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Inhibitory Potential of The Essential Oils of Ginger and Thyme Against *S. aureus* Isolated From Some Meat Products

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Abstract

ABOUT 2500 grams of frozen beef kofta, sausage, burger, and luncheon samples were obtained from a supermarket in El-Bagor, Menoufia governorate, Egypt. In order to confirm sterility of the tested meat products, samples were exposed to UV radiation in a cabinet for duration of 20 minutes. Samples were promptly prepared and artificially-inoculated with *S. aureus* (10^4 cf/g). Samples were blended with the essential oils of ginger or thyme at 0.5% and 1%, respectively for 30 seconds. PBS served as the control. The samples containing oils, as well as the control samples, were placed in polyethylene bags, labelled, and stored at a temperature of 4 ºC. The inhibitory potential of the essential oils of thyme and ginger against *S. aureus* was examined. In addition, the sensory properties of the tested meat products were evaluated during cold storage at 4°C. The evaluation was conducted after 3 hours, 3rd, 6th, 9th, 12th, 15th, 18th, and 21st days. The findings indicated that the treated samples exhibited a decline in *S. aureus* counts and enhanced sensory characteristics in comparison to the untreated samples (control). Furthermore, ginger oil exhibited the most efficacies when used at a concentration of 1%.

Keywords: Essential oils, Thyme oil, Ginger oil, Meat products, *S. aureus*.

Introduction

Meat and meat products are highly appealing to humans due to their rich nutritional content, which includes protein, fat, vital amino acids, minerals, vitamins, and other essential nutrients. Due to their richness in such nutrients, several pathogens and spoilage organisms might attack such products [1]. Foodborne pathogens can remain and become more harmful when surfaces used for meat handling and processing are not adequately cleaned and disinfected [2]. Due to inadequate hygienic measures adopted during preparation of such meat products, *S. aureus* can contaminate such products [3]. Furthermore, it can be challenging to completely eradicate pathogens from food processing environments due to the ability of bacteria to adhere to the surfaces in contact with food and create biofilms, which allow them to persist even after washing and disinfection [4].

The overuse and inappropriate application of traditional antimicrobial drugs have resulted in the rise of foodborne pathogens and other multi-resistant microbes, with grave implications for human health. Essential oils may be able to address the developing issue of antimicrobial resistance due to their natural antimicrobial properties [5]. Since it is a challenge for the food industry to make food that is both safe and free of artificial chemical preservatives, since ancient times, aromatic and medicinal herbs have been extensively used in food preparation, also in recent years, their significance in bio-preservation has come to light [6].

Ginger is used as a spice in foods and drinks because of its characteristic spicy aroma and taste [7]. Ginger and garlic contain active ingredients with several health benefits, including antioxidant, anti-inflammatory, anti-diabetic, immune-modulatory, and antibacterial properties [8]. Thyme is abundant in bioactive components that can effectively suppress the growth of foodborne pathogens [9].

Staphylococcus aureus is a major foodborne pathogen that is responsible for many cases of hospitalizations worldwide and can easily contaminate meat products [9].

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In sight of the previous facts, this study aimed at investigation of the inhibitory effects of the essential oils of ginger and thyme against *S. aureus* isolated from different meat products. In addition, the effects of such essential oils to extend the shelf life of the examined meat products and to enhance the sensory attributes of the final products were also examined.

Material and Methods

Essential oils

The ready-made herbal oils of ginger (Zingiberaceae) and thyme (Thymus vulgaris) used in this study were purchased from Galhoum Co., Menoufia, Egypt.

Meat products

A total of 2500 grams of frozen beef kofta, sausage, burger, and luncheon samples were obtained from a store in El-Bagor, Menoufia governorate, Egypt. To eradicate natural bacterial populations, meat product samples were subjected to 20 minutes of UV light exposure in a cabinet to achieve total sterilization and eliminate any microorganisms that may be present on the surface.

Bacterial Strain and Culture Media

The count of Staphylococcus aureus was measured using Baird Parker agar supplied by Media Unite, Food Hygiene Department, Animal Health Research Institute, Shebin El-Kom, Menoufia, Egypt.

