GSC Biological and Pharmaceutical Sciences

eISSN: 2581-3250 CODEN (USA): GBPSC2

Cross Ref DOI: 10.30574/gscbps

Journal homepage: https://gsconlinepress.com/journals/gscbps/

(Research Article)

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Effect of orange peel essential oil (*Citrus sinensis* l.) on the oxidative and qualitative stability of shea butter (*Vitellaria paradoxa*) produced in northern ivory coast during storage

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GSC Biological and Pharmaceutical Sciences, 2026, 35(03), 206-215

Publication history: Received on 07 May 2026; revised on 13 June 2026; accepted on 16 June 2026

Article DOI: https://doi.org/10.30574/gscbps.2026.35.3.0223

Abstract

Aims: The oxidation of fats is a cause for concern regarding the quality of most oils, particularly shea butter (*Vitellaria paradoxa*). The objective of this study is therefore to contribute to improving the oxidative stability of shea butter (*Vitellaria paradoxa*) produced in northern Côte d'Ivoire during storage at room temperature through the use of orange peel essential oil (*Citrus sinensis* L.).

Methodology: To achieve this, the essential oil was extracted from orange peels using steam distillation. The physical and chemical properties of this essential oil were then determined. The shea butter was then treated with the orange peel essential oil before being stored. Finally, the quality parameters of the treated shea butter and the control shea butter were monitored over a four-month storage period.

Results: The results obtained show relatively high extraction yield, density, and refractive index for orange peel essential oil, at 1.2%, 0.811 ± 0.02, and 1.481 ± 0.02, respectively. Furthermore, the DPPH radical scavenging assay yielded inhibition percentages for the essential oil ranging from 9.69% to 90.27%, corresponding to concentrations between 5 and 20 µg/mL. Furthermore, the EC50 value of the essential oil, determined by linear regression, was 10.23 µg/mL, associated with an antioxidant activity index of 2.15. As for the study of the oxidative stability of shea butter treated with orange peel essential oil, the acidity of the control sample ranged from 5.078% to 6.974%, corresponding to a change in acid value from 9.778 to 13.251 mg/g over four (4) months of storage. The treated butters showed a slight variation in acidity ranging from 4.769% to 5.299%, equivalent to acid values of 9.061 to 10.068 mg/g. The peroxide value of the control sample ranged from 10.254 to 12.322 meq/kg, compared to 10.076 to 11.081 meq/kg for the treated sample. The impurity indices ranged from 0.177 to 0.202% for the control sample, compared to 0.178 to 0.198% for the treated shea butter.

Conclusion: Ultimately, the essential oil significantly reduced the oxidation rate of the treated shea butter compared to the untreated control. The acidity, acid value, peroxide value, and levels of insoluble impurities in the shea butter treated with the essential oil remained relatively low throughout the shelf life, unlike the untreated butter.

Keywords: Conservation; Essential Oil; Shea Butter (*Vitellaria Paradoxa*); Orange Peel (*Citrus Sinensis* L.); Oxidative Stability

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1. Introduction

Shea butter is the fat extracted from the kernel of the fruit of the shea tree (*Vitellaria paradoxa* C.F. Gaerth) (Kapseu *et al.*, 1999). This fat is rich in triglycerides, free fatty acids, palmitic acids, stearic acids, oleic acids, and linoleic acids, which are essential fatty acids for humans. It also contains proteins, wax esters, unsaponifiables, sterols, and tocopherols (UNCTAD, 2006). It plays a role in cosmetics (skin hydration, UV protection, etc.) and pharmaceuticals (wound healing, etc.) (Dandjouma *et al*., 2009 ; Gnangle *et al*., 2012 ; Kouyate *et al*., 2015 ; Soro *et al*., 2011). All these benefits make it a sought-after product for which demand is growing rapidly in international markets (Yé *et al*., 2007).

