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Effect of the Inhibitory Activity of Tarragon Extract on Microorganisms in Preserving Laboratory-Manufactured Tomato Paste

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ABSTRACT

The growing consumer demand for natural food preservatives as alternatives to synthetic additives has intensified research into plant-derived antimicrobial agents. This study investigated the efficacy of tarragon (Artemisia dracunculus L.) oil extract in extending the shelf life of tomato paste and its antimicrobial properties against common foodborne pathogens. Oil and alcoholic extracts of tarragon were prepared using a Soxhlet apparatus and rotary evaporation. Their antimicrobial activity was evaluated against Staphylococcus aureus, Escherichia coli, Candida spp., and Aspergillus niger using the agar well diffusion method. Subsequently, laboratory-manufactured tomato paste was treated with tarragon oil extract at concentrations of 25%, 50%, and 100% and stored at 4°C for 1, 4, 8, and 14 days. Microbial quality (total aerobic bacterial count and yeast/mold count) and sensory attributes were assessed periodically. Results demonstrated that the oil extract exhibited significant dose-dependent antimicrobial activity, with the 100% concentration showing the highest inhibition zones, particularly against fungi. In contrast, the alcoholic extract showed no inhibitory effect. In tomato paste, all extract concentrations effectively suppressed microbial growth compared to the control, with higher concentrations (50% and 100%) demonstrating superior preservation over the 14-day storage period. Sensory evaluation indicated that while the control sample scored highest, all treated samples remained within an acceptable "Good" to "Very Good" range, with the 25% concentration best balancing preservative efficacy and sensory quality. The findings of this study indicate that tarragon oil extract is a promising natural preservative capable of enhancing the microbial stability of tomato paste while maintaining acceptable sensory characteristics.

1. INTRODUCTION

Globally, there is increasing interest in the use of plants and herbs due to their content of bioactive compounds, including essential oils, alkaloids, phenols, and aldehydes. The antimicrobial activity of several herbal extracts has been addressed in numerous studies [1, 2]. Antimicrobial properties have led to the use of various plants in food preservation [3]. The use of natural additives to protect food has become more common as consumers become increasingly aware of the potential risks associated with synthetic chemical preservatives. Preserving food can be achieved by using the essential oils of plants as a source of natural antimicrobial agents. Essential plant oils have been employed to combat bacteria and fungi that cause diseases and are transmitted through food, and some exhibit potent antimicrobial activity **[4].** Food spoilage factors can be categorized as external factors, such as bacteria and fungi, and internal factors, such as enzymatic reactions and associated chemical changes [5].

Tomatoes and their products rank among the most widely cultivated crops globally [6]. They play a vital role in various culinary applications, enhancing dishes such as salads, pasta, ketchup, sauces, and juices. Rich in essential nutrients, tomatoes are an excellent source of vitamin C, fiber, and lycopene. Additionally, they supply essential nutrients including copper, iron, magnesium, manganese, niacin, pantothenic acid, thiamine, and vitamin K, while being fat-free **[7].** This study investigated the effect of tarragon oil extract on the shelf life of tomato paste.*Artemisia dracunculus* L. is the scientific name for tarragon, a perennial herb belonging to the **Asteraceae** family. The plant **has** long **,** narrow leaves and **attains** a height ranging from 0.3 to **1** meter**.** French and Russian are the two main varieties of this plant **[8].** Worldwide, there are between 200 and 400 species within this genus. Tarragon thrives in warm, sunny climates [9].

Tarragon is commonly used in foods to add aroma and taste because of its abundance of aromatic components. Tarragon consists of 24% protein, 45% carbohydrates, 7% fat, and 7% fiber. It also contains numerous minerals and modest amounts of vitamin A and several B vitamins [10].

Among the essential oil components, terpenes account for 24.3% and sesquiterpenoids account for 0.2%. Coumarin is also present in tarragon at concentrations exceeding 1.0%. The plant contains flavonoids at concentrations between 0.5% and 1.9%. Studies have demonstrated the presence of phenolic acids, alkaloids, and peroxidase compounds in tarragon extract [11].