Preparation of sample

The samples were promptly prepared and then artificially-inoculated with S. aureus at a concentration of 104 cfu/g. Next, the inoculated samples were put in sterile plastic bags; the bags were gently compressed by hand, and then left undisturbed for 30 minutes to ensure perfect attachment between the microbe and the sample. To achieve uniform mixing, the meat product groups were blended with ginger and thyme essential oils at concentrations of 0.5% and 1% (v/g), respectively for 30 seconds. PBS served as the control. The oil and control samples were placed in polyethylene bags, labeled, and stored at a temperature of 4 ºC. During storage, we did sensory studies to evaluate color, odor, texture, and overall acceptability. Additionally, we used the serial dilutions and spread plate technique [10, 11] to determine the S. aureus count. These analyses were performed after 3 hours, as well as on the 3rd, 6th, 9th, 12th, 15th, 18th, and 21st days.

Statistical analysis

In this research, all experiments were conducted with three replicates. The statistical data analysis was conducted using SPSS V21.0 software (SPSS, Inc., Chicago, IL, USA). An analysis of variance (ANOVA) with repeated measures was conducted to assess the significant differences in sensory, chemical, and microbiological evaluations. The significance level was set at $P < 0.05$.

Results and Discussion

The recorded results in Table (1) show that the counts (log cfu/g) of *S. aureus* in the control group of kofta samples were 5.13 ± 0.04 after 3 hours,, 5.72 ± 0.09, 5.95 ± 0.14, and 6.01 ± 0.11 after three, six, and nine days, Subsequently, the sample became spoiled. The samples exposed to 0.5% ginger oil showed *S. aureus* counts of 5.04 ± 0.03, 4.44 ± 0.04, 3.06 ± 0.02, 2.82 ± 0.03, 2.11 ± 0.06, and 1.95 ± 0.04 after three hours, three days, six days, nine days, twelve days, fifteen days, and eighteen days after inoculation. The *S. aureus* counts (log cfu/g) were 4.80 ± 0.02, 4.12 ± 0.04, 2.61 ± 0.03, 2.24 ± 0.06, 1.40 ± 0.02, and ND when 1% ginger oil was used. The counts of *S. aureus* were 5.05 ± 0.01, 4.52 ± 0.02, 3.21 ± 0.05, 2.65 ± 0.02, and 2.21 ± 0.05 when 0.5% of thyme oil was used. While, the counts of *S. aureus* were 5.01 ± 0.01, 4.32 ± 0.03, 2.91 ± 0.04, 2.42 ± 0.03, and 2.09 ± 0.05 after three hours, three days, six days, nine days, and twelve days when 1% of thyme oil was used. Results shown in Table (2) showed that when 0.5% ginger oil was used, the counts of *S. aureus* in sausage samples were 4.81 ± 0.06, 3.56 ± 0.04, 3.01 ± 0.03, 2.82 ± 0.05, 2.27 ± 0.05, and 1.94 ± 0.03 log cfu/g after 3 hours, 3rd day, 6th day, 9th day, 12th day, and 15th day of inoculation, respectively. After using 1% of ginger oil, *S. aureus* counts were 4.64 ± 0.04, 3.28 ± 0.03, 2.85 ± 0.08, 2.02 ± 0.06 log cfu/g after 3 hours, 3rd day, 6th day, 9th day, and were not detected after that, respectively. In case of using 1% of thyme oil, *S. aureus* counts were 4.72 ± 0.03, 3.45 ± 0.06, 2.91 ± 0.04, 2.25 ± 0.06, 1.94 ± 0.07, 1.42 ± 0.02 log cfu/g after 3hrs, 3rd day, 6th day, 9th day, 12th day, 15th day, and not detected after that. The results recorded in Table (3) revealed that in the case of using1% of ginger oil in the contaminated burger samples, *S. aureus* counts were 4.98 ± 0.02, 3.65 ± 0.03, 2.44 ± 0.04, 2.22 ± 0.02, 2.01 ± 0.05 after 3 hours, 3rd day, 6th day, 9th day, 12th day, respectively, and then not detected. While when using 1% thyme such counts were 5.04 ± 0.04, 3.92 ± 0.02, 2.58 ± 0.03, 2.30 ± 0.02, 2.08 ± 0.02, respectively. The results recorded in Table (4) revealed that in the case of using1% of ginger oil in the contaminated luncheon samples, *S. aureus* counts were 4.91 ± 0.02, 3.07 ± 0.03, 1.82 ± 0.02, 1.31 ± 0.03, after 3 hours, 3rd day, 6th day, 9th day, respectively, and then not detected. While when using 1% thyme such counts were 4.93 ± 0.02, 3.35 ± 0.03, 2.09 ± 0.04, and 1.61 ± 0.03, respectively. The obtained results of the present study were in

