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THE EFFECT OF ADDING SOME ESSENTIAL OILS TO THE PHYSICOCHEMICAL PROPERTIES OF FROZEN BEEF SAUSAGE WITH DIFFERENT STORAGE PERIODS

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Abstract

This study was conducted for the purpose of determining the effect of adding 1% of essential oils (rosemary leaf oil, oranges peel oil, and lemon peel oil) to T2, T3, and T4, treatments respectively, and the effect of these additives on physicochemical properties, which include water-retaining meat. The loss percentage of exuded liquid, the loss percentage during cooking of frozen beef sausage at (-18 °C), and for different storage periods (1, 15, 30, 45 days), while T1 (Control treatment) was left without any addition. The results of the statistical analysis of the physicochemical properties on the treatments to which essential oils were added, showed no significant differences at (P <0.05) in meat capacity to retain water, the weight lost by dissolving character, and the losing characteristic during cooking, during cold and freeze storing and for the previously mentioned periods. It is concluded from this study that the treatments for which essential oils were added showed the best results in influencing the physicochemical properties of freeze stored beef sausage for a period of (45 days).
Keywords : Essential oils, physicochemical properties, frozen beef sausage, different storage periods

Introduction

Meat and meat products are exposed to deterioration during storing due to their chemical nature and nutrient contents. The nature of damage is either chemical or microbial, which are the two main factors affecting the quality of meat. Fat oxidation is also an important factor affecting Meat quality during meat processing and storage operations. Meat processing will lead to the expansion of the surface area during mincing and exposing the meat to air as well accelerates the loss of internal muscle components (Nam and Ahn, 2003). (Karwowska and Dolatowski, 2007) found that fat oxidation is one of the most common forms of chemical damage. Foods containing a high proportion of fat, such as meat and its products, are exposed to oxidation during long-term storage, which leads to changes in sensory characteristics such as color, taste, smell, a decline in nutritional value. The free radicals resulting from fat oxidation can oxidize oxymyoglobin dye into metmyoglobin dye and thus, change the color of meat products. Due to the transfer of oxygen, the oxidation process within cells, results in significant damage to cells, disruption of the health of the cell and its natural functions, and may lead to the occurrence of diseases (Youssef, 2014). Oxidation can be divided into enzyme oxidation, which includes enzyme action, such as lipoxygenase, and chemical oxidation, which is caused by light, high temperature, exposure to oxygen or to mineral ions, such as iron (Al-Halfi, 2016). In addition, issues that have aroused the attention of the producer and consumer with the development of modern technologies in food manufacturing sector is meeting the growing food requirements of humans, satisfying the desires of consumers through food safety, preserving its nutritional value, extending its storage life, reducing damage while it is circulating and storing, and moving away from what is industrial, and insure the health and safety of the consumer,

The negative effects of oxidation on food quality have increased the interest in antioxidants (Al-Halfi, 2009). The use of antioxidants in meat and its products reduces or inhibits fat oxidation, thus extending its storing life by inhibiting antioxidants through breaking the chain of reaction and maintaining the catalysts of oxidants such as mineral ions. There are many compounds that demonstrate anti-oxidation effectiveness, but few of them can be used in food production and must be subjected to laws and food specifications (Karre *et al*., 2013). Antioxidants are a class of compounds with different chemical structures and various mechanisms of action, and the most important mechanism is to interact with the free radical of fat, thereby producing ineffective products (Pokorný *et al*., 2001). Meat and meat products needed to be equipped with natural antioxidants derived from animal and plant sources (Cheung *et al*., 2012). A natural alternative to chemical preservatives is essential oils and their use in food is according to consumer's demand to provide processed products with high nutritional value and quality, reducing the damage caused by the use of chemical preservatives and replacing them with natural preservatives (Nychas, 1995). Essential oils are defined as volatile substances that evaporate easily upon heating, and comprise a complex group of many different chemicals that give them their separate characteristics and effects, such as terpnes, and a high proportion of alcohol that gives them their sanitization character, as well as esters, phenols, and flavonoids. The progress in their use in natural products, such as antioxidants and microbial activities in food products became necessary and useful for extending storage period and also for preventing food contamination (Grost, 2017).

Materials and Methods

Beef meat

Fat free beef meat obtained from a piece of thigh from local markets was used. The meat was put at 2ºC for 24 hours, then, it was cut into small cubes of 2-3cm^3 dimensions before starting the process of sausage processing for the purpose of facilitating the mincing process (Mahmmod, 2018).

Fat

The fat localized around the pelvic bones and kidney of a calf sacrifice was obtained from the local market (the same sacrifice from which the pure meat was taken) and was put in special plastic bags that do not allow air pass through and kept in freezing until use. Fat bags were kept sealed in a tight manner during the preservation period. Fat was then cut into small cubes with dimensions of 2-3cm^3 before starting the process of making sausage for a smooth mincing process.