Tarragon exhibits antiparasitic, antifungal, soothing, antitussive, immunomodulatory, and antitumor properties, with a long history of use in the food, cosmetic, and pharmaceutical industries. It also possesses anti-inflammatory and chemopreventive effects against cancer and carcinogens. The presence of gallic acid **[12]** contributes to its efficacy as a preservative that prevents food spoilage. Tarragon components also function as appetite stimulants, antiseptics, and vasodilators **[13].** Therefore, this study aimed to:

1. Evaluate the antimicrobial activity of alcoholic and oil extracts of tarragon against selected microorganisms.
2. Investigate the efficacy of tarragon oil extract in extending the shelf life of tomato paste during storage periods of 1, 4, 8, and 14 days.

2. Materials and Methodologies

2-1 Sample collection:

The Attarin market in Baghdad supplied the resources that made it possible to purchase the tarragon plant.

2-2 Preparation of the alcoholic extract:

The extraction process involved filling a thimble with 50 g of tarragon powder and using a Soxhlet apparatus. The extraction was performed at 80°C using 250 ml of 80% ethanol. The extract was concentrated using a rotary evaporator at 45°C [14]. Following drying in an oven at 37°C for 10 hours, the extract was stored under refrigeration until further use [14].

2-3 Preparation of the oil extract:

The oil extract of the tarragon plant was obtained by following the same extraction method described in Section 2.2, except that hexane was used instead of ethanol. After evaporation using a rotary evaporator, the oil was stored until use.

2-4 Cultivation media:

2-4-1 Sterilization:

Glassware and instruments were sterilized using dry heat in an electric oven at 180°C for 4 hours, followed by direct flame exposure with a Bunsen burner. Wet sterilization using an autoclave was employed for sterilizing solutions and culture media at 121°C and 15 psi for 15 minutes **[15].**

2-4-2 Nutrient Broth

The medium was prepared to activate the test bacteria for use in examining the inhibitory effectiveness of microorganisms, and the medium was prepared according to the instructions of the supplying company by dissolving 25 grams in 1 liter of distilled water.

2-4-3 Nutrient agar Medium

The supplier's instructions were followed to prepare the medium for calculating the total count of bacteria. A liter of distilled water was used to dissolve 28 grams in 1 liter of distilled water.

2-4-4 Potato Doxtrose Agar

The supplier's instructions were followed to prepare the medium for calculating the total count of yeasts and molds.

2.5 Antimicrobial Activity of Tarragon Extracts Against Test Bacteria and Molds

The agar well diffusion method was employed following [16] with modifications. Test bacteria were inoculated into nutrient agar medium. Following dispersion, 0.1 ml of bacterial suspension at a concentration of 1.5×10^8 cells/ml (standardized against McFarland solution) was added. Wells of 5 mm diameter were created in the culture medium using a sterile cork borer. Alcoholic and oil extracts of tarragon were prepared at concentrations of 50% and 100%. A volume of 0.1 ml of each extract was introduced into each well; a control well containing diluted ethanol was included. Plates were left at room temperature for 20 minutes to facilitate diffusion, followed by incubation at 37°C for 24 hours. Subsequently, the diameter of the inhibition zones surrounding each well was measured.

For antifungal activity against molds, the same procedure was followed using potato dextrose agar (PDA) inoculated with mold colonies at a concentration of 4×10^8 cells/ml. Plates were incubated for 3–5 days at 25°C. All tests were performed in duplicate.

2.6 Laboratory Production of Tomato Paste

Tomato paste was prepared using oven drying. The oven was preheated to 200°C. Tomatoes were washed with cold water, dried thoroughly using paper towels, and cut into quarters. The cut tomatoes were placed on a tray with space between pieces and placed in the oven until well cooked and reduced in volume by approximately one-third. Cooking time was 2–3 hours, after which the tomatoes were left for 8 hours to complete drying; the duration depended on the moisture content. The dried tomatoes were stored in a refrigerator at 4°C [17].

Table (1): The coding of additive coefficients and concentrations.

