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Research Article

Quality assessment of chicken meatball with different types of antioxidants in short-term preservation

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Abstract

This study evaluated the effect of natural antioxidants—lemon (*Citrus limon*) extract, tulsi (*Ocimum tenuiflorum*) extract, and moringa (*Moringa oleifera Lam.*) extract—on the quality of chicken meatballs. Samples were divided into four groups: T_0 (control), T_1 (0.5% lemon extract), T_2 (0.5% tulsi extract), and T_3 (0.5% moringa extract). Stored at 4°C for 12 days, they were analyzed at 0, 4, 8, and 12 days for sensory, proximate, physicochemical, biochemical, and microbiological properties. T_1 had the best color, flavor, and tenderness, while T_3 showed the least preferable values. Sensory attributes declined over time but remained acceptable. T_3 had the highest dry matter (DM) content, while T0 had the lowest. Crude protein (CP) was highest in T_1 and lowest in T_3. Ether extract (EE) decreased across all treatments, reaching 3.15% by day 12. Ash content increased to 1.43% but remained within acceptable limits. Cooked pH was most stable in T_1 and T_2, slightly decreasing over time due to increased acidity. Cooking loss was lowest in T_2, indicating better retention of moisture and nutrients. Thiobarbituric acid reactive substances (TBARS) values, indicating lipid oxidation, were lowest in T_1 and increased over storage. Microbial analysis showed T_0 had the highest total viable count (TVC, 5.14 log CFU/g), while T_1 had the lowest. Total coliform count (TCC) and total yeast-mold count (TYMC) were also highest in T_0, decreasing significantly in antioxidant-treated samples. Overall, T_1 (0.5% lemon extract) was the most effective in improving sensory, biochemical, and microbial quality. Therefore, lemon extract can enhance chicken meatball quality and shelf life as a natural antioxidant.

Introduction

Bangladesh is predominantly an agricultural nation, with a significant portion of its population engaged in livestock rearing. The country hosts 25.013 million cattle, 27.11 million goats, 3.90 million sheep, and 327.77 million chickens (Livestock Economy, DLS, 2024). The country's economy heavily relies on agriculture, with livestock being a major contributor among its four primary sectors: crops, livestock, fisheries, and forestry (Baset et al., 2002). Meat, which consists of the edible flesh of animals, is rich in protein, iron, zinc, fatty acids, and essential vitamins (Liza et al., 2024; Rahman et al., 2023; Sajib et al., 2023; Sagar et al., 2024). However, due to its biological composition, fresh meat is highly perishable, and contamination often occurs during slaughtering and processing (Das et al., 2022). Microbial activity can negatively impact meat quality, with lactic acid bacteria playing a significant role in spoilage (Bithi et al., 2020). Meat products generally spoil due to two main factors: microbial growth or chemical deterioration. In the case of chemical deterioration, lipid oxidation plays a significant role in the processed meat industry, as it is a primary cause of quality decline. Lipid oxidation can negatively impact sensory qualities such as color, texture, odor, and flavor, as well as reduce the nutritional value of the product. This oxidation is a complex process that occurs in aerobic environments, involving the interaction between molecular oxygen and polyunsaturated fatty acids. To combat lipid oxidation, antioxidants are commonly used. Since lipid oxidation leads to rancid flavors and odors, as well as shortened shelf life, diminished nutritional quality, and compromised safety of meat products, antioxidants have been employed for many years to prevent or slow down the oxidation process (Hashem et al., 2024; Lahucky et al., 2010). The antioxidant capacity of meat is naturally low, but it can be enhanced by adding flavonoids during processing. Synthetic antioxidants, including butylated hydroxyl anisole (BHA), butylated hydroxyl toluene (BHT), tert-butyl hydroquinone (TBHQ), and propyl gallate (PG), have long been used in meat and poultry products. However, concerns about their potential toxic effects, such as liver damage and cancer risks, have prompted the meat industry to seek natural alternatives. Natural antioxidants, particularly those from plants, are gaining popularity for their dual antioxidant and antimicrobial properties, aligning with consumer preferences for safer, more natural options. Ingredients such as fruits, spices, and herbs enhance not only the flavor and aroma of meat products but also improve stability, shelf life, and Citrus fruits, globally consumed for their nutritional and health benefits are rich in bioactive compounds such as flavonoids and vitamin C, which exhibit strong antioxidant properties by scavenging free radicals (Anagnostopoulou et al., 2006). They also contain flavonoid glycosides, coumarins, β and γ sitosterol, glycosides, and volatile oils. Additionally, citrus fruits provide essential macronutrients, including dietary fiber, sugars, potassium, calcium, folate, thiamine, niacin, vitamin B6, phosphorus, magnesium, copper, riboflavin, and pantothenic acid.

