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Essential Oil of *Ocimum gratissimum* L. (Lamiaceae) as Biopreservative of Peanuts in Post-harvest: Application Model and Effects on Quality of Derived Products

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Authors' contributions

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

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ABSTRACT

The aim of this study was to investigate the use of essential oil extracted from leaves of African basil *(Ocimum gratissimum* L.) in post-harvest preservation of peanuts as well as the effects on the physicochemical, technological and organoleptic characteristics of derived peanut products. Preservation tests of peanuts with essential oil of *Ocimum gratissimum* at a concentration of 0.6 µl/g were carried out. Control was without essential oil. Evolution of the fungal flora as well as physicochemical, technological and organoleptic characteristics of preserved peanuts were evaluated. Results of microbiological analyses indicated a significant reduction ($p < 0.005$) of the microbial quantum in peanut samples preserved with essential oil, when compared to the control. The results of physicochemical analyses revealed that preservation of peanuts using essential oil of *Ocimum gratissimum* had little effect on the physicochemical characteristics. However, results from evaluation of the technological aptitudes of preserved peanuts indicated that the essential oil has modified the functional properties of preserved peanut samples, with a particular impact on the elasticity of the pastes as well as the flavor and the texture of the peanut cakes. Organoleptic analyses revealed that preservation of peanuts with the essential oil of *Ocimum gratissimum* affected the aroma and flavor of derived products.

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1. INTRODUCTION

Peanut (*Arachis hypogaea* L.) is an important oilseed worldwide [1]. From South America, the peanut plant spread to China, Africa, Indian, Japan and United States of America [2]. Peanut was one of Benin's main export products but nowadays, it is mostly grown for local consumption and as raw material for peanut oil industries [3]. In west African countries such as Senegal and Nigeria, peanut is a widely consumed food and is also an inexpensive source of protein, fats, minerals and vitamins of rural populations. It is consumed in boiled or roasted form, but also as peanut pastes or cakes [4]. However, peanuts are often contaminated by fungi, including *Aspergillus*, *Fusarium* and *Penicillium*. This contamination not only reduces its marketability but can also lead to the production of mycotoxins [5]. Thus, several studies have reported the contamination of peanut pastes and cakes by mycotoxins, with the co-contamination of aflatoxins and ochratoxin A [6,7]. A strategy to prevent peanut contamination by mycotoxins is therefore necessary because there are few decontamination processes that can eliminate the mycotoxin without denaturing the product [8]. Faced with the numerous challenges associated with the use of synthetic chemical preservatives, industrialized societies increasingly approve of the trend towards green consumption, desiring fewer synthetic additives [9]. Thus, spices and plant extracts including essential oils, as possessing many antibacterial, antifungal, and antioxidants, are increasingly used in food preservation [10]. Several studies have investigated the use of plant extracts against proliferation of fungi and mycotoxin production in peanuts and derived products. Indeed, Kasi et al. [11] reported that the phytoconstituents of ginger extract (*Zingiber officinale*) lead to a substantial reduction in aflatoxin in peanut oil, and improved its organoleptic characteristics. Moreover, Adjou et al. [12] reported that the essential oil extracted from leaves of *Ocimum gratissimum* (Lamiaceae) harvested in Benin, has *in vitro* antimicrobial properties against fungi isolated from peanuts in post-harvest. However, it is not sufficient to identify an effective antifungal agent, it must also be applicable in the food industry context. Thus, the present study aimed to evaluate conditions for the use of the essential oil of African Basil (*Ocimum gratissimum* L.) in the preservation of peanuts in post-harvest as well as its effects on the physicochemical, technological and organoleptic characteristics of derived products.

2. MATERIALS AND METHODS

2.1 Collection of Peanut Samples

A total of twenty-five (25) kilograms of shelled peanut seeds were collected at *Pahou* (South Benin). In this locality, five different collection sites were chosen and five kilograms of shelled peanuts were collected in each site. The collected peanut seed samples were transferred to sterile bags and stored at 4°C in the laboratory.

2.2 Collection of Plant Leaves

Fresh leaves of *Ocimum gratissimum* L. (10 kilograms) were collected at Abomey-calavi (south Benin) and identified at the Benin national herbarium, where a voucher specimen was deposited. Plant materials were shaded dried at laboratory until their used for essential oil extraction.

