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The Effect of Hot Aqueous Extract of Salvadora persica Roots on Microbial Load and Sensory Properties of Minced Meatballs

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Abstract: This study aimed to investigate the effect of the hot aqueous extract of Salvadora persica (miswak) roots on the preservation of minced meatballs. The research was conducted to evaluate the impact of adding the plant extract on improving the quality and sensory properties of refrigerated meatballs stored at 4°C. Additionally, the study examined the effect of the extract on microbial tests. Four different treatments were included: TC (control), T1 (0.05% hot aqueous root extract), T2 (0.1% extract), and T3 (0.15% extract). The treatments were stored for 1, 3, 5, 7, 9, and 11 days. The addition of Salvadora persica root extract to minced meat showed a noticeable effect in all treated samples, contributing to a reduction in total bacterial counts, which reached 8.5, 6.5, 6.3, and 6.4 log cycles, respectively. Psychrophilic bacterial counts were 7.9, 5.3, 5.1, and 5.2 log cycles, respectively. Furthermore, the extract improved the sensory properties of the refrigerated meat compared to the control (TC). The treatments with Salvadora persica root extract (T1, T2, T3) at concentrations of 0.05%, 0.1%, and 0.15% showed less deterioration compared to the control sample (TC).

INTRODUCTION

Miswak is a twig or root derived from a small shrub known as Salvadora persica, used for both health and social purposes (1). The use of miswak has been associated with a reduced need for periodontal treatment, as its sticks possess mechanical and chemical properties that combat microbial inflammation, tooth erosion, and gingival bleeding (2). Extracts of miswak have been employed in dental treatments and mouthwashes to control microbial infections and promote periodontal health (3).

Meat is considered a highly nutritious food for humans due to its sensory attributes and its content of essential nutrients, including high-quality proteins, essential fatty acids, minerals, and vitamins—particularly vitamins B6 and B12 (4). However, processed meat products are known for their high content of fat, saturated fatty acids, cholesterol, and minerals (5). Consequently, processed meats have received significant attention due to their association with various health conditions such as obesity, diabetes, cardiovascular diseases, and several types of cancer

MATERIALS AND METHODS

A. Preparation of Minced Meatballs

Beef meat (2 kg) was obtained and minced using an electric meat grinder. The meat was mixed with the hot aqueous extract of miswak (Salvadora persica) roots and divided into four treatments. The first treatment (TC) served as the control with no extract added. The second, third, and fourth treatments (T1, T2, T3) were prepared by adding the root extract at concentrations of 0.05, 0.1, and 0.15 mg per 100 g of beef, respectively. The meatballs were formed, placed in sterilized dishes, and stored under refrigeration at $4\pm1^{\circ}$ C (8).

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B. Microbiological Tests

a. Total Bacterial Count

The total bacterial count was performed according to the method described by (9). Petri dishes were incubated at 37°C for 24 hours, and bacterial colonies were then counted using a colony count collector.

b. Psychrophilic Bacterial Count

This test followed the procedure described by (10). Nutrient Agar was used as the culture medium, and the plates were incubated at 4°C to determine the psychrophilic bacterial count.

4. Sensory Evaluation

Sensory evaluation was conducted for all treatments over the storage periods (1, 3, 5, 7, 9, and 11 days) by specialists in food science. The sensory attributes evaluated included color, odor, texture, and overall acceptability, following the sensory evaluation form described by (11).

RESULTS AND DISCUSSION

A. Microbiological Tests

1. Total Bacterial Count Estimation

Table (2) presents the results of the total bacterial count for minced meatball samples stored at a refrigerated temperature of 4°C over a storage period of 11 days. The results showed that meatballs treated with different concentrations (0.05%, 0.1%, and 0.15%) of the aqueous root extract of Salvadora persica exhibited a strong antimicrobial effect compared to the control treatment (TC). The table also indicates an increase in bacterial counts with longer refrigerated storage durations. However, samples treated with the extract displayed significantly lower bacterial counts. There were statistically significant differences among the extract concentrations as well as among the different storage periods.

The results demonstrated that the addition of Salvadora persica root extract led to a reduction in total bacterial counts throughout the storage periods. On the first day of production, the bacterial count for the control (TC) was 4.7 log cycles, increasing to 8.5 log cycles by the final storage day. For treatment T1 (0.05% extract), the count started at 3.6 log cycles and rose to 6.5 log cycles. Treatment T2 (0.1%) started at 3.4 log cycles and ended at 6.3 log cycles, while T3 (0.15%) started at 3.5 and reached 6.44 log cycles by day 11. Statistically, treatment T3 showed the lowest average bacterial count of 4.8 log cycles, compared to the control which averaged 6.7 log cycles.

Across the storage periods, the lowest log value was recorded on day 1 (3.7 log cycles), which gradually increased to 6.9 log cycles on the final day. These findings indicate that both extract concentration and storage duration significantly affected the total bacterial count in meatballs.