Tulsi can be applied to meat in various forms—such as extracts, essential oils, or powder—either as a marinade, coating, or spray, creating a protective barrier that helps maintain freshness and reduce microbial growth. Tulsi, known for its antioxidant and antimicrobial properties, serves as a natural and effective alternative to synthetic preservatives, enhancing the safety and shelf life of meat products (Siddiqua et al., 2018). Moringa (Moringa oleifera) is gaining attention as a natural meat preservative due to its rich antioxidant and antimicrobial properties. High levels of vitamin C, vitamin E, and flavonoids in moringa help prevent oxidation, reducing spoilage, off-flavors, and discoloration. Its antimicrobial compounds inhibit harmful bacteria, molds, and pathogens like Salmonella and Escherichia coli, enhancing meat safety (Rasak et al., 2023). Used as a coating or extract, moringa creates a protective barrier against air and moisture, extending shelf life. This makes moringa a promising, sustainable alternative to synthetic preservatives. Based on the above discussion the present study was conducted with the following objectives: i. To examine sensory, proximate, biochemical, and microbiological analysis of chicken meatballs with the addition of lemon extract, tulsi leaf extract, and moringa leaf extract at the same level and ii. To evaluate the effect of different natural antioxidants on the shelf-life and nutritional status of chicken meatball under refrigeration (4ºC temperature) storage conditions and to recommend value added chicken meatballs enriched with natural antioxidants.

Materials and Methods

Materials collection

Boneless chicken broiler meat of 2 kg obtained by slaughtering of poultry by halal method was procured from KR Market, Bangladesh Agricultural University, Mymensingh at 11.00 a.m. The meat samples were immediately transferred to the Animal Science Laboratory.

Preparation of jar and other instruments

All necessary instruments and jars or containers were cleaned with hot water and detergent powder and then dried before starting the experimental activities.

Sample preparation

About 2kg of fresh meat sample was taken for the preparation of chicken meatballs. At first, the chicken meats were properly cleaned with fresh water and all the body fat, tendons, skin and as well as separable connective tissues were trimmed off from the boneless meat with sharp knife. Then the meat was grinded properly and the spices, garam masala, salt, ice flakes, refined vegetable oil, refined corn flower, sauce were mixed with the grinded chicken meat properly as per experimental design. There were four treatment groups, i.e. T_0 = Control group, T_1 = 0.5% Lemon extract, T_2 = 0.5% Tulsi leaves extract, T_3 = 0.5% Moringa leaves extract. Then meatballs of proper shape were prepared separately. It was then boiled in hot water for 2-3 minutes. Then the water was removed from the meatballs properly and was fried in hot oil until reddish brown color obtained. Four types of meatballs were prepared properly about 20-25 gm. Size.

Different Analytical Characteristics of Chicken Meatball Samples

Sensory evaluation

Sensory evaluation of chicken meatball samples was conducted by an 8-member trained panel using a 5-point balanced semantic scale to assess color, flavor, tenderness, juiciness, and overall acceptability. Panelists, selected and trained per AMSA (1995) guidelines, evaluated samples under controlled light, temperature, and humidity conditions. Orientation sessions familiarized panelists with the intensity scale. Sensory qualities were assessed both before and after cooking, with scores ranging from 5 (excellent) to 1 (poor). Evaluations were conducted on days 0, 4, 8, and 12 of refrigerated storage at 4±1°C, with samples served in Petri dishes.

Proximate Composition

Proximate composition such as Dry Matter (DM), Ether Extract (EE), Crude Protein (CP) and Ash were carried out according to the methods (AOAC, 1995). All determination was done in triplicate and the mean value was reported.

Physicochemical properties measurement

Cooked meat pH measurement

pH value of cooked meatballs was measured using pH meter from cooked meatball homogenate. The homogenate was prepared by blending 5g of meatball with 10 ml distilled water.