Production	Transactions	Additional material
Tomato Paste	A	Control treatment without any addition
	B	Oil extract concentration 25%
	C	Oil extract concentration 50%
	D	Oil extract concentration 100%

Following production, tomato paste samples were treated with oil extract according to the concentrations shown in Table (1). The experimental procedure was as follows:

1. **Microbial tests were conducted on Day 1.**
2. **Tomato paste samples were stored under refrigeration at 4°C.**
3. **Microbial tests were repeated at 4, 8, and 14 days.**

2.7 Microbiological Tests

Microbiological tests were performed on tomato paste samples treated with tarragon oil extract at concentrations of 25%, 50%, and 100%. Tests included total aerobic bacterial count and yeast and mold count. A 10 g sample of tomato paste was accurately weighed and diluted in 90 ml of 0.85% physiological saline solution in a 200 ml glass beaker, yielding a 10^{-1} (1:10) dilution. Culturing and microbial enumeration were performed for each sample. All tests were performed in duplicate.

2.8 Sensory Evaluation

Sensory evaluation of tomato paste was conducted following the method described by [7]. The product was assessed by 10 trained panelists at the Food Contamination Research Center, Department of Environment and Water, Ministry of Health. Evaluation was based on a 9-point hedonic scale, where 9 = Excellent, 8 = High Very Good, 7 = Very Good, 6 = High Good, 5 = Good, 4 = High Mid, 3 = Mid, 2 = Acceptable, and 1 = Weak. Attributes evaluated included color, taste, flavor, texture, appearance, and overall acceptability.

Feature Transaction	Color	Taste	Flavor	Tissue	Appearance	General Acceptance
A						
B						
C						
D						

 Evaluation grades: 9 = Excellent, 8 = High Very Good, 7 = Very Good, 6 = High Good, 5 = Good, 4 = High Mid, 3 = Mid, 2 = Acceptable, 1 = weak.

2-9 Statistical analysis:

Data were analyzed to evaluate the effects of each treatment on the studied traits using the Statistical Analysis System (SAS) [18]. Significant differences between means were determined using the least significant difference (LSD) test at $p \leq 0.05$.

3-Results and discussion

3-1Testing the effectiveness of tarragon extracts:

The study evaluated the antimicrobial activity of tarragon oil and alcoholic extracts against *Escherichia coli*, *Staphylococcus aureus*, and *Candida* spp.

3.1.1 Evaluation of the Antimicrobial Activity of Oil and Alcoholic Extracts of Tarragon Against Selected Microorganisms

The antimicrobial activity of tarragon oil and alcoholic extracts was evaluated at concentrations of 50% and 100% against the selected bacterial and fungal isolates, as shown in Table 2. Statistically significant differences ($p < 0.05$) were observed in the inhibitory effects of the oil extract. At a concentration of 50%, the oil extract exhibited inhibition zones of 10 mm against *Staphylococcus aureus*, 14 mm against *Escherichia coli*, and 16 mm against *Candida* spp. **A**t a concentration of 100%, the oil extract produced inhibition zones of 12 mm against *Staphylococcus aureus*, 17 mm against *Escherichia coli*, and 24 mm against *Candida* spp.

In contrast, the alcoholic extract at concentrations of 50% and 100% did not show any inhibitory activity against the tested microorganisms, indicating its ineffectiveness**.** This finding aligns with the study of [19], which reported that tarragon essential oil exhibits significant antimicrobial efficacy against *Escherichia coli* and *Staphylococcus aureus* strains. The antimicrobial mechanism of the extract is attributed to its capacity to disrupt the structural and functional integrity of the microbial cell membrane. Specifically, the oil enhances membrane permeability, facilitating uncontrolled diffusion across the lipid bilayer and ultimately leading to cell death.

The inhibitory effect of tarragon essential oil on microbial growth is primarily mediated by its hydrophobic nature, which enables efficient penetration into the cell membrane's lipid bilayer. This interaction increases membrane permeability, often resulting in the leakage of intracellular components and subsequent cell lysis. Furthermore, the essential oil may interfere with bacterial enzymatic processes, contributing to its antimicrobial activity. Chemically, essential oils are complex mixtures typically comprising 20–60 bioactive compounds at varying concentrations, which collectively enhance their antimicrobial properties.

