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Impact of Liquid Smoke and Thyme Oil on Quality of Chicken and Turkey Chilled Meatballs

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Abstract

The purpose of this study was to see how liquid smoke made from beech sawdust and thyme essential oil (TEO) affected the stability of chicken and turkey meatballs when kept refrigerated. The study looked at three factors: chicken and turkey meatballs, liquid smoke and TEO concentrations (1 % and 0.1 %, respectively), and shelf life at refrigerated temperatures at 4◦ +1 C for (0, 3, 6, 9, 12 and 15 days). Physicochemical (moisture, crude protein, crude fat, ash, pH value, water holding capacity) and total bacterial count parameters were examined. The treatments showed a considerable favorable effect on the overall bacterial count and physicochemical parameters. During storage, the samples treated with liquid smoke, then TEO, and control lost the least amount of moisture, protein, and water holding capacity. During storage, the fat, ash, and total bacterial count increased, with the greatest levels in control and the lowest in liquid smoke, followed by TEO. The pH value has decreased during storage by addition of liquid smoke 1% compared to TEO, and control. The interaction between treatments and storage days showed significant effect ($P \leq 0.05$) on protein, fat, pH value, water holding capacity and total bacterial count. Furthermore, the lowest change was noticed in samples treated with liquid smoke during storage. Applications of liquid smoke in meatballs reduced the rate of microbial damage, maintained the physiochemical properties and were better accepted compared to TEO and control.

Keywords:
Meatballs, liquid smoke, thyme essential oil, physicochemical properties and total bacterial count.

INTRODUCTION

Poultry meat and products have become one of the most popular sources of meat protein, and they are now widely available all over the world Barbut (2002).The most prevalent poultry meat sources are chickens and turkeys (87% and 6.7% of total poultry production, respectively) (FAO, 2010). Meat of chicken and turkey are higher in protein and lower in fat than beef and other red meats. On the other hand, turkey and chicken meat products are increasing in supermarket such as tenderloins, breast, cutlets, and ground turkey (Soriano-Santos, 2010).

Meatballs are one of the most popular meat products in the world, and there are many different sorts (including chicken meat balls, beef meatballs and fish meatballs). The chicken meatballs, on the other hand, are the most popular and widely consumed. They are frequently served with noodles or vermicelli, veggies, and gravy (Indiarto, *et al.,* 2020). However, a significant amount of meat and meat products is lost or wasted each year. These annual losses amount to roughly 20% of the original meat supply (Gram *et al.*, 2002). Meat is constituted of fat, protein, minerals, carbohydrates, and water, among other things. lipids and proteins can degrade, resulting in the formation of new molecules that cause meat to change (Heinz and Hautzinger, 2007; Dave and Ghaly, 2011).

The discovery of new antioxidants from natural sources has been the focus of recent research Nunez de Gonzalez, *et al.* (2008). Food smoking has a long history of being used to preserve food. Recently, liquid smoking has become widely used in the food industry Soldera et *al.* (2008). Due to various advantages, such as more effective control of polycyclic aromatic hydrocarbon (PAH) content, liquid smoke is becoming more widely used as an acceptable replacement for traditional wood smoking Varlet *et al.* (2009). The presence of antibacterial and antioxidant chemicals such as carboxylic acids, aldehyde and phenols in food smoking provides a preservative effect (Leroi and Joffraud, 2000; Rorvik, 2000).

Essential oils (EOs) are complex natural combinations of hydrocarbons (mostly terpenoids) and oxygenated molecules (esters, alcohols, ethers, ketones, aldehydes, lactones, phenols and phenol ethers) (Zygadlo and Juliani, 2003; Pichersky *et al.*, 2006). The majority of thyme essential oils are remarkable for their high concentrations of monoterpenes, particularly the phenolic compounds thymol and its isomer carvacrol, as well as a variety of other biologically active chemicals Stahl-Biskup and Saez (2002). Thyme (*Thymus vulgaris*) essential oil is a rich source of aromatic bioactive components such as thymol and carvacrol, both of which have antioxidative and antibacterial properties (Marino *et al.,* 1999; Aljabeili, *et al.,* 2018). TEO could be added to burgers at a concentration of 0.05 % (v/w), which may be sensory acceptable. But it is not enough to significantly extend the burger’s shelf life. In order to extend the product's shelf life, both EO had to be used at higher concentrations. It's worth noting that in prior investigations on swordfish using TEO, concentrations of 0.1 % (v/w) in fresh fish were shown to be sensory acceptable (Dolea *et al.*, 2018). The objective of this work was to evaluate the effect of adding liquid smoke or TEO on the physiochemical properties and microbiological changes of chicken and turkey meatballs during chilled storage.