However, the processes used to produce shea butter cause oxidation of the final product, which reduces the quality of the shea butter. According to Kpegba *et al.* (2017), the quality of the extracted product depends on the process used to obtain it. More specifically, high extraction temperatures combined with long cooking times for shea butter led to significant oxidation and acidity in the final product (Womeni *et al*., 2006). Furthermore, the processes of auto-oxidation and photo-oxidation that occur in shea butter led to its extensive oxidation during its life cycle (Bentekaya *et al*., 2005). This oxidation limits the use of shea butter in dietary habits and in industry despite its availability and relatively low cost compared to ordinary butter used in food formulations (Government of Côte d'Ivoire, 2018; N'kalo, 2019).

Give adequate information to allow the experiment to be reproduced. Already published methods should be mentioned with references. Significant modifications of published methods and new methods should be described in detail. This section will include sub-sections. Tables & figures should be placed inside the text. Tables and figures should be presented as per their appearance in the text. It is suggested that the discussion about the tables and figures should appear in the text before the appearance of the respective tables and figures. No tables or figures should be given without discussion or reference inside the text.

However, several studies have shown that the use of essential oils derived from aromatic plants can help improve the oxidative stability of certain oils, particularly sunflower oil (Saty *et al.*, 2011). These essential oils, found in large quantities in certain parts of agricultural products such as orange peels, are very often discarded into the environment as organic or food waste. This constitutes a form of environmental pollution visible at certain points along our streets. Thus, the use of orange peel essential oil presents itself as a sustainable biological solution for controlling the oxidation of shea butter.

The objective of this study is therefore to help improve the oxidative stability of shea butter (*Vitellaria paradoxa*) produced in northern Côte d'Ivoire during storage at room temperature through the use of orange peel essential oil (*Citrus sinensis* L.).

2. Material and methods

2.1. Material

The plant material used in this study was shea almond butter (*Vitellaria paradoxa*) obtained in 2022 from northern Côte d'Ivoire (Figure 1). In addition to shea butter, sweet orange peels (*Citrus sinensis* L.) were used. These sweet oranges, grown in Côte d'Ivoire, were purchased in 2024 at physiological maturity at the fruit market in the town of Adjamé on the Banco Road (Abidjan, Côte d'Ivoire). The entire batch was then transported directly to the Laboratory of Food Biochemistry and Tropical Product Processing Technology (LBATTPT) at NANGUI ABROGOUA University for further analysis.

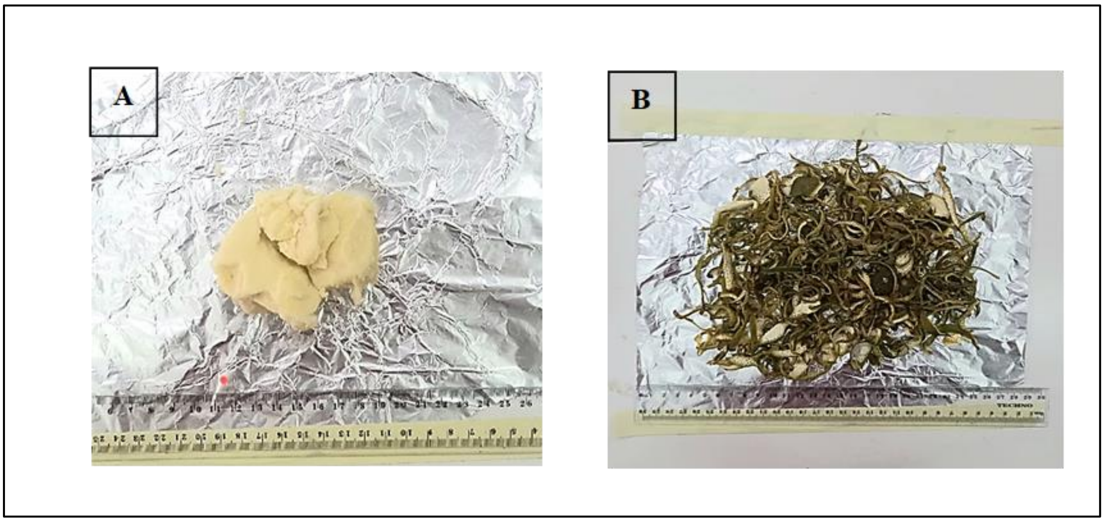

Figure 1 Photograph of shea almond butter (*Vitellaria paradoxa*) (A) and dried orange peel (*Citrus sinensis*) (B)