Supporting these findings, [9] demonstrated that tarragon oil extract effectively inhibits

the growth of *Staphylococcus aureus* and *Candida* species, further underscoring its broad-spectrum antimicrobial potential.

Table (2): Inhibitory Effects of Tarragon Plant Oil and Alcoholic Extract on Selected Microorganisms

Microbiology Condenser		*Candidiasis*	*Escherichia coli*	*Staphylococcus aureus*
Extraction	**%**	**Damping diameter (mm)**		
Oil extract	**50**	**16 ±0.68**	**14 ±0.72**	**10 ±0.46**
	100	**24 ±1.08**	**17 ±0.86**	**12 ±0.61**
Alcoholic extract	**50**	**0 ±0**	**0 ±0**	**0 ±0**
	100	**0 ±0**	**0 ±0**	**0 ±0**
LSD*		**3.95 ***	**3.07 ***	**2.69 ***

*Values represent means of two replicates ± standard deviation. Different superscript ers within the same column indicate significant differences ($p \leq 0.05$).

3.1.2 Antifungal Activity

Against *Aspergillus niger*

The oil extract demonstrated inhibitory effects against Aspergillus niger, as shown in Table 3. At concentrations of 50% and 100%, the oil extract significantly reduced colony diameter compared to the control. The control treatment exhibited a colony diameter of 34 mm, while colony diameters of 15 mm and 8 mm were observed at 50% and 100% concentrations, respectively, with significant differences at $p < 0.05$. A major component of tarragon essential oil exhibits a wide range of antibacterial and antifungal activities contributing to its antifungal properties [20, 19].

In contrast, the alcoholic extract showed no inhibitory effect against *Aspergillus niger*. The control treatment for the alcoholic extract had a colony diameter of 21 mm, with no reduction observed at either 50% or 100% concentrations, which also measured 21 mm.

Table (3): Inhibitory activity of tarragon oil and alcoholic extract against *Aspergillus niger*, quantified by colony diameter (mm).

Type of extract	%	*Aspergillus Niger*
Oil Extract	Control	**34**
	50	**15**
	100	**8**
Alcoholic Extract	Control	**21**
	50	**21**
	100	**21**
LSD	**6.017 ***	

*Values represent means of two replicates ± standard deviation. Different superscript letters indicate significant differences ($p \leq 0.05$).

3.2 Total Aerobic Bacterial Count in Tomato Paste Samples

Table 4 presents the total aerobic bacterial counts in tomato paste samples treated with tarragon oil extract at concentrations of 25%, 50%, and 100%, stored at 4°C for 1, 4, 8, and 14 days. No bacterial growth was observed in any treatment on day 1, with no statistically significant differences among treatments ($p < 0.05$).

At day 4, bacterial growth was observed in the 25% concentration treatment (17×10^2 CFU/g), the 50% concentration treatment (3×10^2 CFU/g), and the 100% concentration treatment (98×10^1 CFU/g). At day 8, bacterial counts increased to 19×10^2 CFU/g for the 25% treatment, 37×10^2 CFU/g for the 50% treatment, and 102×10^1 CFU/g for the 100% treatment. At day 14, bacterial counts reached 21×10^2 CFU/g for the 25% treatment, 44×10^2 CFU/g for the 50% treatment, and 116×10^1 CFU/g for the 100% treatment. Statistically significant differences ($p < 0.05$) were observed among treatments at days 4, 8, and 14.

The control treatment exhibited a progressive increase in bacterial colonies, reaching 38×10^2 CFU/g at day 4, 86×10^2 CFU/g at day 8, and 32×10^3 CFU/g at day 14. Increasing concentrations of the extract resulted in decreased bacterial counts, indicating a dose-dependent antimicrobial effect.

Table (4): Total count of aerobic and anaerobic bacteria in tomato paste samples to which concentrations (100, 50, 25%) of tarragon oil extract were stored (1, 4, 8, 14) days at refrigerator temperature.