MATERIALS AND METHODS

Materials

36 kg of fresh breast chicken and turkey meat were purchased from the main market in Assiut city, Assiut, Egypt during September 2020. Meat of chicken and turkey were washed carefully then deboned. The breast chicken and turkey meat were minced using a meat mincer. Some samples were used directly for physiochemical analysis at zero time, while others were prepared for processing product (Can and Harun, 2015).

Beech sawdust waste

Beech sawdust waste was used in liquid smoke preparation and was obtained from a local furniture manufacturer in Assiut, Egypt.

Essential oils

Thyme (*Thymus vulgaris*) was purchased from Natural oil extraction unit, National Research Centre, Cairo, Egypt. (2020).

Spices

The spice mixture was prepared using equal weights (black pepper, sweet paprika, chili paprika, and cumin). Also, salt, rusk flour, and onions were obtained from the local market during September 2020 in Assiut city, Egypt.

Methods

Preparation of liquid smoke

The beech sawdust waste was moistened to a moisture content of about 20%. A small laboratory smoke generator was used to generate smoke, and the destructive distillation was condensed using a small condenser. The concomitent substance were removed from the obtained smoke condensates by settling about 7 days at 4˚C, followed by centrifugation at 2500 rpm for 10 min., filtration by Watman No.1 papers, and titration by carbonate solution to pH 4- 5.5. Finally it diluted with distilled water in a ratio 1:4 (condensate: distilled water) to obtain liquid smoke (Youssef *et al.*, 2015). Some modification in this method was carried out by passing of filtered liquid smoke through active charcoal filtration to obtain liquid smoke completely free from harmful substances such as benzo[a]pyrene.

Preparation of chicken and turkey meatballs samples

The ingredients for the meatballs include (minced meat of chicken and turkey 71 gm, rusk flour 10 gm, salt 2.00 gm, onion 7gm, black pepper 0.40 gm, sweet paprika 0.25 gm, chilli paprika 0.25 gm, cumin 0.10 gm, and iced water 9.00 ml). (Dinçer *et al.*, 2017).

Preparation of meatballs treated with liquid smoke or essential oil

The samples were divided into three groups; the first group was treated with 1% liquid smoke or 0.1% thyme essential oil for chicken meatballs; the second group was treated with either 1% liquid smoke or 0.1% thyme essential oil for turkey meatballs; and the third group was kept as a control group. Each sample was packed in polyethylene bags and stored under the refrigerator stored at 4±1°C for 15 days at intervals of 0, 3, 6, 9, 12 and 15 days for the refrigerator sample (Pilevar *et al.*, 2017).

Moisture content

Moisture content was determined by using the air oven drying method by AOAC Official Method 990.19 (AOAC, 2016). The moisture content was evaluated by drying 5 gm overnight at 105°C in a Model 600 air oven (Memmert, Germany). The dish was then chilled in a desiccator and weighed again and again until a steady weight was achieved.

The crude protein content:

Kjeldahl Method was used to determine protein content based on the standard procedure in AOAC Official Method 973.48 (AOAC, 2016). This method is performed based on an automated Kjeltec instrument (Foss, Germany) to determine protein content in meatball samples.

The crude fat

The Soxhlet extraction method was used to evaluate fat content as described by AOAC Official Method 960.39 (AOAC, 2016). The weight loss of the sample or the weight of the fat removed was used to determine fat content.

The ash content

As described by AOAC Official Method 999.11 (AOAC, 2016), the dry ashing method was used to determine the ash content of the samples. Ashing of meatball samples (10 g) was done in a Thermo Scientific Thermolyne 62700 muffle furnace at 550°C. The crucible was dried in an oven at 105°C for 3 hours before ashing.

The pH value

The pH of meatballs sample was measured by homogenizing 10 gm of the sample with 100 ml distilled water for 30 sec. The pH of the prepared sample was measured using a pH meter (OAKTON, pH/ mV/°C meter, USA) with a glass electrode at 20P º PC according to the method described by (Turhan *et al*., 2005).

Determination of water holding capacity (WHC)

Water holding capacity (WHC) was measured according to the methods mentioned by (Abraham and Kumar, 2000) as follows:

The minced sample (0.3g) was placed under ashless filter paper (Watman, No.41) and pressed for 10 min using a 1Kg weight. Two zones formed on the filter paper and by the planimeter their surface areas were measured. The area of the outer zone resulted from the water separated from pressed tissues thus indicating the WHC in cmP2P.

