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Natural Alternatives of Nitrate and Nitrite as Preservative Agents in Pastrami

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Abstract

The objective of this study was to compare the effect of nitrates and nitrites as common preservatives and its alternatives to reduce the use of these substances to the absolute minimum required for preservation effect in pastrami. In this work, the effect of preserving pastrami with sodium nitrite (200 ppm), thyme essential oil (0.5%), and smoke liquid (1%) were investigated. Packed samples with additives and a control were stored at 4±1°C for 30 days. Analysis of sensory and biochemical alterations were done over the duration of storage. The outcomes showed that the use of 1% smoke liquid for preserving meat products made the best-preserving properties (TBA value and peroxide value were decreased), lowering the residual nitrite and nitrate content with no existence of N-nitrosamine compounds, and all the studied characteristics were at the completion of the storage terms, within the permitted limitations specified by the Egyptian organization for standardization and quality control.

Keywords: nitrate, nitrite, N-nitrosamine, Natural Alternatives and Pastrami.

INTRODUCTION

Pastrami is a dry-cured meat product preserved by nitrite or nitrate salts, seasoned, and coated with garlic and fenugreek, according to E.O.S. (2005/1042). In meat curing, table salt, nitrate or nitrite with other chemicals, and various spices are used (Aksu et al., 2017). During the preparation and processing of pastrami, it is susceptible to contamination by spoilage bacteria that cause spoilage and pathogenic bacteria that cause foodborne illness (Abd-Elghany et al., 2020). Meat is considered to be a good source of vitamins, essential amino acids, proteins, and minerals, as well as having a low carbohydrate and lipid content. Meat is a high biological value protein source, indicating the amino acids found in the meat are important to human needs, regardless of age, ethnicity, sex, or area of production. Fat is a determinant of meat quality since it affects organoleptic properties, palatability, and nutritional value, but it is also a substrate for lipoperoxidation in meat products (Ribeiro et al., 2019). Meat products, on the other hand, are high in salt, saturated fats, and cholesterol. During processing and storage, meat and meat products are susceptible to lipid and protein oxidation, resulting in quality degradation (Domínguez et al., 2019; Fernandes et al., 2017). As Nitrites, despite their considerable technological utility, (a) contribute to the development of a distinctive pink color, (b) have antioxidative activity, (c) is responsible for the production of the distinctive flavor of cured meats, (d) prevent the growth of germs that cause food to degrade, especially Clostridium botulinum, which produces the potentially fatal toxin botulin (Deda et al., 2007). Methemoglobin formation is the most visible and identifiable symptom of nitrite toxicity in humans. Furthermore, nitrites may combine with certain amines in foods to form N-nitrosocompounds, many of which are known carcinogens. Because the rate of nitrosamine generation is related to the square of nitrite concentration, lowering the latter has a greater impact on the number of nitrosamines created in meat products. Because of the potential health danger posed by nitrites, there is a lot of interest in the creation of natural food colorants, which are regarded to be healthy and of high quality (Deda et al., 2007). As a result, consumers' preference for natural additives is growing by the day. Natural preservatives can be found in plants, microbes, fungi, animals, and algae (Gokoglu, 2019). Natural herbs and spices have preservative properties that vary depending on the organism's form, composition, and concentration. Volatile chemicals can be found in herbs and spices. These chemicals contain compounds that act as self-defense mechanisms in plants (Singh et al., 2010; Gokoglu, 2019). Phenolic compounds, such as thymol from thyme is the major antimicrobial components in plants and their extracts and essential oils (EOs). While novel antimicrobial compounds are often isolated from bacteria and fungi, herbs and spices are considered alternative sources (Hintz et al., 2015). Thyme has been used in meat and meat products for centuries as a flavoring ingredient (Kassem et al., 2011). Common thyme contains 0.8-2.6 % volatile oil, consisting of phenols, monoterpene hydrocarbons, and alcohols in varying amounts. Thymol is usually thought of as a key phenolic component in thyme. Thyme is a natural food preservative, antispasmodic, carminative, antiseptic, antimicrobial, and antispasmodic (Ghasemi et al., 2012). Antibacterial activity of thyme has been demonstrated against bacteria such as Staphylococcus aureus, Bacillus subtilis, Escherichia coli and Shigella sonnei. Only a few flavonoids found in thyme essential oil. Thyme's antioxidant capacity is enhanced by these flavonoids (Kassem et al., 2011). Historically, the smoke has been used to successfully preserve food, frequently in conjunction with other challenges such as boiling and drying. Natural wood smoke is a suspension of vapors, solid particles, and liquid droplets produced by regulated wood burning in the absence of or at reduced oxygen levels (Holley and Patel, 2005). There are around 400 chemical components in smoke liquid that have been recognized. Acids that react with proteins to generate brown and phenol stains, which are the principal ingredients of scent and demonstrate antioxidant activity, are among the components that might alter the flavor, pH, and shelf life of smoked carbonyl goods (Ali and Al Fiqri, 2020). Carbonyl