Cooking loss (%)

The process of measuring cooking loss in chicken meatballs begins by weighing fresh samples. These meatballs are then boiled at 100ºC, followed by mixing with egg albumin and biscuit crumbs. They are subsequently fried in refined soybean oil at 100ºC and cooled to room temperature. Once cooled, the meatballs are covered with thin-walled plastic bags, and surface oil is removed. Cooking loss is measured at different intervals: 0 days, 4 days, 8 days, and 12 days, ensuring the preparation is consistent for accurate results.

Biochemical analysis

Thiobarbituric Acid Values (TBARS) (mg-MDA/kg)

Lipid oxidation in chicken meatballs was evaluated using the TBA method. A 5 g sample was homogenized with trichloroacetic acid, filtered, and mixed with TBA solution. After incubation at 100°C for 30 minutes, absorbance was measured at 532 nm, with results expressed as mg malonaldehyde per kg.

Microbial assessment

Chicken meatball samples (10 g) were aseptically excised, macerated using 0.1% peptone water, and homogenized in a blender to create a 1:10 dilution. Serial dilutions (10^{-2} to 10^{-6}) were prepared using a whirly mixture machine following ISO (1995) guidelines. Plate Count Agar (PCA), MacConkey Agar (MA), and Potato Dextrose Agar (PDA) were used as media for microbial analysis. The media were commercially prepared per manufacturers' instructions, and 0.1% peptone water served as the diluent. Various glassware and appliances, such as test tubes, petri dishes, conical flasks, pipettes, glass spreaders, and

equipment like whirly mixers, incubators, sterilizing instruments, and pH meters, were used for accurate microbial assessments. Media preparation involved dissolving specific amounts of PCA and MA in distilled water, boiling, and sterilizing. PDA was prepared by boiling potato slices, sieving, and adding commercial dextrose and agar before autoclaving at 121°C. Media were adjusted to pH 7.0 and kept ready for pouring at 45°C. Dilutions (0.1 ml) were spread on PCA agar in triplicates, incubated at 35°C for 24-48 hours, and colonies (30-300) were counted using a colony counter. Results were expressed as CFU/g by multiplying average colony counts by the dilution factor. Dilutions (0.1 ml) were spread on MA agar in triplicates, incubated at 35°C for 24-48 hours, and colonies (30-300) were counted. The average colony counts were multiplied by the dilution factor, and results were expressed as CFU/g. Dilutions (0.1 ml) were spread on PDA agar in triplicates, incubated at 25°C for 48-72 hours, and colonies (30-300) were counted. Results were calculated by multiplying average colony counts by the dilution factor and expressed as CFU/g.

Statistical model and analysis

The proposed model for the planned experiment was factorial experiment with two factors A (Treatments) and B (Days of Intervals) is: $yijk = \mu + Ai + Bj + (AB)ij + \varepsilon ijk \; i = 1,...,a; \; j = 1,...,b; \; k = 1,...,n$ .Data were statistically analyzed using SAS Statistical Discovery software, NC, USA. DMRT test was used to determine the significance of differences among treatments means.

Results and Discussion

Sensory Evaluation

The T1 group had the best color, while T3 had the least. Day 0 showed the most preferable color, and day 12 the least, though still consumer-accepted. Color decline at 4°C was due to lipid and pigment oxidation causing browning. The color of beef is determined by the regulation of myoglobin redox state conversion, a reversible process dependent on oxygen availability, active enzymes, and reducing compounds in the muscle (Tushar et al., 2023).

Among four treatments most preferable flavor was observed from T1 group and the lowest flavor from T0 (control group). The most preferable flavor was observed from 0 day and less preferable from 12th day but in terms of consumers view it was accepted. So, it was found that the quality was deteriorated with the increase storage period. Flavor is one of the mor causes of quality deterioration because it can negatively affect sensory attributes such as color, texture, odor and flavor as well as the nutritional quality of the product (Nunez and Boleman, 2008).

Among four treatments, most preferable tenderness was observed from T_1 group and less preferable tenderness was observed from T_0 (control group). The most preferable tenderness was observed at 0 day and less preferable tenderness at 12^{th} day but in terms of consumers view it was accepted. When meatballs were frozen, ice crystals form inside the cells of muscle tissue and puncture the cell walls. That's why meatballs leak moisture when they were cooked. Tenderness is interrelated DM content of the meatballs. With the increasing of storage period DM was increased consequently tenderness was decreased with day's intervals. The result of this experiment is related to (Lui et al., 2010) findings.