2.3 Essential Oil Extraction

The essential oil was extracted by the hydro-distillation method using a Clevenger-type apparatus. The oil recovered was dried over anhydrous sodium sulfate and stored at 4°C until required for use [13].

2.4 Experimental Preservation Model

Preservation trials of shelled peanuts with essential oil of *Ocimum gratissimum* were carried out using modified *triple bagging hermetic technology* [14]. As part of this study, the introduction into the package of blotting paper discs impregnated with the essential oil of *Ocimum gratissimum* and the reduction of the number of bags from three (3) to one (1), were the main changes made to the *triple bagging hermetic technology*. Considering previous studies, that of Adjou et al. [12], the modified packages were filled with 0.6 µL of the essential oil per gram of shelled peanuts. Control was made following the same procedure but without the essential oil. Sampling and periodic inspections were carried out to evaluate the evolution of fungi growth during storage.

2.5 Enumeration of Fungi

The enumeration of fungi was performed using a dilution plating method described by Adjou et al. [12] as follow: 10 g of each shelled peanut samples (whole seed) were added separately to 90 mL of sterile water containing 0.1% peptone water, and thoroughly mixed. Further, 10-fold serial dilutions up were made. 1 mL volume of each dilution was separately placed in Petri dishes, over which, 20 mL of Yeast Extract Sucrose (YES) agar medium was poured. The plates were incubated at 28 ± 2°C for 7 days.

2.6 Determination of Moisture Content

The moisture content of peanut samples was determined by desiccation using the method described by De Knegt and Brink [15] as follow. 5 g of peanut samples were weighed and spread on dish previously dried and weighed. The dish containing the sample was then transferred into the air oven (model: DNP-9082A, Hunan, China) at 105°C to dry until a constant weight was obtained and the loss in mass was determined.

2.7 Anti-nutritional Factors Analysis

Total oxalate of peanut samples was determined as described by Day and Underwood [16] and the oxalate content was calculated as described by Chinma and Igyor [17]. Phytate was determined using the method of Reddy and Love [18] and the phytic acid content was calculated as described by Okon and Akpanyung [19].

2.8 Processing of Preserved Peanuts

To evaluate the effect of the essential oil on the functional properties of peanuts, preserved peanuts samples were used to produce peanut pastes and cakes according to the method described by Adjou et al. [6]. During the processes, all constraints encountered were recorded.

2.9 Organoleptic Analysis

Organoleptic analyses were performed according to the hedonic test base on a 9-point linear scale [20]. These organoleptic tests concerned peanut pastes and cakes produced from preserved peanut samples, in order to assess the consumer preference on the different peanut derivative products.

2.10 Statistical Analysis

Statistical Analysis Software (SAS) and SYSTAT 5.05 were used to analyse the data generated from this study. The statistical analyses carried out were mean and standard deviation, and analysis of variance (ANOVA) [21].

3. RESULTS AND DISCUSSION

Results of the microbiological analyses of peanut samples during treatment tests showed a significant reduction ($p < 5\%$) in fungal flora of peanut samples treated with *Ocimum gratissimum* L. essential oil, when compared to the control (Table 1). After three months (90 days) of treatment with the essential oil, no fungal growth was observed in the peanut samples. However, in the control samples, a progressive growth of fungi was observed. These results show the antifungal properties of the essential oil of *Ocimum gratissimum* L. Several research studies reported that the efficacy of *Ocimum gratissimum* essential oil is due to the presence of bioactive molecules with proven antimicrobial properties such as terpenoids, which constitute a large group of broad-spectrum antimicrobial compounds [22]. However, considering that the composition and structure of foods have an effect on the different interactions that take place in food products, application in food industry of essential oils as antimicrobial agents could be affected by the intrinsic composition of food products.