Daily Storage periods / Extract concentration %	1	4	8	14	value LSD
(A) Data Control	0 ±0	$10^2 \times 38$ ±1.6	$10^2 \times 86$ ±6.28	$10^3 \times 32$ ±2.1	28.69 *
(B) 25%	0 ±0	$10^2 \times 17$ ±2.8	$10^2 \times 19$ ±1.6	$10^2 \times 21$ ±1.17	15.07 *
(C) 50%	0 ±0	$10^2 \times 3$ ±0.47	$10^2 \times 37$ ±1.42	$10^2 \times 44$ ±2.87	29.36 *
(D) 100%	0 ±0	10×98 ±6.7	10×102 ±6.09	10×116 ±7.02	14.88 *
Value LSD	0.00 NS	35.74 *	22.61 *	51.86 *	---

* ($P \leq 0.05$).

CFU = Colony for Unit

3-1-3 Number of molds in tomato paste samples:

Table (5) shows the inhibitory effect of tarragon extract at different concentrations compared to the control treatment, where no numbers of yeasts and molds appeared during (1) day for all treatments, while a gradual increase in the growth of molds and yeasts appeared in the control treatment, which reached (29, 73, 620) cells/mm during the storage period (4, 8, 14) days, respectively.

While the treatment with 25% concentration showed growth of yeast and mold numbers (18,

16, 20) cells/mm in storage periods (4, 8, 14) days respectively, and the concentration of 50% during storage periods (4, 8, 14) days, the number of yeast and mold reached (11, 12, 15) cells/mm, while the concentration of 100% for the same storage periods, the number of yeast and mold reached (7, 9, 9) cells/mm.

From the results of the statistical analysis indicated in the table, we can conclude that there are significant differences between the various treatments at the significance level ($p<0.05$), as

3.3 Yeast and Mold Count in Tomato Paste Samples

Table 5 presents the yeast and mold counts in tomato paste samples treated with tarragon oil extract at concentrations of 25%, 50%, and 100%, stored at 4°C for 1, 4, 8, and 14 days. No yeast or mold growth was observed in any treatment on day 1.

A gradual increase in yeast and mold growth was observed in the control treatment, reaching 29 CFU/g at day 4, 173 CFU/g at day 8, and 620 CFU/g at day 14.

The 25% concentration treatment exhibited yeast and mold counts of 18 CFU/g at day 4, 16 CFU/g at day 8, and 20 CFU/g at day 14. The 50% concentration treatment showed

treatments to which oil extract was added outperformed the control treatment, during the various storage periods, and specifically, the greater the concentration of the extract, the greater its role in preserving the tomato paste during storage.

Tarragon demonstrates notable antibacterial and antifungal properties, exhibiting greater bactericidal efficacy against Gram-positive bacteria and fungi compared to Gram-negative bacteria [21].

counts of 11 CFU/g at day 4, 12 CFU/g at day 8, and 15 CFU/g at day 14. The 100% concentration treatment demonstrated the lowest counts, with 7 CFU/g at day 4, 9 CFU/g at day 8, and 9 CFU/g at day 14.

Statistically significant differences ($p < 0.05$) were observed among treatments at days 4, 8, and 14. Treatments containing oil extract outperformed the control treatment across all storage periods, with higher extract concentrations correlating with greater preservation efficacy.

Tarragon demonstrates notable antibacterial and antifungal properties, exhibiting greater bactericidal efficacy against Gram-positive bacteria and fungi compared to Gram-negative bacteria [21].

Table (5): total count of molds in tomato pastes which concentrations of tarragon oil extract (100, 50, 25%) were stored (1, 4, 8, 14) days at refrigerator temperature.

Daily Storage periods

Extract concentration %	1	4	8	14	Value LSD
(A) Control sample	0 ±0	29 ±1.07	173 ±8.02	620 ±32.5	37.08 *
(B) 25%	0 ±0	18 ±0.74	16 ±0.81	20 ±1.06	13.95 *
(C) 50%	0 ±0	11 ±0.52	12 ±0.67	15 ±0.75	9.63 *
(D) 100%	0 ±0	7 ±0.32	9 ±0.46	9 ±0.46	7.31 *
Value LSD	47.02 *	31.66 *	18.53 *	0.00 NS	---

* (P≤0.05).