Among four treatment groups, most preferable juiciness score was observed at T_1 group and less preferable juiciness was observed at T_0 (control group). The most preferable juiciness was observed at 0 day and less preferable juiciness at 12^{th} day but in terms of consumers view it was accepted. The result of this experiment is also related to Lui et al. (2010) findings.

Among four treatment groups, most preferable overall acceptability was observed at $T_1$and T_2 group and less preferable at T_0 (control group).The range of different days of intervals of overall observation of overall acceptability score was 3.33 to 5.00. The most preferable overall acceptability was observed at 0 day and less preferable overall acceptability at 12^{th} day but in terms of consumers view it was accepted. The overall acceptability decreased during storage because of the decline in the sensory score. Similar finding was confirmed by Malav et al. (2015)

Table 1. Effect of different antioxidant on sensory parameters in chicken meatballs

Parameters	DI	Treatments					Level of significance		
		T_0	T_1	T_2	T_3	Mean	Treatment	DI	T×DI
Color	0	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00^{a}	*	**	NS
	4	5.00±0.00	5.00±0.00	5.00±0.00	4.67±0.58	4.92^{a}			
	8	4.00±0.00	4.33±0.57	4.00±0.00	3.67±0.58	4.00^{b}			
	12	3.00±0.00	3.33±0.57	3.33±0.57	3.00±0.00	3.17^{c}			
	Mean	4.25^{ab}	4.42^{a}	4.33^{ab}	4.08^{b}				
Flavor	0	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00^{a}	**	**	NS
	4	4.33±0.58	5.00±0.00	5.00±0.00	4.67±0.58	4.75^{a}			
	8	3.67±0.58	4.33±0.58	4.00±0.00	4.00±0.00	4.00^{b}			
	12	3.00±0.00	3.67±0.58	3.33±0.58	3.00±0.00	3.25^{c}			
	Mean	4.00^{c}	4.50^{a}	4.33^{ab}	4.17^{bc}				
Tenderness	0	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00^{a}	NS	**	NS
	4	4.33±0.58	4.67±0.58	4.67±0.58	4.67±0.58	4.58^{b}			
	8	3.67±0.58	4.33±0.58	4.00±0.00	3.67±0.58	3.92^{c}			
	12	3.00±1.00	3.33±0.58	3.33±0.58	3.00±0.00	3.17^{d}			
	Mean	4.00^{a}	4.33^{a}	4.25^{a}	4.08^{a}				
Juiciness	0	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00^{a}	*	**	NS
	4	4.00±0.00	4.67±0.58	4.67±0.58	4.33±0.58	4.41^{b}			
	8	3.67±0.58	4.33±0.58	4.00±0.00	3.67±0.58	3.91^{c}			
	12	3.00±1.00	3.67±0.58	3.33±0.58	3.33±0.58	3.33^{d}			
	Mean	3.92^{b}	4.42^{a}	4.25^{ab}	4.08^{ab}				
Overall acceptability	0	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00^{a}	NS	**	NS
	4	4.33±0.58	4.67±0.58	4.67±0.58	4.33±0.58	4.50^{b}			
	8	3.67±0.58	4.33±0.58	4.33±0.58	4.00±1.00	4.08^{b}			

12	3.00±0.00	3.67±0.58	3.67±0.58	3.00±1.00	3.33[c]	
Mean	4.00[a]	4.42[a]	4.42[a]	4.08[a]		

Sensory scores were 5 for excellent, 4 for very good, 3 for good, 2 for fair, and 1 for poor. Same superscripts in different treatments groups and days of interval did not differ significantly ($p>0.05$) whereas different superscripts in different treatments groups and days of interval differ significantly ($p<0.05$). T_0 = Control group, T_1 = 0.5% Lemon extract, T_2 = 0.5% Tulsi leaves extract, T_3 = 0.5% Moringa leaves extract, DI=Days of Intervals, Treat= Treatment, T*DI=Interaction of Treatment and Day Intervals.

Proximate analysis

Dry Matter

Among the four treatments, T_3 had the most preferable DM content, while T_0 (Control group) had the least. Higher DM content was less desirable, while lower DM content indicated preference. DM content increased over the storage period due to moisture loss but remained acceptable to consumers, with the best DM content observed on day 0 and the least on day 12. The DM content increased over storage time, possibly due to decreased moisture loss, as reported by Boby et al. (2021).