Each 1cmP2 Pof outer zone area is equivalent for 8.4 mg. free water. The expressible water % and W.H.C % were calculated as follows:-

Expressible water %= (Cm^2 x 8.4/ 0.3 x1000) x100

Water holding capacity (WHC %) = Moisture % - Expressible water%

Determination of total bacterial count in chicken and turkey meatballs

The total bacterial count in chicken and turkey meatballs were determined using the total plate count (TPC) method, according to the study Nelce *et al.(*2017), with a few modifications. Total bacterial count in chicken and turkey meatballs were determined as fellow: 1g meatballs pounded with pestle and mortar, dissolved in 9 mL of physiological sodium chloride solution (0.9 % w/v) to obtain a 10^{-1} dilution. Next, 1 mL of the sample solution is pipetted and put into a test tube containing 9 mL of physiological sodium chloride solution and so on, until a dilution of 10^{-5} is obtained. Dilution samples 10^{-3}, 10^{-4}, and 10^{-5} were pipetted 1 mL and then put into sterile Petri dishes. The petri dish is then added with 15 mL of plate count agar media (PCA). After the PCA media solidified, the Petri dish is kept upside down in an incubator at 30° C for 24 hrs.

Statistical analysis

Data were analyzed by analysis of variance (ANOVA) using a completely randomized factorial design. Basic statistics and ANOVA were performed to test the significance within replications and between treatments (SPSS, 2011). (L.S.D) tests were used to determine the differences among means at the level of 0.05%.

RESULTS AND DISCUSSION

Effect of liquid smoke and thyme essential oil on moisture content of chicken and turkey meatballs

The data in Table (1) illustrates the moisture content of chicken and turkey meatballs-controlled samples (without the addition) and with the addition of liquid smoke 1% or thyme essential oil 0.1% during storage at 4±1°C for up to 15 days.

The moisture content determination of all chicken and turkey meatballs samples showed that the heights significant difference ($P \leq 0.05$) was observed between meat types. This may be due to the change in structure of muscles. Jukna *et al.*, (2012) showed that the hardness of the meat is a good indicator of its quality. Firmness depends on the muscle tissue and its protein structure. When comparing the hardness of different species of meat, it was shown that broiler meat had the lowest firmness compared to turkey and ostrich meat, while turkey meat had the highest water holding capacity compared to chicken and ostrich meat.

Additionally, the ultimate moisture content of chicken and turkey meatballs samples treated by liquid smoke and TEO at the end of storage periods was 60.77, 61.18, and 60.95, 61.64%, respectively. These samples had a lower moisture level than the control sample during storage with a significant difference ($P \leq 0.05$). The same results were observed by Biswas *et al.* (2011) they found that gradual decrease in moisture content of duck patties with increase of storage period. Indiarto *et al.* (2020) reported that the ability of derived phenolic compounds to establish a hydrogen bond with water may impact the water-binding capacity of meat.

The overall moisture content was significantly ($P \leq 0.05$) lowest in the liquid smoked meatballs than those with TEO and control during storage periods. This change could be related to acetic acid in the liquid smoke component. The meatballs ion H^+ was bound with acetic acid. As a result, protein solubility decreased, which caused a reduction in the meatballs water content (Mahmoud, 2013; Arnim *et al.*, 2012). Liquid smoke plays a function in loosening the bound of myofibrils fiber to form area that is filled with water in a semi-free form consequently water the ability of the meat to bind water increases Abustam *et al.* (2016).

Effect of liquid smoke and thyme essential oil on protein content of chicken and turkey meatballs

Data presented in Table (2) outlines the changes in protein content during the storage period at 4˚C±1 for 15 days in different meatballs samples.

The results showed that a decrement that lowest in protein content for all samples during storage periods. Overall, treatments means showed significant difference ($P \leq 0.05$) in protein content amongst control, liquid smoke and TEO for chicken and turkey meatballs during storage periods. The interaction between treatments × storage days showed highest significant effect ($P \leq 0.05$). Data showed that protein content was reduced with a significant difference ($P<0.05$) in meatballs samples during storage periods. The highest decrease in protein content was observed in control samples during storage periods. At zero time, it was (66.07 and 63.41%) for chicken and turkey meatballs, whereas at the end of the storage period, protein content was (59.14 and 57.39%), respectively. Keryanti *et al.* (2020) found that because of the activity of bacteria that use nitrogen, the overall protein level of white tofu reduced during storage.

Moreover, it is interested to indicate that addition of liquid smoked to chicken and turkey meatballs minimized the observed decrease in protein contents than those with TEO and control sample during storage periods. At zero time, it was (65.66 and 62.22 %) for chicken and turkey meatballs treated by liquid smoked, whereas at the end of the storage period, protein content was (63.31and 60.42%), respectively with a significant difference ($P<0.05$). The minimized decrease in protein content in chicken and turkey meatballs treated by the liquid smoke during storage might be due to a slight loss of nitrogen (as volatile nitrogen) as a result of slight protein breakdown. Also, Arnim *et al.* (2012) found that liquid smoke had antibacterial activity is a factor that determines this state. Because the growth of proteolytic bacteria is inhibited by cold temperatures alone. At refrigeration temperatures, any psychrotrophic bacterium can grow. However, the impacts of liquid smoke were observed as a reduced number of microbes. According to Saloko *et al.* (2014), certain phenolic compounds can assist in the destruction of bacterial cells. Because it can non activate necessary enzymes, koagulation SH group and NH group protein. Phenol and derivatives can be bactericide (Rahayu *et al.*, 2014; Ariestya *et al.*, 2016).