2.2. Methods

2.2.1. Method for producing dried orange peels

The peels were obtained according to the procedure described in Figure 2. To do this, fifty (50) oranges from each bag were sorted by stage of ripeness, washed with tap water, and peeled using a stainless-steel knife to obtain the peels. The freshly collected peels were spread out on aluminum foil and then dried at 16 °C for 48 hours. The resulting dried orange peels (Figure 2) were stored in Stomacher bags prior to any analyses.

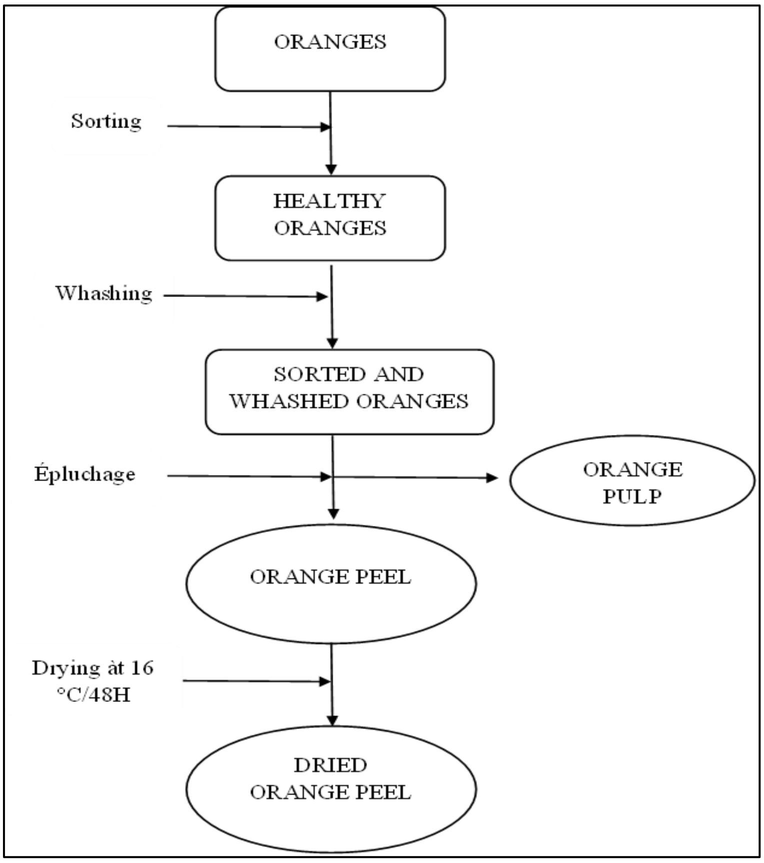

Figure 2 Flowchart for the production of dried orange peel (*Citrus sinensis* L.)

2.2.2. Method for obtaining essential oils from orange peel: Extraction by hydrodistillation

The method used to extract essential oils from *Citrus sinensis* is hydrodistillation using a Clevenger-type apparatus. Dried orange peel (*Citrus sinensis*), placed on a rack, is heated on a hotplate until the solvent boils. The water vapour carries the volatile components of the orange peel up through the cylindrical column and into the condenser, where the vapours condense and flow into the essential oil collector. The essential oil (EO) separates from the water due to a difference in density and is collected in a graduated cylinder by decantation.