The results confirm that the addition of miswak root extract clearly reduced the microbial load in the meatball samples. Notably, treatment T2 (0.1% extract) significantly outperformed the other treatments in reducing bacterial counts, followed by T3 (0.15%). These findings suggest that Salvadora persica root extract acts as a natural antimicrobial agent, contributing to an extended shelf life of the refrigerated meatballs.

These results are in agreement with those of (12), who reported a decline in viable cell counts in treated samples during six days of refrigerated storage, with counts rising from 3.2×10^3 to 2.5×10^5

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CFU/g. Statistical analysis showed significant differences between treated and control groups across all storage periods, supporting the antimicrobial efficacy of the plant extract.

In conclusion, the addition of miswak extract helped preserve the microbial quality of minced meatballs throughout refrigerated storage, keeping bacterial counts within the acceptable limits for high-quality meat products, which are defined as not exceeding 7 log cycles (13).

Table (2): Logarithmic Total Bacterial Count of Meatball Treatments during Refrigerated Storage

Rate of Transactions	11	9	7	5	3	1	Days Treatments
6.7	8.5	7.8	7.6	6.3	5.2	4.7	T _c
5	6.5	6.3	5.1	4.7	3.8	3.6	T_1
4.9	6.3	6.2	5.1	4.6	3.6	3.4	T_2
4.8	6.4	6.3	5.1	4.7	3.5	3.5	T_3
LSD = 0.0173 Treatments					0.0291 =	Interact	ion Effect L.S.D
days LSD =0.0105	6.9	6.6	5.7	5.1	4	3.7	Interval Rate

2 - Psychrotrophic Bacterial Counts

Table (3) illustrates the effect of treatments (TC, T1, T2, T3) on the counts of Psychrotrophic bacteria in refrigerated meatballs stored at 4°C for a storage period of 11 days. The results showed significant differences among treatments. The control treatment (TC) recorded a count of 3.3 log cycles on the first day of processing, which increased to 7.9 log cycles by the end of the storage period.

Treatment T1, supplemented with Salvadora persica (miswak) extract at a concentration of 0.05%, recorded 2.5 log cycles on the first day and increased to 5.3 log cycles by the final day. Treatment T2, with 0.1% miswak extract, showed 2.4 log cycles initially and increased to 5.1 log cycles. Treatment T3, containing 0.15% miswak extract, started at 2.5 log cycles and rose to 5.2 log cycles at the end of storage.

There were also significant differences among storage intervals in terms of Psychrotrophic bacterial counts. These findings indicate that the plant extract of Salvadora persica exhibited strong inhibitory activity against Psychrotrophic bacteria and contributed to prolonging the shelf life of meatballs. The microbial activity is responsible for the spoilage of meat products; thus, the inhibitory effect of the extract helped reduce bacterial activity.

Regarding treatment averages, treatment T2 was significantly superior in recording the lowest mean log value of 3.8 log cycles compared to the control treatment TC, which recorded 5.9 log cycles. Significant differences were also observed in the mean values across storage intervals, with the lowest log count recorded on the first day of processing at 2.6 log cycles, which increased to 5.9 log cycles by the end of the storage period.

The reduction in bacterial counts in the miswak-treated meatballs may be attributed to the presence of phenolic compounds, which act as antimicrobial agents. Treatments containing the plant extract of miswak maintained the Psychrotrophic bacterial counts within the permissible limit, while the control treatment exceeded the acceptable threshold, reaching 6.9 log cycles during storage.

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According to the Iraqi Standard Specification for Quality Control (13), the permissible limit for Psychrotrophic bacteria in meat is 6 log cycles.

Table (3): Logarithmic Counts of Psychrophilic Bacteria in Meat Treatments during Refrigerated Storage

Rate of Transactions	11	9	7	5	3	1	days Treatments
5.9	7.9	7.2	6.9	5.4	4.5	3.3	Tc
3.9	5.3	4.8	4.2	3.7	3.3	2.5	T_1
3.8	5.1	4.6	4.1	3.5	3.2	2.4	T_2
3.9	5.2	4.8	4.1	3.6	3.1	2.5	T_3
L.S.D =0.018 Treatments					0.0412=	Interact	ion Effect L.S.D
L.S.D =0.021 days	5.9	5.3	4.8	4	3.6	2.6	Interval Rate

3 - Visual Evaluation of Meatballs

Table (4) presents the characteristics and visual evaluation of refrigerated meatballs stored for different periods (1, 3, 5, 7, 9, and 11 days). The samples treated with Salvadora persica (miswak) plant extract (T1, T2, T3) at concentrations of 0.05%, 0.1%, and 0.15%, respectively, were less affected compared to the control sample (TC). The control sample exhibited a rapid deterioration in quality, becoming visibly unacceptable after the fifth day of storage and unsuitable for consumption after the seventh day.

