdiarticle5.com/review-history/80480