2.2.3. Physical and chemical properties of orange peel essential oil

Determination of extraction yield

According to the AFNOR standard (1986), the essential oil yield (r %) is defined as the ratio of the mass of essential oils obtained after extraction to the mass of the plant material. It is given by the following formula:

$$r = \frac{m}{M} \times 100$$

r: yield (%); m (EO): mass of essential oil; (g) M: mass of plant material (g)

Determination of relative density

The relative density (d) of the essential oil was determined in accordance with French Standard NF ISO 279. One milliliter of distilled water and one milliliter of each essential oil (lemon/orange) were weighed, and the density was measured as the ratio of the mass of the distilled water to the mass of the Citrus sinensis essential oil.

$$d = m(EO)/m(water)$$

m (EO): the test sample of essential oil; m (water): the test sample of distilled water.

Determination of relative density

The refractive index of *Citrus sinensis* essential oil was determined in accordance with the French standard NF ISO 280. The refractive indices were measured using a refractometer. The refractometer’s prism was cleaned with ethanol after each measurement and calibrated with distilled water. The refractive index n, at the reference temperature t = 20 °C, is given by the following equation:

$$n = n' + 0{,}0004\ (t' - 20)$$

n’: Reading value; t’: Temperature at which the measurement was taken

Determination of the antioxidant activity of orange peel essential oil (*Citrus sinensis* L.)

The antioxidant activity of *Citrus sinensis* orange peel essential oil was determined by assessing its free radical scavenging potential, which measures the percentage of a free radical (DPPH•) neutralised by the antioxidants present in the extract. The antioxidant properties of the samples were assessed using the DPPH method (Scherer and Godoy, 2009). In the presence of free radical scavengers, the violet-coloured DPPH is reduced to yellow-coloured 2,2-diphenyl-1-picrylhydrazine. Two millilitres (2 mL) of each essential oil dilution at different concentrations ranging from two to twenty microlitres (2–20 µg/mL) were added to 1 mL of the DPPH ethanol solution (22 µg/mL). The absorbance is measured at 517 nm after 30 minutes of incubation in the dark at room temperature. For each concentration, the test is repeated three times. The results are expressed as inhibition potential (% IP).

Determination of inhibitory potential (Khoudali *et al*., 2014):

The percentage reduction in free radicals (PI %) is calculated using the following formula:

$$PR\ (\%) = \frac{(\text{Abscontrôle} - \text{Abstest}) \times 100}{\text{Abscontrôl}}$$

Abstest: sample absorbance; Abscontrol: positive control absorbance.

The curve showing the relationship between essential oil concentrations and DPPH radical inhibition potential was used to determine the reduction concentration CR50. This parameter is defined as the concentration of antioxidant required to reduce the initial DPPH concentration by 50%.

Measurement of the antioxidant activity index

The antioxidant activity index (AAI) of orange peel essential oil was calculated using the following method described by Scherer and Godoy (2009).

$$\mathrm{IAA} = \frac{[\mathrm{DPPH}]\mathrm{final}}{\mathrm{CR50}}$$

[DPPH]final = final concentration of DPPH in the reaction (22 µg/mL); CR50 = reduction concentration

2.2.4. Storage and analysis of quality parameters

The various samples of untreated butter (control) and butter treated with orange peel essential oil (*Citrus sinensis* L.) were stored in sterilised, colored glass jars, sealed and kept away from light, where they will be used for analysis from month 0 to month 4.

The analyses covered quality parameters as described in Codex Alimentarius standard CXS 325R-2017, amended in 2020.

2.2.5. Methods for deodorising shea butter

Deodorisation of shea oil

The deodorisation of the shea oil product was carried out during the cooling phase of the shea oil using orange peel essential oil. The method involved neutralizing the aromas responsible for the shea odor whilst stabilizing the quality parameters through the application of orange peel essential oils with high antioxidant potential. To this end, 100 g of liquid shea butter, heated to a temperature of 35 °C in a water bath, was taken. Next, 400 to 500 ppm (Saty *et al*., 2011) of orange peel essential oil was added, and the mixture was homogenised using a magnetic stirrer for 15 to 20 minutes. Finally, the oil was cooled whilst being stirred regularly until it solidified to form shea butter, before being packaged in sealed, light-proof glass containers.