components in smoke contribute to the color and flavor of smoked items, giving them a distinct caramel-like scent. In contrast, acidic components serve as antibacterial agents and contribute to the smoked product's taste (Ariestya, 2016).

MATERIALS AND METHODS

Materials

Meat

Fresh lean round (long muscles) of cow meat was purchased from a slaughterhouse at Sohag city, Egypt, visible fats and connective tissues were manually removed, and the lean round samples were used to make Pastrami (Ibrahim, 2001).

Other ingredients:

Fresh garlic, fresh tomato, fresh onion, salt, eggs, wheat flour, bread crust powder, paprika, and fenugreek were bought from a neighborhood market in the Egyptian city of Sohag. In addition, rehydrated extruded soy was purchased from a local herbalist.

Chemicals:

Some chemicals were purchased from Sigma – Aldrich (St Louis, MO, USA). The other chemicals were either of analytical grade or of the highest quality was purchased from El-Gomhoria Company, Egypt.

Smoke liquid:

Smoke liquid prepared from Beech sawdust was obtained from the Food Science and Technology Department, Faculty of Agriculture, EL-Azhar University, Assiut, Egypt (2021).

Essential oils:

Thyme (*Thymus vulgaris*) was purchased from the essential oils laboratory of the National Research Center, Giza, Egypt (2021).

Preparation of pastrami samples:

Pastrami samples were prepared and manufactured in two phases, according to Ibrahim (2001).

1. Salting and aging:

The lean round (long muscles) was cut into equal sections, measured, and fractured on the surface using a knife to allow the salting and curing mixture to penetrate through the tissues. The meat samples were then separated into eight portions, the first of which was left unchanged (control), and the other seven of which were individually mixed with various preservative ratios as shown in Table (1). The samples were set on a stainless-steel rack, and the following salting (curing) combination was equally applied to its surfaces: 150 g of NaCl, 1 g of ascorbic acid, and 0.1 g of sodium nitrite (g/kg meat). The samples were then covered with cotton tissue, a hardwood plate, and a heavyweight weighing around 30 kg. The samples were dried overnight (~12 h) after being rinsed with distilled water to remove extra salt and left to drain for two days at room temperature (around 25° C).

Table (1): Additional ingredients used in pastrami formula

Treatments	Salting mixture ingredients (g/kg meat)
Control	Only salt (without any additional ingredients)
Treatment $_2$ (basal pastrami formula)	NaCl 150 g + ascorbic acid 1 g + sodium nitrite 0.1 g
Treatment $_3$	NaCl 150 g + 5 ml thyme essential oil
Treatment $_4$	NaCl 150 g + 10 ml smoke liquid

2. Acceleration of curing:

Before curing, three layers of the following curing paste were applied to the entire surface of the sample, which was thoroughly mixed with a sufficient amount of water to form a soft paste (see Table 2). After each layer application, the samples were hung for 1 h. The meat samples were then heated in an electric oven at 88° C for partial drying until the internal temperature reached ~71° C. Next, the internal temperature of the meat samples was measured using an electronic thermometer. To finish the curing and drying process, the samples were hung in the air for 6 days at room temperature.

Pastrami samples were minced and kept cool for 30 days in polyethylene bags at 4 °C.

Table (2): Curing paste formula.

Ingredients	g/kg meat
Salt and curing mixture	20.0 g
Paprika	90.0 g
Fenugreek	90.0 g
Wheat flour	4.0 g
Fresh garlic	180.0 g

Analytical methods

Chemical analysis of meat products:

Moisture, ash, protein, fat, and carbohydrate contents were determined according to A.O.A.C. (2016).