Spice

Different types of non-grinded spice from local markets in Baghdad was obtained and used. Each spice was milled separately by a coffee mill, then, the spice mixture was prepared according to (Mahmmod, 2018) method, which was later used for making sausage as shown in Table 5. The spice mixture was then placed in sealed plastic containers until use.

NaCl Salt

Pure, crystal and processed salt supplied by Merck was used.

Wrappers

Natural wrappers were used sausages filling and large intestine of the sheep was obtained, and the following steps were followed in the cleaning and preparation process. According to (Mahmmod, 2018), large intestine was washed with running water for disposing waste. The inner part of the large intestine was scratched using the blunt edge of the knife to remove the mucous layer. The remaining part of the large intestine (collagenous material) was washed with running water in order to remove the coatings, mucus, and other suspended matter. Then, the large intestine was placed in a saline solution at a concentration of 15% in order to stop the growth of microscopic organisms. Acetic acid was added to the solution (1%) in order to obtain the desired white color of the large intestine.

Sausage processing and treatments division

The following steps were followed in the manufacture of sausage according to (Mahmmod, 2018) study. The weights of fat free meat were prepared with ratio of (3:1), indicating that the weight of the manufactured meal is 12 kg, the pure meat and the fat have been minced using an electric chopper, using a 1.5 cm diameter pores disk for the first mincing and a 0.8 cm diameter pores disk for the second mincing process. Essential oils were added to the minced meat mixture and each treatment weighted (3 kg) as follows: T1 (control treatment) without adding essential oils, T2 1 ml/kg meat of lemon peel oil was added , T3 1 ml/kg meat of orange peel oil was added, and T4, 1 ml/kg meat of rosemary leaf oil was added. Table salt was added to the pure meat and fat mixture with a ratio of (1.5%) and spices for the experiment parameters. The mixture was manually mixed using sterile medical gloves to obtain the initial homogeneity of the materials used. The prepared minced sausages mixture of four treatments was filled using natural wrappers in a specific way, which included the use of an electric mincing machine with removing the knife and the disk. A 15cm stainless steel metal pipe with a 2cm diameter locally made was attached to the mincing machine with taking into consideration two basic conditions, which are homogeneity and uniformity to avoid light filling of air gaps in the casing, as well as to avoid severe packing that obstructs the evaporation process during the ripening stage. The sausage was sealed by using wax-coated cotton thread, as the sausage was 20 cm in length and 2.5 cm in diameter for all treatments. Sausage samples were hanged in a sequenced manner by using hocks and placed in a refrigerated incubator at a temperature of 4°C for 1, 3, and 7 days.

Sausage frying method

The method described by (Peryam, 1990) was applied for frying the sausage samples assigned for sensory testing, as the sausage was cut into small rounded slices of 1.5cm in thickness and put in 99% pure corn oil at 120 °C (boiled) for two minutes. Then, the slices were placed on N.1 Whatman filter paper for a short time to remove excess oil in sausages.

Physical examinations of meat

Loss through exuded liquid

Weight loss after refrigerating was assessed according to the method described by (Young and Lyon, 1997). 5 kg of meat samples was taken after removing the packaging material and left in the refrigerator at 4°C for 24 hours. Samples were re-weighed after drying them and the liquids on the meat sample surface were removed using a filter paper. The weight loss ratio was calculated using the following equation:

$$\text{Loss after refrigerating (\%)} = \frac{\text{(Weight of meat sample before refrigerating - Weigh of the sample after refrigerating)}}{\text{Weight of meat sample before refrigerating}} \times 100$$

Loss during cooking

Loss percentage during cooking was estimated according to the method described by (Dolatowski and Stasiak, 1998). Meat samples were placed in refrigerator at (4°C) for 24 hours prior to cooking. 20 g of meat samples were taken and placed on aluminum foil and cooked in an electric oven at 200 °C for 10 minutes. After cooking, meat samples were dried using filter papers and then cooled for 30 minutes at room temperature and weight loss percentage during cooking was calculated by the difference in the weight of the sample before and after cooking according to the following equation

$$\text{Loss after cooking (\%)} = \frac{\text{(Weight of meat sample before cooking - Weigh of the sample after cooking)}}{\text{Weight of meat sample before cooking}} \times 100$$

Water Holding Capacity (WHC)