Statistical analysis method

Statistical analysis of the data was carried out using STATISTICA version 7.1. The results are expressed as mean ± standard deviation. Significant differences were identified using one-way analysis of variance (ANOVA) followed by Duncan's test. Differences were considered significant at the 5% level.

3. Results and discussion

3.1. Physical analysis of orange peel essential oil

Table 1 presents the results of the physical analysis of orange peel essential oil. The results of the physical analysis carried out on the essential oil revealed a relatively high extraction yield of 1.2%. This yield is lower than that reported by Didi and Yakoubi (2021) during the extraction of essential oils from *Citrus sinensis* peel by hydrodistillation in Algeria; or the extraction yield of 6.55–8.61% for *Citrus sinensis* essential oil obtained by ethanol extraction in the United States, as reported by Ogo *et a*l. (2024). It remains, however, higher than the value obtained by Bousbia (2011) in the case of the extraction of orange essential oils by hydrodistillation. This author obtained a yield of 1.1% for an extraction time of 3 hours. This difference from our results can be explained, on the one hand, by the growing region, the growth stage and even the harvest period (Boutakiout *et al*., 2023); and, on the other hand, by the drying and extraction methods, which are thought to affect the extraction yield of essential oils (Verkiari *et al*., 2002). Indeed, according to Boukhatem *et al.* (2019), the quantities of essential oils produced by plants are minimal, resulting in extremely low extraction yields, generally below 2%.

Relative density is one of the physical properties used to characterise essential oils. The results showed a relative density of 0.811 ± 0.02. This density value was close to that obtained by Didi & Yakoubi (2021) during the extraction of essential oils from *Citrus sinensis* peel by hydrodistillation in Algeria, which was 0.82. This value is also below the limit of 0.925 recommended by AFNOR for the density of essential oils.

The refractive index varies primarily with the content of monoterpenes and oxygenated derivatives; a high monoterpene content will result in a high refractive index (Boukhatem et al., 2010). The refractive index of *Citrus sinensis* essential oil is relatively high, with a value of 1.471 ± 0.01. This index value was lower than the values of 1.473 and 1.472 obtained respectively by Hellal (2011) and Marouf (1990) for the same orange variety. However, it falls within the range of 1.464 to 1.474 recorded by the European Pharmacopoeia in 2008. A low refractive index of an essential oil indicates its low refraction of light, which could favour its use in cosmetic products (Kanko *et al*., 2004; Boukhatem *et al*., 2010).

Table 1 Physical characteristics of orange peel essential oil (*Citrus sinensis***)**

Variables	Value	Methods
Rendement (%)	1.2	ratio of the mass (AFNOR, 1986)
Density	0.8114	Picometre method (French standard NF ISO 279, 2000)
Refractive index	1.471	Refractometer method (French standard NF ISO 280, 2000)

3.2. Chemical analysis of orange peel essential oil

Figure 3 illustrates the antioxidant activity of orange peel essential oil. The inhibition percentages of the essential oil ranged from 9.69% to 90.27%, corresponding to concentrations between 5 and 20 µg/mL.The antioxidant concentration required to reduce the initial DPPH concentration by 50% (IC_{50}) is 10.23 µg/mL. Furthermore, the antioxidant activity index showed a relatively high value of 2.15 (Table 2). This low RC50 value, combined with a relatively high antioxidant activity index, indicates the essential oil's strong antioxidant power. According to Kholkhal (2014), the most effective DPPH radical scavengers are those with a lower RC50. Furthermore, the antioxidant activity index (AAI) is a suitable indicator for measuring an antioxidant's ability to scavenge the DPPH radical, regardless of the matrix's origin (Scherer & Godoy, 2009). Thus, with an AAI of 2.15 (AAI > 2), Citrus sinensis essential oil exhibits very high antioxidant activity. Indeed, according to Scherer & Godoy (2009), plant extracts have low antioxidant activity when the AAI is less than 0.05 (AAI < 0.05), moderate when the AAI is between 0.05 and 1 (0.05 < AAI < 1.0), high when the AAI is between 1 and 2 (1 < AAI < 2.0) and very high when the AAI is greater than 2 (AAI > 2). This suggests potential applications in the food industry. Indeed, according to Diaglia *et al*. (2008), antioxidant capacity helps to inhibit the formation of chemical toxins in cooked foods caused by the production of free radicals, thereby reducing the risk of degenerative diseases induced by these toxins. The essential oil from the peel of the orange (*Citrus sinensis*) could therefore play an important role in the preservation of foods such as shea butter, where oxidation is the primary cause of accelerated spoilage (Ouis, 2015).