Determination of minerals content:

The A.O.A.C. (2016) states that the samples were burned at 550 °C in a muffle furnace before being mixed with 5 mL of hydrochloric acid Conc. and diluted to 50 mL with deionized water. Then an atomic absorption spectrophotometer was used to quantify the amounts of zinc, copper, cadmium, and lead. They were established in the Faculty of Agriculture's Water and Soil Department at Sohag University in Egypt.

Determination of thiobarbituric acid value (TBA):

TBA value in meat products can be determined directly using the method described by EOS (2006). In a 500 ml distillation flask, combine 10 gm minced meat with 50 ml distilled water, then add (47.5 ml distilled water + 2.5 ml 4N hydrochloric acid) to the flask until the pH exceeds 1-5. Then add a few small pieces of a pumice stone to the beaker to prevent foaming. Within 10 minutes of boiling, the beaker containing the mixture is heated on an electric heater to distill around 50 mL of the distiller. Then, in a test tube with a cap, put 5 ml of the distillate and 5 ml of the thiobarbituric acid preparation (0.2833 g of the acid dissolved in100 ml glacial acetic acid 90 %). The tube's head is sealed with a sanded cap or a strong cotton stopper, and it's put in a water bath that boils for 35 minutes from start to finish. The optical absorption force is measured at 538 nm after the tubes have been soaked in running water for 10 minutes. The results were measured and expressed in milligrams of malonaldehyde per kilogram of meat tissues. The following equation was used to measure the quantity of malonaldehydes:

TBA = A×7.8

A= Absorbance of the sample.

Determination of peroxide value:

According to A.O.A.C. (2016), the peroxide value as a lipid oxidation was measured as follow:

In a conical flask, 0.5 g of samples are combined with 25 ml of a mixture of glacial acetic acid and chloroform (3:2, v/v), followed by 1 ml of saturated potassium iodide. After 10 minutes in the dark, 30 mL distilled water and 1 mL freshly formulated 1 % starch solution were applied to the mixture. The samples were titrated with 0.01 N sodium thiosulphate until the blue color was gone after shaking. The peroxide concentrations were measured in meq/kg of sample.

Determination of nitrate, nitrite and N-nitrosamine:

As in our earlier work, measurements of nitrate, nitrite, and N-nitrosamine were acquired and made (Srour *et al.,* 2022).

RESULTS AND DISCUSSION

Chemical composition of pastrami:

Data presented in Table (3) revealed the chemical composition of pastrami. It could be noted that pastrami contains 58.49% moisture, 13.74% total ash, 33.30% crude protein, and 4.71% crude fat. These results are in the line with those reported by (Abdallah *et al.*, 2017; Abd-Elghany *et al.*, 2020). These results are also in line with EOS (2005) for pastrami. Furthermore, the results in this Table showed that several heavy metals such as Zn, Cu, Pb, and Cd were found in pastrami in amounts of 0.580, 0.041, 0.034, and 0.008 ppm; respectively. These results were in line with (Alturiqi and Albedair, 2012; Korish and Attia, 2020; Maky *et al.,* 2020). In addition, the EOS

(2010) indicated that Pb concentration in pastrami has to be less than 0.1 mg /kg and Cd concentration has to be less than 0.05 mg /kg. According to FDA (2001) Zn and Cu concentration in fresh pastrami should be 0.3-1.00 mg /kg and 0.05- 0.5 mg /kg; respectively. Furthermore, in Egypt, the permissible level of Cu in meat and offal should not exceed 15 ppm, according to EOS (2010).

Table (3): Gross chemical composition of fresh pastrami.

	Moisture %	Ash %	Protein %	Fat %	Carbs* %	Zn (ppm)	Cu (ppm)	Pb (ppm)	Cd (ppm)
Mean	57.73	13.47	33.30	0.24	0.00	2.627	10.33	0.901	0.026
SD	0.114	0.289	0.301	0.029	0.00	1.388	0.895	0.692	0.015

*Carbs refers to carbohydrates

Thiobarbituric acid (TBA) values of pastrami treated with different preservatives during storage at 4±1°C for 30 days:

The formation of oxidative rancid flavor in processed meat created during refrigeration and frozen storage has a negative impact on its desirability. To assess the level of oxidative rancidity (malonaldehyde formation) in beef meat, the thiobarbituric acid (TBA) test is utilized. A sensitive test for highly unsaturated fatty acid breakdown products that are absent from the peroxide value calculation is the (TBA) test. (Mohamed, 2005; Mohamed, 2011; Mahmoud, 2013). The results in Table (2) indicate the changes in thiobarbituric acid (TBA) values of pastrami samples treated with different preservatives during storage at 4±1°C. According to the data in Table (4), the thiobarbituric acid (TBA) value was 0.32 mg malonaldehyde/Kg sample. This result was in line with (Ayas *et al.*, 2020). There were no differences in TBA values between treated samples and untreated samples (control samples) at zero day. However, there were significant differences (P≥ 0.05) in TBA values between the treatments throughout the storage periods. TBA values in the control sample gradually increased with time, peaking at 0.969 mg malonaldehyde/Kg sample at the end of the storage period. The other treatments showed a slight increase in TBA levels during storage at 4±1 °C for up to 30 days. However, these gains were far lower than those observed in the control group. According to Mahmoud, (2013); Milly *et al.*, (2008); Akköse *et al.*, (2018); Malelak *et al.*, (2019), The slight increase in pastrami samples treated with various preservatives could be due to their effects on microbial activity, which increases oxidative deterioration in pastrami, such as carbonyl components in smoke liquid, or the antioxidant activity of some compounds, such as thymol in thyme, and phenolic compounds in smoke liquid. According to the gathered data, the initial TBA value reported in the control sample was 0.356 mg malonaldhyde /Kg sample. Meanwhile, it measured 0.2; 0.287 and 0.2 mg malonaldhyde/Kg for samples made with nitrite 200 ppm, thyme 0.5% and smoke liquid 1%. At the end of the storage period, the TBA values for samples prepared with nitrite 200ppm, thyme 0.5%, and smoke liquid 1%, were 0.43, 0.757 and 0.435 mg of malonaldhyde/Kg, respectively. The TBA values of the control sample increased gradually throughout cold storage at 4±1 °C up to 30 days, peaking at 0.969 mg malonaldehyde /kg of the sample at the end of the cold storage periods. Although the TBA values of the other pastrami treatments were generally lower than the control sample, there were some notable variances in some cases. For example, pastrami samples created with nitrite 200 ppm and smoke liquid 1% had the lowest TBA values at the end of cold period storage (0.431 and 0.435 mg malonaldehyde/Kg sample, respectively). which might be ascribed to the antibacterial and antioxidant chemicals in smoke liquid, including aldehydes, carboxylic acids, and phenols. The phenolic and carbonyl components determine smoke liquid 's antibacterial activity; however, carbonyls are more effective antibacterial than phenols (Milly *et al.*, 2008; Malelak *et al.*, 2019). The ability of nitric oxide to bind to and stabilize heme iron of meat pigments during the

curing process is credited with nitrite's antioxidant effect. Nitric oxide reacts quickly with oxygen and other reactive oxygen species, sequestering them (Ford and Lorkovic, 2002; Alahakoon *et al.*, 2015). In contrast, those made with thyme EOs 0.5% had higher TBA value. The antioxidant and antibacterial activity of several preservative components, especially volatile oils and phenol compounds, could explain the slight increase in TBA values of numerous pastrami treatments compared to control (with no additives).After 30 days of storage, all pastrami samples, including the control samples, were confirmed to be safe to consume, with the highest TBA value (0.969 mg malonaldehyde/Kg sample). The thiobarbituric acid (TBA) test is widely used in stores as a lipid oxidation indicator. The rancid flavor is initially detected in meat items with TBA levels of 0.5 to 2.0 (Raharjo and Sofos, 1993; Mahmoud, 2013).

Table (4): Thiobarbituric acid (TBA) values (mg malonaldehyde/Kg) of pastrami treated with different preservatives during storage at 4±1°C for 30 days.