Crude Protein

Among the four treatments, T_1 had the highest and most preferable CP content, while T_3 had the lowest. CP content decreased with increased storage time, with the best observed on day 0 and the lowest on day 12. However, the CP content remained acceptable to consumers. Protein content decreased significantly in wheat bran (WB) and dried carrot pomace (DCP) incorporated chicken sausage (Yadav et al., 2018) which is similar to the present findings.

Ether Extract

Among the four treatments, T_1 had the most preferable EE content, while the control group had the least. EE content decreased over the storage period, reaching 3.15% across all treatments by day 12. Verma et al. (2013) observed a decrease in the fat content of mutton nuggets by the incorporation of guava powder. Ether extract content of the products showed significantly ($p<0.05$) decreasing trend with increasing levels of incorporation of pumpkin in chicken sausages reported by Zargar et al. (2014).

Ash

T_2 had the most preferable ash content, while the control group had the least. Ash content increased over storage, reaching 1.43% in all treatments by day 12 but remained acceptable to consumers. Akter et al. (2009) reported a decrease in ash content in beef, likely caused by the loss of volatile minerals during preservation. Zargar et al. (2017) reported that the ash content of the products showed significant ($P<0.05$) decreasing trend with increasing levels of incorporation of carrot in chicken sausages.

Table 2. Effect of different antioxidant on proximate components in chicken meatballs

Parameters	DI	Treatments					Level of significance		
		T_0	T_1	T_2	T_3	Mean	Treatment	DI	T×DI
DM (%)	**0**	33.60±0.60	32.39±0.87	32.23±0.68	32.54±0.82	32.69[c]	*	**	NS
	4	34.04±0.53	32.72±0.58	32.52±0.59	32.89±0.36	33.04[bc]			
	8	34.49±0.36	32.94±0.67	32.70±0.64	32.23±0.60	33.34[ab]			
	12	34.88±0.38	33.18±0.42	32.85±0.73	33.50±0.42	33.60[a]			
	Mean	34.25[a]	32.81[b]	32.58[b]	32.04[b]				
CP (%)	**0**	22.93±0.53	22.83±0.10	22.80±0.30	22.69±0.02	22.81[a]	NS	*	NS
	4	22.27±0.64	22.51±1.03	22.66±0.72	22.58±0.98	22.50[a]			
	8	21.93±0.39	22.26±0.68	21.39±0.26	21.35±0.37	21.73[b]			
	12	21.57±0.29	22.14±0.63	21.25±0.41	21.20±0.26	21.54[b]			
	Mean	22.18[a]	22.44[a]	22.08[a]	21.96[a]				
EE (%)	**0**	3.31±0.14	3.21±0.10	3.22±0.15	3.23±0.10	3.24[a]	NS	*	NS
	4	3.23±0.13	3.17±0.08	3.27±0.00	3.18±0.09	3.21[a]			
	8	3.18±0.13	3.13±0.08	3.15±0.11	3.14±0.08	3.15[ab]			
	12	3.12±0.14	3.07±0.11	3.10±0.14	3.09±0.17	3.09[b]			
	Mean	3.21[a]	3.15[a]	3.19[a]	3.16[a]				
Ash (%)	**0**	1.32±0.09	1.26±0.06	1.25±0.07	1.27±0.06	1.27[b]	*	*	NS
	4	1.38±0.09	1.30±0.03	1.28±0.08	1.31±0.05	1.32[b]			
	8	1.49±0.09	1.35±0.06	1.34±0.07	1.37±0.09	1.39[a]			
	12	1.55±0.07	1.38±0.04	1.37±0.09	1.40±0.05	1.43[a]			
	Mean	1.44[a]	1.33[b]	1.31[b]	1.34[b]				

Different superscripts in different treatments groups and days of interval differ significantly ($p<0.05$), T_0 = Control group, T_1 = 0.5% Lemon extract, T_2 = 0.5% Tulsi leaves extract, T_3 = 0.5% Moringa leaves extract, DI=Days of Intervals, Treat= Treatment, T*DI=Interaction of Treatment and Day Intervals.

Physicochemical properties

Cooked pH

T_1 and T_2 had the most preferable cooked pH, which decreased slightly over 12 days of storage due to increased acidity from free fatty acids caused by rancidity. Sharker et al. (2024) reported similar findings, attributing the pH increase in the control group to bacterial consumption of acids during protein degradation.