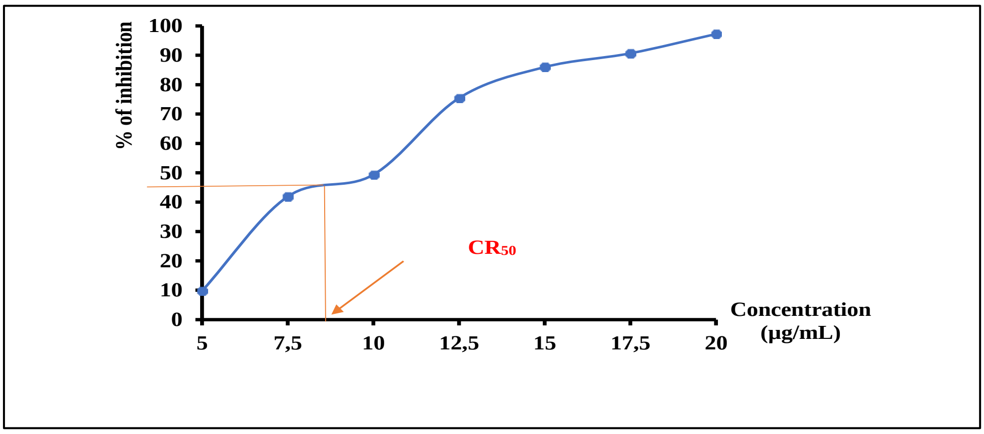

Figure 3 Percentage reduction in DPPH radical activity of orange peel (*Citrus sinensis*) essential oils

Table 2 Antioxidant activity of orange peel essential oil (*Citrus sinensis*)

Variables	Value	Methods
CR_{50} (µg/mL)	10.23	DPPH method (Scherer et Godoy, 2009)
	2.15	Calculation method (Scherer et Godoy, 2009)

3.3. Butter quality parameters during storage

Figures 4 and 5 show the changes in acidity and acid value of the control butter samples and the treated butter samples over the course of storage. Both parameters increased significantly to the 5 % threshold over time in both the control sample and the treated butter. The acidity of the control sample ranged from 5.078% to 6.974%, corresponding to a change in acid value from 9.778 to 13.251 mg/g over four (4) months of storage. In contrast, there was little variation in the acidity of the treated butter, ranging from 4.769% to 5.299%, equivalent to acid values of 9.061 to 10.068 mg/g. These acidity values remained below 5% until the second month of storage before rising to reach 5.299% by the fourth month. The rapid increase in the acid value and acidity in the sample without essential oil could be explained by its greater vulnerability to light. Indeed, according to Saty *et al*. (2011), exposure to light of sunflower oil without antioxidants accelerates acid formation in the oil. The essential oil from the peel of the orange (*Citrus sinensis*) appears to have acted as an inhibitor of this acid formation in shea butter. These results are consistent with those of Byrd (2001). Furthermore, the antioxidant properties of the essential oil are thought to have helped neutralise the fatty acids released during the extraction of shea butter, thereby reducing the acidity levels and acid values in shea butter treated with orange peel essential oil. It would therefore be of great interest in ensuring our butter complies with the quality standards set by the Codex Alimentarius. It should be noted that the Codex Alimentarius recommends acidity levels of less than 5% in free fatty acids (FFA) for acid values of less than 10 mg/g.