Table 1. Fungal flora in peanut samples during storage

Shelf life (Days)	Controls	Samples treated with essential oil
0	$2.6.10^{1}$ ufc/g	$2.6.10^{1}$ ufc/g
30	$3.1.10^{2}$ ufc/g	4 ufc/g
60	$1.4.10^{3}$ ufc/g	1 ufc/g
90	$2.9.10^{3}$ ufc/g	No fungal growth
120	$5.4.10^{3}$ ufc/g	No fungal growth
150	$2.3.10^{5}$ ufc/g	No fungal growth

Table 2. Moisture and antinutrient contents of preserved peanut samples

Peanut samples	Moisture content (%)	Oxalate (%)	Phytate (%)
Control	8.19 ±0.01[a]	0.11 ±0.04[a]	0.67 ±0.09[a]
Treated samples	7.89 ±0.25[a]	0.20 ±0.06[a]	0.68 ±0.05[a]

Values are means (n = 3). The means followed by different superscript letters in the same column are significantly different according to ANOVA and Tukey's multiple comparison test

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Table 3. Observations made by producers during processing of treated peanut samples

Process stages	Findings	Rendering
Reception- Sorting-Roasting	Absence of major technological constraints	Treatment of peanuts with the essential oil has no effect on these stages of the process when compared to control samples
Peeling, Milling	Absence of major technological constraints	Treatment of peanuts with the essential oil has no effect on these stages of the process when compared to control samples
Mixing-Pressing	Pastes from peanut samples treated with essential oils of *Ocimum gratissimum* are less consistent than pastes from control samples.	The essential oil of *Ocimum gratissimum* modifies the functional characteristics of proteins contained in peanut pastes, in particular the structuring properties involving protein-protein type relationships.
	Peanut samples treated with essential oil requires more water than pastes from control samples	The essential oil of *Ocimum gratissimum* modifies the functional characteristics of the proteins contained in peanut pastes, in particular the properties of hydration (relations of the protein with water)
Seasoning-Homogenization-Shaping-Frying-Draining-Cooling	Absence of major technological constraints	Treatment of peanuts with the essential oil has no effect on these stages of the process when compared to control samples

The results of moisture, oxalate and phytate factors contents of treated peanut samples (Table 2) indicated that the treatment of peanuts with essential oil has a minimal effect on these parameters ($p < 5\%$). Thus, the risk of alteration of peanut seeds, in particular the loss of the germination potential (due to the reduction of water availability) of the seeds would therefore be greatly reduced.

Table 3 presents constraints encountered by producers during processing of peanut samples treated with *Ocimum gratissimum* essential oil. According to producers, pastes from peanut samples treated with essential oil are less consistent and required more water during processing than the pastes from control samples. These results indicated that the use of the essential oil of *Ocimum gratissimum* has modified the functional characteristics such as hydration properties (absorption, retention or swelling) of proteins contained in peanut pastes.

From results of the organoleptic analyses of peanut products (Figs. 1 and 2), it appeared that the preservation of peanuts using essential oil did not modified the texture and the color of different products obtained. However, there is significant difference ($p < 5\%$) in the aroma and taste of the different peanut products. These findings are in concordance with the observations made by the producers during the mixing-pressing process of the pasta obtained from peanut samples treated with essential oil (Table 3). Furthermore, results based on the preference of tasters revealed that the majority (75%) of them accepted products derived from peanut seeds treated with the essential oil due to their flavored characteristics. The constituents of this essential oil (mostly aromatic molecules), have improved the organoleptic characteristics of peanut products. Several studies have confirmed the importance of using essential oils in improving the organoleptic characteristics of food products. Indeed, Tsigarida et al. [23] reported that the addition of 0.8% v/w essential oil improves the flavor of beef meat. According to Harpaz et al. [24], essential oils of *Thyme* and *Oregano* spread throughout Asian sea bass flesh at a dose of 0.05% v/w gave it a herbal aroma. Furthermore, Santos et al. [25] reported that, above certain concentrations, some essential oils may no longer be viable for food use because they become odoriferous. According to Mejlholm and Dalgaard [26], the concentrations required for several essential oils for extending shelf-life resulted in overly strong flavors, which limited their use. It is for this reason that it is always desirable, after the determination of the Minimal Inhibitory Concentrations, to also evaluate the efficacy of the essential oil by direct application in real food systems. Indeed, the application of essential oils in real food systems as antibacterial agents, despite its many constraints, are now emerging at laboratory scale [26] and several innovative essential oil applications have been proposed to provide effective solutions for the food preservative challenges.