Cooking loss

Among these four treatments most preferable cooking loss was observed at T_2 group. The lowest amount of cooking loss indicates this product is most preferable for consumers' choices than other treatment groups. The cooking loss was decreased with the increase storage period. Cooking loss refers to the reduction in weight of meatballs during the cooking process (Jama et al., 2008) is the similar trend with

the experiment. Major components of cooking losses are thawing, dripping and evaporation. Thawing loss refers to the loss of fluid in meatballs resulting from the formation of exudates following freezing and thawing (Jama et al., 2008) is the similar report. Such losses are lower following a rapid freezing compared with slow freezing.

Table 3. Effect of different antioxidant on physicochemical properties in chicken meatballs

Parameters	DI	Treatments					Level of significance		
		T_0	T_1	T_2	T_3	Mean	Treatment	DI	T×DI
Cooking pH	0	6.12±0.01	6.14±0.01	6.14±0.01	6.13±0.01	6.13^{a}	**	**	NS
	4	6.10±0.01	6.12±0.01	6.12±0.00	6.10±0.01	6.11^{b}			
	8	6.07±0.01	6.11±0.02	6.12±0.02	6.09±0.01	6.10^{c}			
	12	6.06±0.02	6.09±0.02	6.09±0.01	6.08±0.02	6.08^{d}			
	Mean	6.09^{c}	6.12^{ab}	6.12^{a}	6.10^{b}				
Cooking loss	0	28.24±0.16	27.57±0.47	27.19±0.42	27.94±0.69	27.73^{a}	**	*	NS
	4	27.79±0.64	27.37±0.46	26.55±0.41	27.54±0.53	27.31^{a}			
	8	27.69±0.84	27.23±0.69	26.46±0.70	27.30±0.72	27.17^{a}			
	12	27.33±0.84	26.41±1.09	25.96±0.83	26.55±0.60	26.56^{b}			
	Mean	27.76^{a}	27.15^{b}	26.54^{c}	27.34^{ab}				

Same superscripts in different treatments groups and days of interval did not differ significantly ($p>0.05$) whereas different superscripts in different treatments groups and days of interval differ significantly ($p<0.05$) T_0 = Control group, T_1 = 0.5% Lemon extract, T_2 = 0.5% Tulsi leaves extract, T_3 = 0.5% Moringa leaves extract, DI=Days of Intervals, Treat= Treatment, T*DI=Interaction of Treatment and Day Intervals.

Biochemical properties

Thiobarbituric Acid Value

Among these four treatments, the most preferable TBARS value was observed from T_1. The lowest amount of TBARS value indicates the product is most preferable for consumers' health. The TBARS values increased significantly ($p<0.05$) during storage in all treatments. Similar findings were reported by Chidanandaiah et al. (2009) in meat patties during refrigerated storage.

Table 4. Effect of different antioxidant on biochemical parameters in chicken meatballs

Parameters	DI	Treatments					Level of significance		
		T_0	T_1	T_2	T_3	Mean	Treat	DI	T×DI
TBARS	0	0.150±0.020	0.113±0.025	0.130±0.010	0.127±0.015	0.130^{d}	**	**	NS
	4	0.206±0.025	0.136±0.015	0.153±0.025	0.157±0.015	0.163^{c}			
	8	0.256±0.025	0.156±0.025	0.183±0.040	0.193±0.025	0.197^{b}			
	12	0.317±0.025	0.203±0.045	0.216±0.035	0.230±0.020	0.241^{a}			
	Mean	0.232^{a}	0.152^{c}	0.171^{bc}	0.177^{b}				

Same superscripts in different treatments groups and days of interval did not differ significantly ($p>0.05$) whereas different superscripts in different treatments groups and days of interval differ significantly ($p<0.05$). T_0 = Control group, T_1 = 0.5% Lemon extract, T_2 = 0.5% Tulsi leaves extract, T_3 = 0.5% Moringa leaves extract, DI = Days of Intervals, Treat = Treatment, T*DI = Interaction of Treatment and Day Intervals.

Microbiological assessment

Total viable count

The T_0 group had the highest plate count (5.14 log CFU/g), while T_1 showed the lowest, making it the most preferable for consumers' health. TVC increased over storage time, but antioxidant compounds reduced bacterial growth by inhibiting fat deterioration and metabolism. However, a number of studies have demonstrated that compounds existing in many spices also possess antimicrobial activity (Zhang et al., 2010).