The table shows statistically significant differences among the treatments (TC, T1, T2, T3). Treatment T2, which contained 0.1% miswak extract, significantly outperformed all other treatments. It was observed that although the treated meatballs exhibited slight changes in general characteristics during refrigerated storage, they retained acceptable sensory qualities, making them acceptable to consumers up to 11 days of storage. After this period, all samples became unacceptable, except for treatment T2, which maintained its sensory attributes due to the 0.1% concentration of Salvadora persica root extract.

The results suggest that the miswak extract treatments extended the shelf life of the product up to approximately 11 days under refrigeration. This may be attributed to the active compounds in the extract that inhibit or delay lipid oxidation due to their strong antimicrobial and enzymatic activity-reducing properties. This prevents the breakdown of meatballs, which would otherwise result in the formation of undesirable compounds and odors that lower consumer acceptance (14).

The addition of Salvadora persica extract improved the sensory characteristics of the treated food product, increased solubility, reduced undesirable flavor and odor development, and mitigated oxidative processes (15). These findings align with those of reference (7), who reported that the addition of green walnut peel extract to beef sausages improved their sensory properties and extended their shelf life

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Table (4): Sensory and Visual Evaluation of Meatballs during Refrigerated Storage at 4°C

Total out of °20	Overall Acceptab °5 ility	Texture °5	Aroma °5	Color °5	Treatments	Stora ge Perio ds
18.7	4.7c	4.6 ^d	4.7°	4.7 ^{ab}	Control Treatment (TC)	
19.1	4.9a	4.7 ^b	4.8ª	4.7 ab	Treatment T1: Salvadora persica Plant Extract at 0.05% Concentration	
19.5	4.9 ^b	4.9ª	4.8ª	4.9 a	Treatment T2: Salvadora persica Plant Extract at 0.1% Concentration	1 days
18.5	4.7 ^d	4.7°	4.6 ^d	4.5 ^{bc}	Treatment T3: Salvadora persica Plant Extract at 0.15% Concentration	
14.7	3.6 ¹	3.7 ^k	3.6 ¹	3.8 ^g	Control Treatment (TC)	
17.7	4.4 ^f	4.4°	4.4 ^f	4.5 bc	Treatment T1: Salvadora persica Plant Extract at 0.05% Concentration	
18.3	4.5°	4.7°	4.5°	4.6 ^{abc}	Treatment T2: Salvadora persica Plant Extract at 0.1% Concentration	3 days
17.4	4.3 ^g	4.4°	4.3 ^g	4.4 ^{cd}	Treatment T3: Salvadora persica Plant Extract at 0.15% Concentration	
12.7	3.3 ^p	3.1 ⁿ	3.1 ^q	3.2 ^h	Control Treatment (TC)	
16.4	4.2 ^h	4 ^h	4.1 ^h	4.1 ^{ef}	Treatment T1: Salvadora persica Plant Extract at 0.05% Concentration	
16.9	4.2 ^h	4.2 ^f	4.3 ^g	4.2 ^{de}	Treatment T2: Salvadora persica Plant Extract at 0.1% Concentration	5 days
16.1	4.1 ⁱ	4.1 ^g	4.1 ⁱ	3.9 ^{fg}	Treatment T3: Salvadora persica Plant Extract at 0.15% Concentration	
10.9	3 ^r	2.7°	2.5 ^u	2.7i	Control Treatment (TC)	
14.5	3.6 ^m	3.3 ^m	3.8 ^k	3.8 ^g	Treatment T1: Salvadora persica Plant Extract at 0.05% Concentration	
14.7	3.8 ^j	3.8 ⁱ	3.9 ^k	3.2 ^h	Treatment T2: Salvadora persica Plant Extract at 0.1% Concentration	days
14.5	3.7 ^k	3.8 ^j	3.71	3.2 ^h	Treatment T3: Salvadora persica Plant Extract at 0.15% Concentration	

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	Control Treatment (TC)	2.2 ^j	2^{w}	2.1 ^p	2.4 ^u	8.7
9 days	Treatment T1: Salvadora persica Plant Extract at 0.05% Concentration	2.6 ⁱ	2.9°	2.7°	3.2 ^q	11.4
	Treatment T2: Salvadora persica Plant Extract at 0.1% Concentration	2.8i	3.3°	3.5 ¹	3.3°	12.9
	Treatment T3: Salvadora persica Plant Extract at 0.15% Concentration	2.6 ⁱ	3.2°	3.3 ^m	3.4 ⁿ	12.5
	Control Treatment (TC)	1.5 ¹	1 ^x	1.2°	1.1 ^w	4.8
11 days	Treatment T1: Salvadora persica Plant Extract at 0.05% Concentration	2.1 ^{jk}	2.1°	1.6°	2.4°	8.2
	Treatment T2: Salvadora persica Plant Extract at 0.1% Concentration	2.3 ^j	3.1 ^r	3.1 ⁿ	2.7°	11.2
	Treatment T3: Salvadora persica Plant Extract at 0.15% Concentration	1.9 ^k	2.7 ^t	2.1 ^q	2.6 ^t	9.3

Different letters within the same column indicate significant differences at the 5% level (p < 0.05)

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