agreement with previous reports [1, 12-15]. Thyme is frequently employed in culinary applications primarily because of its distinctive taste and fragrance. Thymol, a compound present in thyme, has been used in mouthwash products for more than a hundred years. In addition, it exhibits activity against *S. aureus, Escherichia coli,* and the microorganisms responsible for deterioration in meat products [16]. The ginger essential oil exhibited the highest efficacy in reducing the count of *S. aureus* [17]. The findings demonstrate that the essential oils under investigation, namely ginger and thyme at a concentration of 1% have noteworthy antibacterial effects against *S. aureus*. Over time, their antimicrobial action intensifies. Thus, these crucial oils can be chosen as prospective food bio-preservatives and anti-*S. aureus* agents in meat products. Essential oils possess antibacterial qualities that result in the deterioration of bacterial membranes, leading to the release of internal cell components and finally causing the death of the cells. Typically, Gram-negative bacteria have a lower susceptibility to antimicrobials. However, it should be noted that Gram-positive bacteria are not necessarily more sensitive to these substances [18, 19]. The inherent characteristics of food, such as its fat, protein, and water composition, together with its antioxidants, preservatives, pH level, salt content, and other additions, can all influence the efficacy of plant essential oils in eradicating germs. However, temperature, packaging environment, and bacterial features can all influence their sensitivity. Moreover, there is a widespread belief that diets that are rich in fat and/or protein provide protection to bacteria from the impacts of essential oils in some manner [20, 21].

Results in Table (5) showed that in the case of using ginger oil at concentrations of 1%, the scores of sensory attribute evaluation for kofta were 9, 8.5, 8, 7.5, 7, 6.5, 5 & 4 after 3hrs, 3rd, 6th, 9th, 12th, 15th, 18th, and 21st day of the storage period, respectively. But in the case of using thyme oil at the concentration of 1%, the scores were 9, 8.5, 8, 7.5, 6, 5.5, 5, and 3.5 after the same storage periods, respectively.

Results in Table (6) showed that in the case of using ginger oil at concentrations of 1%, the scores of sensory attribute evaluation for sausage were 9, 8.5, 8, 7.5, 7, 6.5, 5 & 4 after 3hrs, 3rd, 6th, 9th, 12th, 15th, 18th, and 21st day of the storage period, respectively. But in the case of using thyme oil at the concentration of 1%, the scores were 9, 8.5, 8, 7.5, 6, 5.5, 4.5, and 3 after the same storage periods, respectively.

Results in Table (7) showed that in the case of using ginger oil at concentrations of 1%, the scores of sensory attribute evaluation for burger were 9, 8.5, 8, 7.5, 7, 6.5, 5, and 4 after 3hrs, 3rd, 6th, 9th, 12th, 15th, 18th, and 21st day of the storage period, respectively. But in the case of using thyme oil at the concentration of 1%, the scores were 9, 8.5, 8, 7, 6, 5.5, 5, and 3.5 after the same storage periods, respectively.