Method described by (Dolatowski and Stasiak, 1998) was followed for the measuring water holding capacity. 50g of sausage samples were taken and homogenized with (50 ml) of distilled water for 1 minute using Ho4 Tafesa homogenizer supplied by Hannover company. Homogeneous mixture was centrifuged using IEC Beckman centrifuge at (4°C) at a speed of (5000 rpm for 10 minutes); the water holding capacity was calculated as follows:

$$\text{Water holding capacity (\%)} = \frac{\text{Distilled water weight - weight of filtrate after centrifuging}}{\text{Sample weight (g)}} \times 100$$

Result and Discussion

Water Holding Capacity (WHC)

Table (1) shows the effect of interference between (T1, T2, T3, and T4) treatments of chilled beef sausage stored for different time periods (1, 15, 30, and 45 days) in the capacity of meat to hold water (WHC). It is noted from the table that there are no significant differences on ($p \leq 0.05$) level in (WHC) values. T4 treatment (rosemary leaf oil) was the highest in (WHC) values among other T2 and T3 treatments for the chilled beef sausage. T1 (control) treatment, which recorded a decrease in cold and freeze storage periods reaching 34.77%. On the other hand, T3 treatment (orange peel oil) ranked second in the average values of (WHC) reaching an average of 36.48% compared to T4 treatment, while T2 treatment (lemon peel oil) ranked third in the average values of (WHC) for with a rate of 75.35% and for the same storage periods. It is noted from Table (1) that T4 treatment (rosemary leaf oil) recorded a significant increase in the values of (WHC) compared with all other treatments (T3 and T2) compared to T1 control treatment, which recorded the lowest (WHC) values for the same cold and freeze storage periods. These outcomes can be attributed to the active compounds in these treatments that have the ability to protect cellular membranes from breaking down and thus protect proteins from decomposition and prevent the water from exiting outside and remaining associated with protein-water bonds (Viuda-Martos *et al*., 2015). As for the effect of the cold and freeze storage period, 9T2, T3, and T4 treatments showed higher levels in (1 day) of cold and freeze storage period compared to the control treatment (T1), then, started to reduce in varying rates as the storage period progresses until it reaches lower values in the 45-day period respectively, which recorded higher rates than control treatment as shown in tables (1). The high WHC values can be attributed to the increase in the pH and the protein content in the meat, which contributes to the ability of the meat to retain and hold large amount of water, and therefore it is not possible to raise the values of the (WHC) as indicated by (Tahir, 1990).

In addition to the foregoing, (Acton *et al*., 1983) suggested that the increase in meat proteins solubility increases the binding ability to water, while the decrease in water holding ability due with the progress of storage periods for all treatments is due to some changes in the meat pH and meat proteins during cold and freeze storage. As these proteins decompose and break down, the non-protein nitrogen compounds (NPN) increase and so all these factors may lead to a decrease in the ability to hold and/or recover water lowers humidity (Taher, 1983). On the other hand, a study by (Salama, 1993) indicated that there was a decrease in the ability to hold water for a frozen chicken meat sausage (-18 °C) for a storage period of 90 days, and the reason for this was the occurrence of protein denaturation as well as some biochemical changes associated with meat freezing . A direct relationship between the protein content and the ability to hold water, is present as the higher the protein content in meat and meat products, the greater its ability to hold water. This indicates that protein is the main component of water binding in meat (Shalaby, 1986).

Table 1 : the effect of adding essential oils (rosemary leaf oil, lemon peel oil, orange peel oil) on the ability of meat to hold water for beef sausage during storage by freezing for different periods.

Character	Treatments	Storage period (day)				Average
		1	15	30	45	
Water holding capacity (%)	T1	41.15	35.50	32.66	29.78	34.77
	T2	42.16	35.68	33.34	31.82	35.75
	T3	43.56	36.22	34.13	32.02	36.48
	T4	45.60	37.84	35.33	32.12	37.72
LSD		NS	NS	NS	NS	---

* ($P < 0.05$), NS: non-significant, (values in the table are averages for three replicates) T1: Without any addition, T2: 1% lemon peel oil, T3: 1% orange peel oil, T4: 1% Rosemary leaf oil.

Weight loss rate through exuded liquid

Table (2) shows the effect of the intersection between (T1, T2, T3, T4) treatments for (1, 15, 30 and 45 days) freezing storage periods on the rate of weight loss through exuded liquid of frozen beef sausage. Results from the table shows that there are no significant differences at the level of $p \leq 0.05$ in the loss ratio in exuded liquid during freeze storage. T4 treatment (rosemary leaf oil) recorded the lowest percentage of loss through exuded liquid from the rest of T2 and T3 treatments, which came first in the rates of the percentage of loss through exuded liquid from the rest of the treatments. On the other hand, T1 treatment (control), recorded an increase in the rate of loss reaching 1.61%.