Effect of liquid smoke and thyme essential oil on fat content of chicken and turkey meatballs

The total fat content of chicken and turkey meatballs samples control and treated with liquid smoke or TEO and control sample during storage at 4˚C±1 for 15 days was evaluated and the obtained results are presented in Table (3). The results showed that initial fat content was increased with significant difference ($P<0.05$) of chicken and turkey meatballs. The interaction between treatments × storage days showed highest significant effect ($P \leq 0.05$). These increases in crude fat in chicken and turkey meatballs samples during storage could be attributed to moisture and protein content decrement. These findings are similar to those reported by Ali *et al.* (2008).

The lowest increase in fat content was observed in chicken and turkey meatballs treated with liquid smoke than those with TEO and control sample during storage periods. At zero time, it was (28.30 and 32.38 %) for chicken and turkey meatballs treated by liquid smoked, whereas at the end of the storage period, fat content was (30.69 and 32.94 %), respectively. This might be due to that liquid smoke separates into three phases when condensed: an aqueous phase, an oily phase, and a water-insoluble high-density tar phase Brimer (2011). Liquid smoked products are less susceptible to rancidity. The components disseminated, rather than the dispersion phase, are attributed to the smoke's antioxidizing properties. The phenols and organic acids in liquid smoke are effective antioxidants, which retard the auto-oxidation of fats Daveidson *et al.* (2005).

Effect of liquid smoke or thyme essential oil on ash content of chicken and turkey meatballs

Ash content in the three types of meatballs samples (C, LS, and TEO) during cold storage is shown in Table (4). Initially, ash content at zero time for control, 1% liquid smoke and 0.1% TEO of chicken and turkey meatballs samples was (5.14, 5.50, 5.42%, and 5.00, 4.83, 5.18%), respectively with significant differences between control sample and treatments.

However, at the end of storage period for 15 days at 4±1°C there were increases in the final ash contents in samples treated with essential oils and the control with significant difference ($P<0.05$) of chicken and turkey meatballs. The highest significant difference ($P<0.05$) was observed between the samples and storage periods. This may be due to moisture loss. Similar effects were obtained by Abd-El-Aziz (2001), who indicated that the loss of moisture was associated with increases in other components such as fat, ash, and carbohydrates.

Table (1) Changes in moisture content of chicken and turkey meatballs treated with liquid smoke and TEO during storage at 4±1°C up to 15 days.

Var.	Treat.	Storage periods (days)						Mean
		0	3	6	9	12	15	
Chicken	C	66.04	65.31	63.91	63.48	62.38	61.83	**63.83**
	LS	65.72	64.95	64.33	62.95	62.02	60.77	**63.46**
	TEO	65.64	65.07	64.33	63.15	61.99	60.95	**63.52**
Mean		**65.80**	**65.11**	**64.19**	**63.19**	**62.13**	**61.19**	**63.60**
Turkey	C	66.26	65.69	64.60	64.04	63.34	62.82	**64.46**
	LS	65.74	65.11	64.37	63.58	61.61	61.18	**63.60**
	TEO	65.78	65.22	64.69	63.82	62.23	61.64	**63.90**
Mean		**65.93**	**65.34**	**64.55**	**63.81**	**62.40**	**61.88**	**63.99**
Main effects	C	66.15	65.50	64.25	63.76	62.86	62.33	**64.14**
	LS	65.73	65.03	64.35	63.27	61.82	60.98	**63.53**
	TEO	65.71	65.14	64.51	63.49	62.11	61.29	**63.71**
Mean		**65.86**	**65.22**	**64.37**	**63.50**	**62.26**	**61.53**	
F-test A(Var.)=**		**L.S.D 0.05**	**B (Tre.)=** 0.34	**AB=** n.s.	**C (P)=** 0.48	**AC=** n.s.	**BC=** n.s.	**ABC=** n.s.

* C: control sample, LS: liquid smoke, TEO: thyme essential oil.
F-test (A= Types meat **; B= Treatments 0.34; C= Storage periods 0.48).
L.S.D0.05 (AB= interaction between types meat x treatments n.s; AC= interaction between types meat x storage periods n.s; BC= interaction between treatments x storage periods n.s; ABC= interaction between types meat x treatments x storage periods n.s).

Table (2) Changes in crude protein content of chicken and turkey meatballs treated with liquid smoke or TEO during storage at 4±1°C up to 15 days.