Results in Table (8) showed that in the case of using ginger oil at concentrations of 1%, the scores of sensory attribute evaluation for luncheon were 9, 8.5, 8, 7.5, 7, 6.5, 5.5, and 4 after 3hrs, 3rd, 6th, 9th, 12th, 15th, 18th, and 21st day of the storage period, respectively. But in the case of using thyme oil at the concentration of 1%, the scores were 9, 8.5, 8, 7.5, 7, 6.5, 5.5, and 4 after the same storage periods, respectively. These results were in agreement with previous reports [13, 18, 22]. In recent times, plant-based compounds have emerged as natural preservatives capable of prolonging the shelf life of numerous food products [23]. Thyme (Thymus vulgaris L.) has been found to be a valuable source of bioactive compounds, such as essential oil (EO) and thymol that have the ability to hinder the growth and dissemination of harmful microbes [24]. The results indicate that the use of both 0.5% and 1% concentrations of ginger oil and thyme oil have a significant and satisfying impact on the sensory characteristics and shelf life of the tested meat product. The sensory attribute scores rise in correlation with the escalating oil concentration, hence enhancing the shelf life of these beef items.

Conclusion

The study's findings indicated that the ginger and thyme oils have antimicrobial properties that can be employed as natural meat preservatives against food-borne pathogens. As a result, they may be helpful in the meat industry to improve meat safety and shelf life by reducing the growth of bacteria that can cause food poisoning. Thus, it is advised to use natural preservatives like ginger and thyme oils in place of chemical ones.

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Conflicts of interest

The authors declared no competing interests.

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Authors' contribution:

All authors contributed equally to the present study.

TABLE 1. Influence of different concentrations of natural oils (ginger and thyme) against *S. aureus* artificially inoculated in kofta samples.

Groups/ Storage period	Control	Ginger 0.5%	Ginger 1%	Thyme 0.5%	Thyme 1%
Zero day	5.13 ± 0.04[a]	5.04 ± 0.03[a]	4.80 ± 0.02[b]	5.05 ± 0.01[a]	5.01 ± 0.01[a]
3rd day	5.72 ± 0.09[a]	4.44 ± 0.04[b]	4.12 ± 0.04[c]	4.52 ± 0.02[b]	4.32 ± 0.03[bc]
6th day	5.95 ± 0.14[a]	3.06 ± 0.02[b]	2.61 ± 0.03[c]	3.21 ± 0.05[d]	2.91 ± 0.04[b]
9th day	6.01 ± 0.11[a]	2.82 ± 0.03[b]	2.24 ± 0.06[c]	2.65 ± 0.02[d]	2.42 ± 0.03[e]
12th day	S	2.11 ± 0.06[a]	1.40 ± 0.02[b]	2.21 ± 0.05[a]	2.09 ± 0.05[a]
15th day	S	1.95 ± 0.04[a]	ND*	2.07 ± 0.04[a]	1.41 ± 0.02[c]
18th day	S	S	ND*	S	1.22 ± 0.04[a]
21th day	S	S	S	S	S

Different superscripted letters are significantly different at $p \leq 0.05$.
*S= Spoiled.
**ND= Not detected.

TABLE 2. Influence of different concentrations of natural oils (ginger and thyme) against *S. aureus* artificially inoculated in sausage samples.

Groups/ Storage period	Control	Ginger 0.5%	Ginger 1%	Thyme 0.5%	Thyme 1%
Zero day	5.22 ± 0.11[a]	4.81 ± 0.06[a]	4.64 ± 0.04[b]	4.86 ± 0.02[a]	4.72 ± 0.03[a b]
3rd day	6.21 ± 0.12[a]	3.56 ± 0.04[b]	3.28 ± 0.03[c]	3.63 ± 0.07[b]	3.45 ± 0.06[b]
6th day	6.63 ± 0.13[a]	3.01 ± 0.03[b]	2.85 ± 0.08[c]	3.11 ± 0.05[b]	2.91 ± 0.04[b]
9th day	7.11 ± 0.11[a]	2.82 ± 0.05[b]	2.02 ± 0.06[c]	2.89± 0.04[b]	2.25 ± 0.06[b]
12th day	S	2.27 ± 0.05[a]	ND*	2.51 ± 0.03[c]	1.94 ± 0.07[d]
15th day	S	1.94 ± 0.03[a]	ND*	2.01 ± 0.05[a]	1.42 ± 0.02[c]
18th day	S	S	ND*	S	ND*
21th day	S	S	S	S	S

TABLE 3. Influence of different concentrations of natural oils (ginger and thyme) against *S. aureus* artificially inoculated in burger samples.