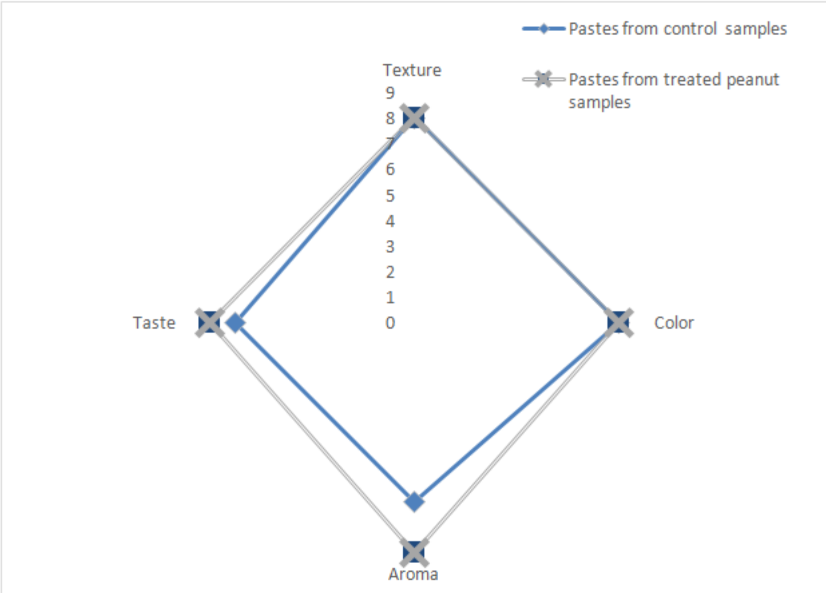

Fig. 1. Results of organoleptic analyses of pastes produced from preserved peanuts

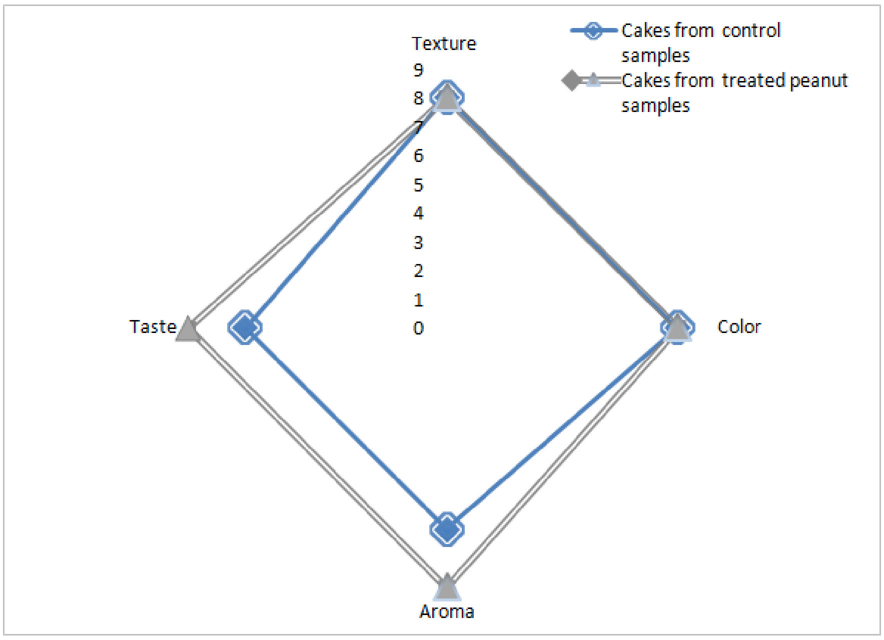

Fig. 2. Results of organoleptic analyses of cakes produced from preserved peanuts

4. CONCLUSION

The use of essential oil with techniques involving treatment and food safety has nowadays a great interest for food industry because it is not sufficient to identify a antifungal agent, but it must also be applicable in the food industry context. This study underlined the treatment potential of essential oil of *Ocimum gratissimum* in post-harvest preservation of peanuts against spoilage factors from fungi origin. *Ocimum gratissimum* has the potential to produce a biopesticides in large post-harvest systems. Future research should therefore focus on other technologically innovative applications of essential oils in food industry, using scientific knowledge-based information.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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