Var.	Treat.	Storage periods (days)						Mean
		0	3	6	9	12	15	
Chicken	C	66.07	65.23	64.14	62.84	60.36	59.14	**62.96**
	LS	65.66	65.20	65.19	64.41	63.03	63.31	**64.47**
	TEO	66.01	65.17	64.01	62.66	61.55	60.33	**63.29**
Mean		**65.91**	**65.20**	**64.45**	**63.30**	**61.65**	**60.93**	**63.57**
Turkey	C	63.41	61.96	60.55	59.15	58.81	57.39	**60.21**
	LS	62.22	62.30	61.49	61.07	60.97	60.42	**61.41**
	TEO	62.74	61.67	60.19	59.30	58.69	58.42	**60.17**
Mean		**62.79**	**61.98**	**60.75**	**59.84**	**59.49**	**58.74**	**60.60**
Main effects	C	**64.74**	**63.60**	**62.35**	**60.99**	**59.59**	**58.27**	**61.59**
	LS	**63.94**	**63.75**	**63.34**	**62.74**	**62.00**	**61.87**	**62.94**
	TEO	**64.38**	**63.42**	**62.10**	**60.98**	**60.12**	**59.38**	**61.73**
Mean		**64.35**	**63.59**	**62.60**	**61.57**	**60.57**	**59.84**	
F-test A(Var.)=**		**L.S.D0.05**	**B = 0.15**	**AB= 0.21**	**C = 0.21**	**AC=0.30**	**BC=0.37**	**ABC=0.52**

* C: control sample, LS: liquid smoke, TEO: thyme essential oil.
F-test (A= **; B= 0.15; C= 0.21).
L.S.D 0.05 (AB= 0.21; AC= 0.30; BC= 0.37; ABC= 0.52).

Table (3): Changes in crude fat content of chicken and turkey meatballs treated with liquid smoke and TEO during storage at 4±1°C up to 15 days.

Var.	Treat.	Storage periods (days)						Mean
		0	3	6	9	12	15	
Chicken	C	28.07	28.16	29.04	30.18	32.43	33.55	**30.24**
	LS	28.30	28.54	28.57	29.42	30.34	30.69	**29.31**
	TEO	28.10	28.40	29.45	30.56	31.72	32.88	**30.19**
Mean		**28.15**	**28.37**	**29.02**	**30.05**	**31.50**	**32.37**	**29.91**
Turkey	C	31.18	32.32	33.50	34.22	34.31	34.99	**33.42**
	LS	32.38	32.14	32.49	32.63	32.72	32.94	**32.55**
	TEO	31.28	32.40	33.43	34.21	34.60	35.02	**33.49**
Mean		**31.61**	**32.29**	**33.14**	**33.69**	**33.88**	**34.32**	**33.16**
Main effects	C	29.63	30.24	31.27	32.20	33.37	34.27	**31.83**
	LS	30.34	30.34	30.53	31.03	31.53	31.81	**30.93**
	TEO	29.69	30.40	31.44	32.39	33.16	33.95	**31.84**
Mean		**29.88**	**30.33**	**31.08**	**31.87**	**32.69**	**33.35**	
F-test A =**		**L.S.D0.05**	**B =0.17**	**AB= n.s.**	**C =0.24**	**AC= 0.34**	**BC= 0.41**	**ABC=0.58**

* C: control sample, LS: liquid smoke, TEO: thyme essential oil.

Table (4): Changes in ash content of chicken and turkey meatballs treated with liquid smoke and TEO during storage at 4±1°C up to 15 days.

Var.	Treat.	Storage periods (days)						Mean
		0	3	6	9	12	15	
Chicken	**C**	5.14	5.90	6.07	6.14	6.32	6.35	**5.99**
	LS	5.50	5.59	5.63	5.52	5.63	5.51	**5.56**
	TEO	5.42	5.91	5.88	5.94	5.98	5.99	**5.85**
Mean		**5.36**	**5.81**	**5.86**	**5.87**	**5.97**	**5.95**	**5.80**
Turkey	**C**	5.00	5.29	5.50	5.88	5.96	6.44	**5.68**
	LS	4.83	4.96	5.59	5.94	5.85	5.88	**5.51**
	TEO	5.18	5.30	5.76	5.74	5.97	6.01	**5.66**
Mean		**5.01**	**5.19**	**5.62**	**5.86**	**5.93**	**6.11**	**5.62**
Main effects	**C**	**5.07**	**5.60**	**5.78**	**6.01**	**6.14**	**6.40**	**5.83**
	LS	**5.16**	**5.28**	**5.61**	**5.73**	**5.61**	**5.69**	**5.51**
	TEO	**5.30**	**5.61**	**5.82**	**5.84**	**5.98**	**6.00**	**5.76**
Mean		**5.18**	**5.49**	**5.74**	**5.86**	**5.91**	**6.03**	
F-test A=**		**L.S.D0.05**	**B= 0.14**	**AB= n.s.**	**C= 0.20**	**AC= 0.28**	**BC= n.s.**	**ABC= n.s.**

* C: control sample, LS: liquid smoke, TEO: thyme essential oil.