Groups/ Storage period	Control	Ginger 0.5%	Ginger 1%	Thyme 0.5%	Thyme 1%
zero day	5.11 ± 0.03[a]	5.04 ± 0.03[a]	4.98 ± 0.02[a]	5.06 ± 0.06[a]	5.04 ± 0.04[a]
3rd day	5.56 ± 0.14[a]	3.98 ± 0.02[b]	3.65 ± 0.03[c]	3.99 ± 0.05[b]	3.92 ± 0.02[b]
6th day	6.22 ± 0.11[a]	2.61 ± 0.03[b]	2.44 ± 0.04[c]	2.75 ± 0.03[b]	2.58 ± 0.03[b]
9th day	6.98 ± 0.11[a]	2.44 ± 0.05[b]	2.22 ± 0.02[c]	2.68± 0.02[d]	2.30 ± 0.02[c]
12th day	S	2.12 ± 0.04[a]	2.01 ± 0.05[b]	2.47 ± 0.05[c]	2.08 ± 0.02[b]
15th day	S	1.77 ± 0.02[a]	ND*	2.09 ± 0.03[c]	1.62 ± 0.01[b]
18th day	S	S	ND*	S	ND*
21th day	S	S	S	S	S

TABLE 4. Influence of different concentrations of natural oils (ginger and thyme) against *S. aureus* artificially inoculated in luncheon samples.

Groups/ Storage period	Control	Ginger 0.5%	Ginger 1%	Thyme 0.5%	Thyme 1%
Zero day	5.01 ± 0.09[a]	4.95 ± 0.03[a]	4.91 ± 0.02[a]	4.95 ± 0.04[a]	4.93 ± 0.02[a]
3rd day	5.45 ± 0.08[a]	3.64 ± 0.05[b]	3.07 ± 0.03[c]	3.70 ± 0.04[b]	3.35 ± 0.03[d]
6th day	6.11 ± 0.09[a]	2.23 ± 0.02[b]	1.82 ± 0.02[c]	2.43 ± 0.05[d]	2.09 ± 0.04[b]
9th day	6.58 ± 0.07[a]	1.97 ± 0.03[b]	1.31 ± 0.03[c]	2.05± 0.04[b]	1.61 ± 0.03[d]
12th day	7.20 ± 0.08[a]	1.62 ± 0.04[b]	ND*	1.64 ± 0.03[b]	ND*
15th day	S	ND*	ND*	ND*	ND*
18th day	S	ND*	ND*	ND*	ND*
21th day	S	S	S	S	S

TABLE 5. Influence of different concentrations of natural oils (ginger and thyme) on sensory attributes of kofta samples during cold storage.

Groups/ Storage period	Control	Ginger 0.5%	Ginger 1%	Thyme 0.5%	Thyme 1%
Zero day	9	9	9	9	9
3rd day	7.5	8	8.5	8	8.5
6th day	5.5	7	8	7	8
9th day	4.5	6	7.5	6	7.5
12th day	3	5	7	5.5	6
15th day	2.5	4.5	6.5	4.5	5.5
18th day	2	3.5	5	3.5	5
21th day	1	2.5	4	2	3.5

Excellent: 9 –Very very good: 8 - Very good: 7 - Good: 6 -Medium: 5 -Fair: 4 - Poor: 3 -Very poor:2 - Very very poor: 1.

TABLE 6. Influence of different concentrations of natural oils (ginger and thyme) on sensory attributes of sausage samples during cold storage.