Furthermore, the peroxide value (Figure 6) measured during the storage of shea butter (*Vitellaria paradoxa*) increased over time, with significantly higher values recorded in untreated shea butter compared to that treated with orange peel essential oil (*Citrus sinensis*). The rapid increase in these values could be attributed to the phenomena of auto-oxidation and photo-oxidation occurring in shea butter, which lead to extensive oxidation (Bentekaya *et al*., 2005). Furthermore, Womeni (2004), in a study examining the relationship between traditional treatments and the quality of shea butter to identify the critical stages of traditional processes, demonstrated that the high peroxide values in shea butter could also be explained by the cooking process, which predisposes the shea butter to extensive oxidation (Womeni, 2004) . The antioxidant power of the essential oil from the peel of the orange (*Citrus sinensis*) is thought to have contributed significantly to delaying or even significantly preventing the oxidation of the treated shea butter. This observation was made by Saty *et al.* (2011), who described a reduction in the rate of oxidation in sunflower oils containing natural antioxidants from aromatic plants.

Finally, the impurity index values of the treated shea butter all remained below those of the control sample, with the exception of month 0. These values were low during the first two months for the treated butter. A significant change was then observed after the second month of storage. This demonstrates the ability of this essential oil to preserve the nutritional value of shea butter during the first two months following its production. Indeed, insoluble impurities are harmful oxidation products that arise in a food matrix following significant oxidation of the matrix (Billek, 2000).