Meanwhile, the ash content of chicken and turkey meatballs treated with liquid smoke showed the lowest increase in ash content. This may be due to the higher the water-binding capacity. Indiarto *et al* (2020) reported that as the amount of free water decreases, Due to the presence of phenolic compounds in liquid smoke, the binding capacity of the liquid smoke treatment will be higher. As a result, the less free water that comes out as a percentage of the total amount of free water, the higher the water-binding capacity.

Effect of liquid smoke or thyme essential oil on pH values of chicken and turkey meatballs

Results in Table (5) showed the pH values of chicken and turkey meatballs untreated (control sample) and treated with liquid smoke or essential oil under levels 1% and 0.1% at zero time and during storage at 4±1°C up to 15 days.

These results revealed that the pH value of untreated chicken and turkey meatballs (control sample) was higher than that of the thyme essential oil. At zero time, the pH values of the control sample and TEO were 5.87, 5.72, and 5.73, 5.67, respectively; and at the end of storage periods, they were 6.77, 6.56, and 6.04.5.96, respectively. These results are consistent with Shaltout *et al.* (2017) who found that during different periods of evaluation, samples treated with thyme essential oil had lower pH values than control samples. Salem-Amany *et al.* (2010) who reported that this could be owing to the activation effect of TEO as an antibacterial agent producing protein breakdown and the development of alkyl groups.

Furthermore, the addition of liquid smoke resulted in a significant decrease in pH, with pH values of 5.63, 5.54 at zero time and 5.55, 5.33 at the end of storage periods. Amaral *et al.* (2018) suggest that due to the decomposition of fatty acids in meat, the oxidation process can decrease the pH of meat during storage. Unsaturated fatty acids decompose into saturated fatty acids, resulting in the creation of short-chain fatty acids, which accumulate in large amounts (Den Besten *et al.*, 2013). Because liquid smoke contains acidic qualities, the higher the liquid smoke concentration applied, the lower the pH value of the smoked product (acid) (Indiarto *et al.*, 2020).

Effect of liquid smoke or thyme essential oil on water holding capacity of chicken and turkey meatballs

Data given in table (6) displayed the effect of addition 1% liquid smoke, and 0.1% TEO for chicken and turkey meatballs compared to control sample on water holding capacity (W H C) during storage at 4±1°C up to 15 days.

The obtained results showed that the WHC of control for chicken and turkey meatballs samples were 83.87 and 79.55%, respectively, at zero time. Meanwhile, with the addition of 1% liquid smoke or 0.1% thyme essential oil for chicken and turkey meatballs, the WHC was 84.62, 81.64, and 82.96, 84.25%, respectively. The ability of derived phenolic compounds to establish a hydrogen bond with water may impact the water-binding capacity of meat (Maga, 1987; Indiarto *et al*,. 2020).

During storage, the water holding capacity decreased. The highest loss of water holding capacity was observed after 12 days of storage periods, with a significant effect ($p<0.05$) for chicken and turkey meatballs. The above-mentioned findings are consistent with those obtained by Yosi *et al.* (2018) who found that during the storage process, anaerobic glycolysis occurs, causing the meat's capacity to bind water to decrease, resulting in more water escaping the meat. Hegazy (2004) reported that the decreased of water holding capacity during storage might be attributed to protein denaturation and losses in protein solubility.

Generally, the lowest loss in water holding capacity was observed in samples treated by liquid smoke then TEO compered to control. Liquid smoke plays a function in loosening the bound of myofibrils fiber to form area that is filled with water in a semi-free form consequently water the ability of the meat to bind water increases (Abustam *et al.*, 2016; Indiarto *et al.*, 2020).

Effect of liquid smoke or thyme essential oil on total bacterial count of chicken and turkey meatballs

The result given in Table (7) showed total bacterial count in different chicken and turkey meatballs samples compared with control samples during the storage period of 15 days at 4±1°C.

The results showed that the total bacterial count of control, 1%liquid smoke, and 0.1% TEO meatballs samples for chicken and turkey meatballs were 4.51, 4.36, 4.47x10^3, and 4.43, 4.16, 4.30 x10^3, respectively, at zero time. After 3 days, total bacterial count decreased significantly ($p<0.05$) for chicken and turkey meatballs of control, liquid smoke, and TEO meatballs samples to 3.74, 3.16, 3.42 x 10^3 and 3.52, 3.48, 3.23 x 10^3, respectively. This may be due to cold temperatures. Arnim *et al.* (2012) reported that proteolysis bacteria are inhibited by cold temperatures alone. Fresh chicken meat is highly perishable and has a short refrigerated storage shelf life. The main causes of psychrotrophic microbial growth and physicochemical changes in refrigerated chicken meat are psychrotrophic microbial growth and physicochemical changes (Rukchon *et al.*, 2014).