Total coliform count

The control sample had a significantly higher total coliform count (1.33 log CFU/g) compared to the treated samples. A lower TCC value is preferred for consumer health. During storage, the TCC value decreased, as antioxidant compounds helped prevent fat deterioration and bacterial metabolism. This resulted in lower bacterial growth in the chicken meatballs treated with antioxidants. Similar findings were observed by (Singh et al., 2011) of raw chicken meat emulsion incorporated with clove powder, ginger and garlic paste at refrigerated storage (4±1ºC).

Total yeast-mold count

The total yeast-mold count in the control sample (2.05 logCFU/g) was significantly higher than in the samples treated with antioxidant group among four treatments. The less amount of TYMC value indicates the product is most preferable for consumers' health. The most preferable TYMC in 0 day and minimum in 12 days. The reduced TYMC in the treated meat samples could be due to the antifungal properties of the oils (Akter et al., 2022).

Table 5. Effect of different antioxidants on different microbe's population in chicken meatballs

Parameters	DI	Treatments					Level of significance		
		T_0	T_1	T_2	T_3	Mean	Treat	DI	T×DI
TVC (logCFU/g)	0	4.63±0.05	4.55±0.05	4.59±0.10	4.58±0.09	4.59^{c}	*	**	**
	4	4.90±0.09	4.63±0.12	4.64±0.07	4.66±0.09	4.72^{c}			
	8	5.17±0.10	4.75±0.05	4.76±0.09	4.75±0.08	4.86^{b}			
	12	5.84±0.51	4.84±0.15	4.83±0.15	4.89±0.19	5.11^{a}			
	Mean	5.14^{a}	4.70^{b}	4.72^{b}	4.73^{b}				
TCC (logCFU/g)	0	1.22±0.05	1.17±0.04	1.18±0.02	1.17±0.04	1.19^{c}	*	**	NS
	4	1.28±0.05	1.20±0.05	1.22±0.06	1.21±0.04	1.23^{bc}			
	8	1.34±0.07	1.22±0.04	1.23±0.07	1.24±0.07	1.26^{ab}			
	12	1.46±0.08	1.24±0.03	1.26±0.08	1.25±0.07	1.30^{a}			
	Mean	1.33^{a}	1.21^{b}	1.22^{b}	1.22^{b}				

TYMC (logCFU/g)	**0**	1.88±0.05	1.74±0.09	1.76±0.10	1.78±0.05	1.79[a]	*	NS	**
	4	1.97±0.07	1.71±0.07	1.73±0.06	1.73±0.07	1.78[a]			
	8	2.12±0.04	1.68±0.06	1.70±0.07	1.70±0.08	1.80[a]			
	12	2.26±0.08	1.66±0.05	1.68±0.02	1.68±0.06	1.83[a]			
	Mean	2.05[a]	1.70[b]	1.72[b]	1.72[b]				

Same superscripts in different treatments groups and days of interval did not differ significantly ($p>0.05$) whereas different superscripts in different treatments groups and days of interval differ significantly ($p<0.05$) T_0 = Control group, T_1 = 0.5% Lemon extract, T_2 = 0.5% Tulsi leaves extract, T_3 = 0.5% Moringa leaves extract, DI=Days of Intervals, Treat= Treatment, T*DI=Interaction of Treatment and Day Intervals

Conclusions

The sensory quality like color, odor, juiciness, tenderness and overall acceptability were increased with different treatment levels and decreased with increase of days of intervals. DM content decreased with different treatment levels and increased with the increase of days of intervals. CP content decreased with different treatment levels. EE and Ash content decreased with different treatment levels. Cooked meat pH value and cooking loss was decreased with different treatment levels and similar trend was observed with increase days of intervals. Biochemical components TBARs value were decreased with different treatment levels but increased with the increase days of intervals. Microbial assessment like TVC, TCC and TYMC were decreased with different treatment levels. From the study it reveals that chicken meatballs can be preserved for 12 days using same levels of natural antioxidant (Lemon extract, Tulsi leaves extract, Moringa leaves extract). On the basis of sensory, physicochemical, biochemical and microbial properties indicates 0.5% Lemon extract was more acceptable and therefore it can be recommended for formulation of value-added chicken meatballs enriched with natural antioxidant.

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