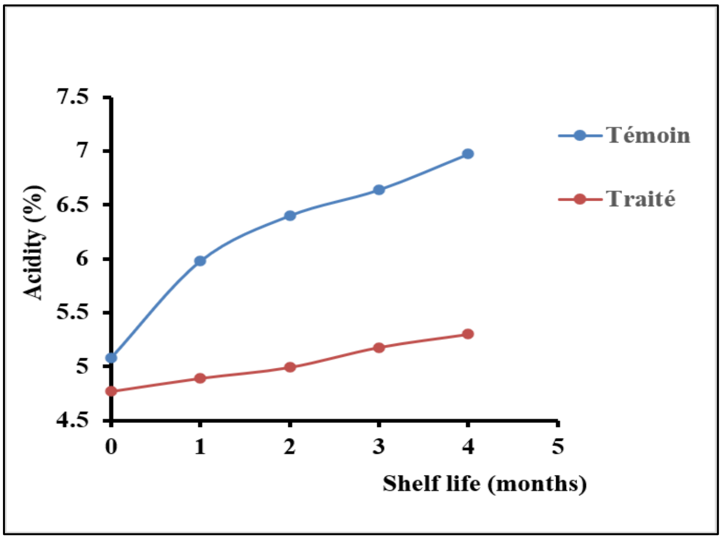

Figure 4 Changes in the acidity of shea butter samples during storage

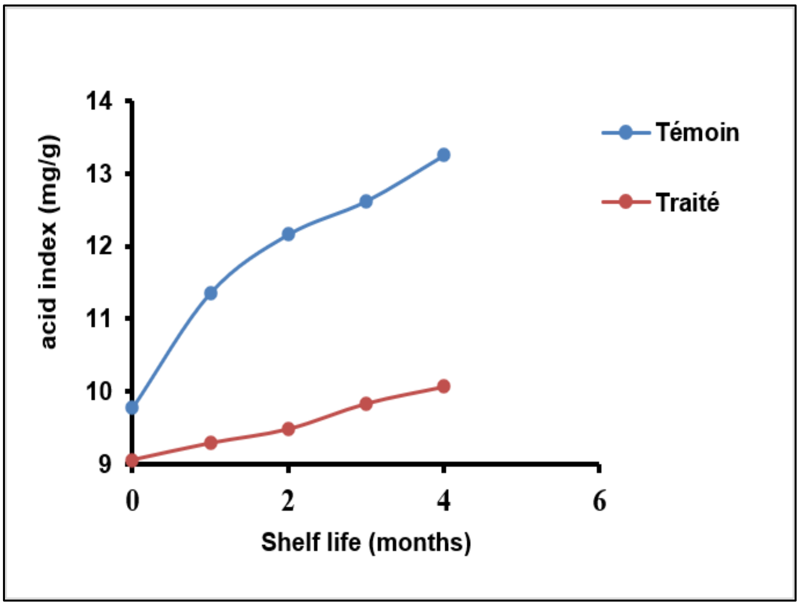

Figure 5 Changes in the acid value of shea butter samples during storage

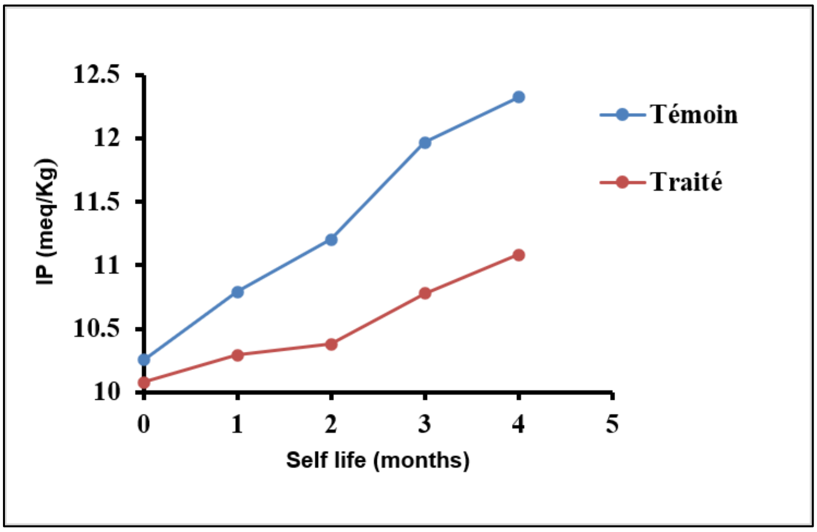

Figure 6 Changes in the peroxide value of shea butter samples over time

4. Conclusion

[The essential oil of *Citrus sinensis* orange peel, used to improve the oxidative stability of *Vitellaria paradoxa* shea butter, was extracted using the hydrodistillation method on a Clevenger-type apparatus. The extraction yield was approximately 1.2%. The density and refractive index were relatively high at 0.811 ± 0.02 and 1.471 ± 0.01, respectively. Furthermore, the essential oil of Citrus sinensis orange peel exhibited strong antioxidant activity, characterised by a low IC50 value of 10.23 µg/mL and a high antioxidant activity index of 2.15, as demonstrated by the DPPH free radical scavenging assay. Furthermore, treating shea butter with this oil during the cooling process enabled the study of its oxidative and qualitative stability during four months of storage. It appears that the essential oil facilitated a significant reduction in the oxidation rate of the treated shea butter compared to the untreated control. The acidity, acid value, peroxide value and levels of insoluble impurities in the shea butter treated with essential oil remained at relatively low levels throughout the shelf life, unlike the untreated butter. More specifically, during the first two months of the study, the values remained in line with those of the Codex Alimentarius standard. This therefore makes it feasible to use this shea butter in food formulations to enhance its value.

Compliance with ethical standards

Acknowledgments

The authors are grateful to the following units: the Laboratory of Food Biochemistry and Tropical Product Processing Technology (LBATTPT), the Laboratory of Biocatalysis and Bioprocesses (LBB), and the Laboratory of Bioorganic Chemistry and Natural Substances (LCBOSN) for making their facilities available to them.

Disclosure of conflict of interest

Authors have declared that no competing interests exist.

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