After 15 day of cold storage the total bacterial count of control, liquid smoke and TEO meatballs samples for chicken and turkey meatballs were 7.03, 5.19, 5.20 x10^5 and 6.47, 4.80, 5.01 x10^5, respectively. The interaction between treatments × storage days showed highest significant effect ($P\leq0.05$).Also, data in Table (7) indicated that, the increase in the total bacterial count by adding 1% liquid smoke and 0.1% TEO to chicken and turkey meatballs during storage is slower compared to control. This might be attributed to the antimicrobial effect of liquid smoke or TEO. These results agreed with those reported by Mohamed *et al.,* (2011); Kjallstrand and Petersson (2001).

In general, liquid smoke had a larger inhibitory effect than TEO on microbial growth. (Pilevar *et al.,* 2017) who recorded that experiments revealed that liquid smoke has better antibacterial effect than *platyloba* essential oil against *Staphylococcus aurous*. Furthermore, adding essential oils to liquid smoke had no influence on its antibacterial properties. Wahyuningsih (2006) suggested that acetic acid in the liquid smoke component could be the cause of the alteration. The most common organic acid found in smoke has been identified as acetic acid. Minatel et *al.* (2017) stated that antimicrobial action is found in acid compounds, particularly acetic acid. Acetic acid has the ability to enter cell membranes and neutralize the trans membrane pH gradient effectively. Phenolic and acid chemicals, in combination with their bactericidal and bacteriostatic capabilities, cause bacterial cell proteins to denature, causing damage to the cell membrane's hydrophobic bonding components such as protein and phospholipids. This increases cell permeability, allowing the entry of phenolic compounds and organic ions into the cell as well as the release of cell components like proteins and nucleic acids, resulting in cell death.

CONCLUSION

The treatments showed a considerable favorable effect on the overall bacterial count and physiochemical characteristics. During cold storage periods, the chicken and turkey meatballs treated with 1% liquid smoke and 0.1 % thyme essential oil showed more enhancement than the control samples. The most significant effect was seen on samples treated with liquid smoke and then with thyme essential oil, which reduced microbiological damage, preserved physicochemical properties, and improved sensory qualities. As a result, liquid smoke can be used instead of preservatives in some cases.

Table (5) Changes in pH values of chicken and turkey meatballs treated with liquid smoke or TEO during storage at 4±1°C up to 15 days.

Var.	Treat.	Storage periods (days)						Mean
		0	3	6	9	12	15	
Chicken	C	5.87	5.93	6.06	6.19	6.59	6.77	**6.23**
	LS	5.63	5.62	5.60	5.59	5.57	5.55	**5.59**
	TEO	5.73	5.77	5.82	5.88	5.96	6.04	**5.86**
Mean		**5.74**	**5.77**	**5.82**	**5.88**	**6.04**	**6.12**	**5.89**
Turkey	C	5.72	5.79	5.96	6.45	6.35	6.56	**6.14**
	LS	5.54	5.625	5.63	5.55	5.395	5.33	**5.51**
	TEO	5.67	5.70	5.775	5.82	5.87	5.96	**5.80**
Mean		**5.64**	**5.70**	**5.78**	**5.94**	**5.87**	**5.95**	**5.81**
Main effects	C	5.80	5.86	6.01	6.32	6.47	6.66	**6.18**
	LS	5.58	5.62	5.61	5. 57	5. 48	5.44	**5.56**
	TEO	5.70	5.73	5.79	5.85	5.91	6.00	**5.83**
Mean		**5.69**	**5.73**	**5.80**	**6.08**	**6.19**	**6.03**	
F-test A(Var.)=**		**L.S.D0.05**	**B = 0.038**	**AB= n.s.**	**C = 0.054**	**AC= *****	**BC= *****	**ABC=***

* C: control sample, LS: liquid smoke, TEO: thyme essential oil.

Table (6) Changes in water holding capacity (WHC %) of chicken and turkey meatballs treated with liquid smoke or TEO during storage at 4±1°C up to 15 days.

Var.	Treat.	Storage periods (days)						Mean
		0	3	6	9	12	15	
Chicken	C	83.87	77.32	74.19	72.43	70.80	54.38	**72.17**
	LS	84.62	82.78	82.59	82.25	77.16	70.03	**79.91**
	TEO	81.64	79.17	77.00	76.01	73.44	61.37	**74.77**
Mean		**83.37**	**79.76**	**77.93**	**76.90**	**73.80**	**61.93**	**75.62**
Turkey	C	79.55	77.45	73.45	71.54	71.80	40.45	**69.04**
	LS	82.96	82.37	81.30	77.72	74.37	58.81	**76.26**
	TEO	84.25	82.83	81.80	80.19	71.88	53.46	**75.74**
Mean		**82.25**	**80.88**	**78.85**	**76.48**	**72.68**	**50.91**	**73.68**
Main effects	C	81.71	77.39	73.82	71.99	71.30	47.41	**70.60**
	LS	83.79	82.57	81.94	79.99	75.76	64.42	**78.08**
	TEO	82.94	81.00	79.40	78.10	72.66	57.41	**75.25**
Mean		**82.81**	**80.32**	**78.39**	**76.69**	**73.24**	**56.42**	
F-test A(Var.)=**		**L.S.D0.05**	**B = 0.690**	**AB= 0.98**	**C = 0.98**	**AC= 1.39**	**BC= 1.70**	**ABC=2.4**

* C: control sample, LS: liquid smoke, TEO: thyme essential oil.