Groups/ Storage period	Control	Ginger 0.5%	Ginger 1%	Thyme 0.5%	Thyme 1%
Zero day	9	9	9	9	9
3rd day	7.5	8.5	8.5	8	8.5
6th day	5.5	7	8	7.5	8
9th day	4.5	6.5	7.5	6	7.5
12th day	3.5	5	7	5.5	6
5th day	2.5	4.5	6.5	4.5	5.5
18th day	2	3	5	3	4.5
21th day	1.5	2	4	2	3

Excellent: 9 -Very very good: 8 - Very good: 7 - Good: 6 -Medium: 5 -Fair: 4 - Poor: 3 Very poor: 2- Very very poor: 1.

TABLE 7. Influence of different concentrations of natural oils (ginger and thyme) on sensory attributes of burger samples during cold storage.

Groups/ Storage period	Control	Ginger 0.5%	Ginger 1%	Thyme 0.5%	Thyme 1%
Zero day	9	9	9	9	9
3rd day	7.5	8	8.5	8	8.5
6th day	5.5	7	8	7	8
9th day	3	6	7.5	6	7
12th day	2.5	5.5	7	5	6
15th day	2	4.5	6.5	4.5	5.5
18th day	1.5	3.5	5	3	5
21th day	1	2.5	4	2	3.5

Excellent: 9 -Very very good: 8 - Very good: 7 - Good: 6 -Medium: 5 -Fair: 4 - Poor: 3 Very poor: 2- Very very poor: 1.

TABLE 8. Influence of different concentrations of natural oils (ginger and thyme) on sensory attributes of luncheon samples during cold storage.

Groups/ Storage period	Control	Ginger 0.5%	Ginger 1%	Thyme 0.5%	Thyme 1%
Zero day	9	9	9	9	9
3rd day	7.5	8	8.5	8	8.5
6th day	6	7.5	8	7.5	8
9th day	5	7	7.5	7	7.5
12th day	4.5	6.5	7	6.5	7
15th day	3	6	6.5	6	6.5
18th day	2	5	5.5	5	5.5
21th day	1	3	4	3	4

Excellent: 9 -Very very good: 8 - Very good: 7 - Good: 6 -Medium: 5 -Fair: 4 - Poor: 3 Very poor: 2- Very very poor: 1.

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النشاط المقاوم للبكتريا للزيوت الأساسية للزنجبيل والزعتر ضد المكورات العنقودية الذهبية المعزولة من بعض منتجات اللحوم

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الملخص

تم تجميع2500 جرام من عينات كفتة اللحم البقري المجمدة، والنقانق، والبرغر، واللانشون من سوبر ماركت في الباجور، محافظة المنوفية، مصر. لتأكيد تعقيم منتجات اللحوم المختبرة، تم تعريض العينات للأشعة فوق البنفسجية في دقيقة. تم تحضير العينات بسرعة وتم تلويثها صناعيًا بـ المكور العنقودي الذهبي .تم خلط العينات مع الزيوت الأساسية للزنجبيل أو الزعتر بنسبة 0.5% و1%، على التوالي، لمدة 30 ثانية. تم وضع العينات التي تحتوي على الزيوت، بالإضافة إلى العينات الضابطة، في أكياس بولي إيثيلين، وتم وضع علامات عليها، وتخزينها عند درجة حرارة 4 تم فحص القدرة التثبيطية للزيوت الأساسية من الزعتر والزنجبيل ضد المكورات العنقودية الذهبية. بالإضافة إلى ذلك، تم تقييم الخصائص الحسية لمنتجات اللحوم المختبرة أثناء التخزين البارد عند 4 درجات مئوية. تم إجراء التقييم بعد 3 ساعات، في اليوم الثالث، السادس، التاسع، الثاني عشر، الخامس عشر، الثامن عشر، والحادي والعشرين. أشارت النتائج إلى أن العينات المعالجة أظهرت انخفاضًا في عدد مستعمرات المكورات العنقودية الذهبية وخصائص حسية محسنة مقارنة بالعينات غير المعالجة .علاوة على ذلك، أظهر زيت الزنجبيل أعلى فعالية عند استخدامه بتركيز 1.%

الكلمات الدالة: الشوارد الحرة، الزيوت الاساسية، الزنجبيل ، الزعتر، المكورات العنقودية الذهبية>