*Values represent means of two replicates ± standard deviation. Different superscript letters within the same column indicate significant differences ($p \leq 0.05$).

CFU: Colony-forming units; NS: Non-significant.

3.4 Sensory Evaluation of Tomato Paste

Table 6 summarizes the results of the sensory evaluation conducted on the laboratory-produced tomato paste treated with three concentrations of tarragon oil extract (25%, 50%, and 100%) and a control treatment. Attributes evaluated included color, taste, flavor, texture, appearance, and overall acceptability. Significant differences ($p < 0.05$) were observed among treatments for all attributes.

The control treatment received the highest scores across all attributes, followed by the 25% concentration treatment, then the 50% concentration treatment. The 100% concentration treatment received the lowest sensory scores; however, all scores remained within the "Good" to "Very Good" range, indicating consumer acceptability of tarragon addition to food products.

In the food industry, sensory evaluation is a valuable tool for assessing consumer acceptance of food. The study of [22] suggests that tarragon's aromatic nature makes it an effective flavor enhancer.

Table (6): Sensory evaluation of laboratory tomato paste

Feature Treatment	Color	Taste	Flavor	Tissue	Appearance	General Acceptance
A	±0.548.9	±0.548.9	±0.488.6	±0.558.9	±0.598.9	±0.628.8
B	±0.628.8	±0.378.2	±0.447.9	±0.378.2	±0.468.1	±0.588.7
C	±0.387.5	±0.417.9	±0.356.8	±0.437.5	±0.297.2	±0.568.3
D	6.8 ±0.41	7.1 ±0.38	6.5 ±0.26	7.3 ±0.39	6.9 ±0.34	7.2 ±0.25
LSD	1.27 *	1.34 *	1.19 *	1.25 *	1.22 *	1.37 *

*Values represent means of 10 panelist scores ± standard deviation. Different superscript letters within the same column indicate significant differences ($p \leq 0.05$).

Evaluation scale**:** 9 = Excellent, 8 = High Very Good, 7 = Very Good, 6 = High Good, 5 = Good, 4 = High Mid, 3 = Mid, 2 = Acceptable, 1 = Weak.

4.Conclusions

This study demonstrates that tarragon (*Artemisia dracunculus* L.) oil extract is an effective natural preservative for tomato paste. The oil extract exhibited significant, concentration-dependent antimicrobial activity against *Staphylococcus aureus*, *Escherichia coli*, *Candida* spp., and *Aspergillus niger*, whereas the alcoholic extract showed no inhibitory effects. The antimicrobial efficacy was particularly pronounced against fungi, with the 100% oil extract concentration producing inhibition zones of 24 mm against *Candida* spp. and reducing *Aspergillus niger* colony diameter from 34 mm (control) to 8 mm. When incorporated into tomato paste at concentrations of 25%, 50%, and 100%, the oil extract significantly reduced total aerobic bacterial counts and yeast/mold counts over a 14-day refrigerated storage period compared to the control. The 100% concentration exhibited the strongest antimicrobial effect, with bacterial counts of 116×10^1 CFU/g and yeast/mold counts of 9 CFU/g after 14 days,

compared to 32 × 10^3 CFU/g and 620 CFU/g, respectively, in the control. Sensory evaluation revealed that although the control sample received the highest scores, all tarragon-treated samples remained within an acceptable sensory quality range ("Good" to "Very Good"). The 25% concentration offered the optimal balance between antimicrobial protection and sensory acceptability, achieving overall acceptability scores of 8.7 out of 9.0 after 14 days of storage. In conclusion, tarragon oil extract represents a promising natural alternative to synthetic preservatives for enhancing the microbial safety and shelf life of tomato paste and other food products. Further studies are recommended to investigate the stability of the active compounds during extended storage and to evaluate the efficacy of this extract in other food matrices.

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