Table (7) Changes in total bacterial count (log CFU/g) of chicken and turkey meatballs treated with liquid smoke and TEO during storage at 4±1°C up to 15 days.

Var.	Treat.	Storage periods (days)						Mean
		0	3	6	9	12	15	
Chicken	C	4.51	3.74	4.31	5.40	6.81	7.03	**5.304**
	LS	4.36	3.16	3.94	4.23	4.46	5.19	**4.228**
	TEO	4.47	3.42	3.53	3.84	4.53	5.20	**4.170**
Mean		**4.45**	**3.45**	**3.93**	**4.49**	**5.27**	**5.81**	**4.567**
Turkey	C	4.43	3.52	4.20	4.49	5.25	6.47	**4.730**
	LS	4.16	3.48	4.04	4.03	4.25	4.80	**4.132**
	TEO	4.30	3.23	4.15	4.25	4.90	5.01	**4.311**
Mean		**4.30**	**3.41**	**4.13**	**4.26**	**4.80**	**5.43**	**4.388**
Main effects	C	4.47	3.63	4.25	4.94	6.03	6.75	**5.017**
	LS	4.38	3.33	3.84	4.13	4.71	5.00	**4.236**
	TEO	4.26	3.32	3.99	4.05	4.36	5.11	**4.185**
Mean		**4.37**	**3.43**	**4.031**	**4.37**	**5.03**	**5.62**	
F-test A(Var.)=**		**L.S.D 0.05**	**B (Tre.)= 0.05**	**AB= 0.07**	**C (P)=0.07**	**AC= 0.10**	**BC= 0.12**	**ABC= 0.18**

* C: control sample, LS: liquid smoke, TEO: thyme essential oil.

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تأثير الدخان السائل وزيت الزعتر على جودة كرات لحم الدجاج والديك الرومي المبردة

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أجريت هذه الدراسة لتقييم تأثير سائل التدخين الناتج من نشارة خشب الزان وزيت الزعتر العطري على جودة كرات لحم الدجاج والديك الرومي أثناء التخزين المبرد. وتم استخدام ثلاثة عوامل في البحث وهي: كرات لحم الدجاج والديك الرومي، تركيز سائل التدخين وزيت الزعتر العطري (1٪ و 0.1٪ على التوالي) و التخزين علي درجة حرارة التبريد لمدة (0 ، 3 ، 6 ، 9 ، 12 و 15 يومًا). وتم تقدير الخواص الفيزوكيميائية للمنتجات (الرطوبة، البروتين، الدهون، الرماد، قيمة الأس الهيدروجيني والقدرة على الإحتفاظ بالماء) والعدد الكلي للبكتيريا. كان للمعاملات تأثيراً معنوياً على الخواص الفيزوكيميائية والعدد الكلي للبكتيريا. كما اوضحت الدراسة ان أقل معدل فقد في الرطوبة و البروتين والماء المفقود في العينات المعامله بسائل التدخين يليها زيت الزعتر العطري مقارنة بعينة الكنترول أثناء التخزين. كما زادت قيمة الدهون الكلية و الرماد والعدد الكلي للبكتيريا أثناء التخزين، وكانت أعلى نسبة في عينات الكنترول وأقلها في العينات المعاملة بسائل التدخين ثم زيت الزعتر العطري والتي لم تصل الي مستويات التدهور. انخفضت قيمة الأس الهيدروجيني أثناء التخزين بإضافة سائل التدخين بنسبة 1٪ مقارنة بزيت الزعتر العطري وعينات الكنترول. كما أظهر التفاعل بين المعاملات وفترات التخزين تأثيراً معنوياً على البروتين والدهون وقيمة الأس الهيدروجيني والقدرة على الاحتفاظ بالماء والعدد الكلي للبكتيريا. علاوة على ذلك ، لوحظ أقل تغيير في العينات المعامله بسائل التدخين أثناء فترة التخزين. كما أدي استخدام سائل التدخين إلى تقليل معدل النمو الميكروبي والحفاظ على الخواص الفيزوكيميائية، وتم قبولها بشكل أفضل مقارنةً بزيت الزعتر وعينات الكنترول.