As for T3 (orange peel oil) treatment, it came second in the rate of weight loss from exuded liquid for the same freeze storage periods reaching an average of 1.52% compared with T4 treatment, while T2 (lemon peel oil) treatment came third in the rate at a rate of 1.56% and for the same freeze storage periods. In addition, T4 (rosemary leaf oil) treatment recorded a marked decrease in the rate of loss compared to T2 and T3 treatments compared to T1 Treatment (control) which recorded the highest loss rate and for all the freeze storage periods.

These outcomes can be attributed to the effect of essential oils, which have a high effectiveness in protecting the cellular wall from damage caused by oxidation and increasing constancy. The solution lies in the lack of clearing of the exudate fluid from inside the cell and this was confirmed by (Viuda-Martos *et al*., 2015) study where an improvement in color and a reduction in fat oxidation was observed compared with the control treatment after freeze storing the meat for 14 days. On the other hand, a study by (Al-Alwani, 2017) reported a decrease in the amount of exuded liquid. The reason for this is the digestion of meat proteins by enzymes (cathepsins), which are responsible for some subtle changes in the permeability of the cell membrane or the fundamental structure of the protein, and therefore the reduction in the ability of meat to hold water, especially protein-related water, and therefore the increase in its rate (Al-Alwani, 2017).

A study by (Salama, 1993) had previously reported a decrease in the ability to hold water for chicken meat sausage freezing stored at (-18°C) for a period of 90 days. These observations can be due to the occurrence of a protein denatioration, as well as some biochemical changes related to frozen meat. There is a direct relationship between the protein content and the ability to hold water, as the higher the protein content in meat and meat products, the greater its ability to hold water. This indicates that protein is the main component of water binding in meat (Shalaby, 1986).

Table 2 : Effect of adding essential oils (rosemary leaves oil, lemon peels oil and orange peel oil) to the loss rate through exuded liquid of beef sausage during freeze storage for different periods.

Character	Treatments	Storage period (day)				Average
		1	15	30	45	
Water holding capacity (%)	T1	1.20	1.52	1.76	1.96	1.61
	T2	1.17	1.48	1.74	1.87	1.56
	T3	1.15	1.42	1.70	1.81	1.52
	T4	0.98	1.29	1.66	1.72	1.41
LSD		NS	NS	NS	NS	---

* (P <0.05), NS: non-significant, (values in the table are averages for three replicates) T1: Without any addition, T2: 1% lemon peel oil, T3: 1% orange peel oil, T4: 1% Rosemary leaf oil.

Weight loss after cooking

Table (3) shows the effect of the interference between (T1, T2, T3, and T4) treatments for (1, 15, 30 and 45 days) freeze storage periods on the loss percentage during cooking for frozen beef sausage. No significant differences were observed at (p≤0.05) level in the percentage of loss during cooking after freeze storage.

T4 treatment (rosemary leaf oil) showed the lowest in loss percentage during cooking compared to the other T2 and T3 treatments, which came first in the rates of loss during cooking compared to the rest of the treatments reaching an average of 21.37% for all different storage periods and compared to T1 (control) treatment that recorded a high significant increase (P <0.05) in freeze storage periods reaching 23.59% of loss during cooking.

Furthermore, T3 treatment (orange peel oil) came second in the rates of loss during cooking for the same freeze storage periods, which amounted to 21.81% compared to T4 treatment, while T2 treatment (lemon peel oil) recorded third in rates of loss during cooking at a rate of 24.22%.

A significant decrease in loss percentage during cooking and shrinkage rate of in meat tablets treated with plant extracts was observed by (Al-Athari, 2016). This is probably due to the interaction activity of natural antioxidant in increasing the ability of meat tissue to hold water and reduce water lost during storage and cooking (Al-Rubaie *et al*., 2008).

This observation can be attributed to the decrease in humidity as a result of evaporation of the surface water on meat. On the other hand, the degradation of meat proteins by the enzymes that break down the bonds that link the protein with the water reduce the ability of the meat to bind to water and hence, be subject to evaporation (Juárez *et al*., 2010).

Table 3 : Effect of adding essential oils (rosemary leaf oil, lemon peel oil, orange peel oil) on the loss percentage of beef sausage during cooking for different freeze storage periods.

Character	Treatments	Storage period (day)				Average
		1	15	30	45	
Water holding capacity (%)	T1	36.80	20.65	19.50	17.43	23.59
	T2	36.30	19.55	17.60	15.54	22.24
	T3	35.90	18.94	17.10	15.32	21.81
	T4	35.50	18.11	16.90	14.98	21.37
LSD		NS	NS	NS	NS	---

* (P <0.05), NS: non-significant, (values in the table are averages for three replicates) T1: Without any addition, T2: 1% lemon peel oil, T3: 1% orange peel oil, T4: 1% oil Rosemary leaves.

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