Treatments / Storage period	T1	T2	T3	T4	LSD 5%
0d	$^{J}0.32^{A}$	$^{C}0.32^{A}$	$^{FG}0.32^{A}$	$^{B}0.32^{A}$	**NS**
3d	$^{I}0.356^{A}$	$^{H}0.2^{C}$	$^{H}0.287^{B}$	$^{I}0.2^{C}$	**0.004**
6d	$^{H}0.397^{A}$	$^{G}0.212^{CD}$	$^{HG}0.297^{B}$	$^{HI}0.209^{D}$	**0.003**
9d	$^{H}0.415^{A}$	$^{F}0.233^{C}$	$^{HG}0.302^{B}$	$^{HG}0.212^{D}$	**0.008**
12d	$^{G}0.455^{A}$	$^{FE}0.24^{B}$	$^{FG}0.311^{C}$	$^{G}0.223^{D}$	**0.007**
15d	$^{F}0.489^{A}$	$^{E}0.241^{CD}$	$^{FE}0.396^{B}$	$^{F}0.234^{D}$	**0.012**
18d	$^{E}0.542^{A}$	$^{D}0.259^{C}$	$^{E}0.453^{B}$	$^{E}0.245^{D}$	**0.011**
21d	$^{D}0.62^{A}$	$^{D}0.265^{CD}$	$^{D}0.526^{B}$	$^{D}0.262^{D}$	**0.017**
24d	$^{C}0.765^{A}$	$^{C}0.355^{C}$	$^{C}0.574^{B}$	$^{C}0.299^{D}$	**0.01**
27d	$^{B}0.856^{A}$	$^{B}0.406^{C}$	$^{B}0.655^{B}$	$^{B}0.343^{D}$	**0.018**
30d	$^{A}0.969^{A}$	$^{A}0.431^{D}$	$^{A}0.757^{B}$	$^{A}0.435^{CD}$	**0.017**
LSD 5%	**0.0188**	**0.0076**	**0.012**	**0.0147**	

T1: control sample, T2: nitrite 200ppm, T3: thyme 0.5%, T4: smoke liquid 1%.

Data within the same raw with different letters on the right side denote significant differences (P≥ 0.05) according to LSD.

Data within the same column with different letters on the left side denote significant differences (P≥ 0.05) according to LSD.

Changes in peroxide values of pastrami treated with different preservatives during storage at 4±1°C for 30 days:

The changes in peroxide values of pastrami samples treated with different preservatives during storage at 4±1°C for up to 30 days, as shown in Table (19). Peroxide values in all pastrami treatments tended to increase significantly as the cold storage period progressed. Until day 12, then peroxide values decreased in all treatments after day 12 except for the samples formulated with smoke liquid 1%, which increased until day 15. These results are consistent with those obtained by Swastawati *et al.*, (2019). According to the data in Table (5), the peroxide value (PV) was 0.26 m. equiv. /kg fat at day zero. Additionally, the control sample and samples treated with nitrite 200ppm, thyme 0.5%, and smoke liquid 1% were found to be 1.55, 0.23, 1.47 and 2.77 m. equiv. /kg fat; respectively, after 3 days of preservation. At the end of cold storage (30 days), peroxide values reached 2.43 m. equiv. /kg fat for the control sample, and changed to 2.66, 0.96 and 2.75 m. equiv. /kg fat, for samples manufactured with nitrite 200ppm, thyme 0.5%, and smoke liquid 1%; respectively. Furthermore, the peroxide value increased in all treatments from day 3 to day 12, except for samples treated with 1% smoke liquid, which increased till day 15. The peroxide value dropped in all samples at the end of the storage period, but the samples made with

smoke liquid 1% had the highest peroxide value (2.75 m. Equiv. /kg fat) at 30 days. This result was in line with Swastawati *et al.,* (2019), who mentioned that peroxide values gradually increased till day 4 then decreased until day 10. Except for samples formulated with thyme 0.5%, it could be inferred that the addition of various preservatives to pastrami samples decreased peroxide levels compared to samples without preservatives (control samples). This could be due to the low levels added from this preservative, affecting its antioxidant ability refence.

Table (5): Changes in peroxide values (m. Equiv. /kg fat) of pastrami samples treated with different preservatives during storage at 4±1°C for 30 days.

Treatments / Storage period	T1	T2	T3	T4	LSD 5%
0d	$^{E}0.26^{A}$	$^{H}0.26^{A}$	$^{G}0.26^{A}$	$^{G}0.26^{A}$	**NS**
3d	$^{F}1.55^{A}$	$^{H}0.23^{B}$	$^{EF}1.47^{AB}$	$^{F}2.77^{A}$	**1.371**
6d	$^{C}4.5^{A}$	$^{G}1.21^{C}$	$^{CD}3.1^{B}$	$^{CED}3.77^{AB}$	**0.495**
9d	$^{C}4.94^{A}$	$^{C}4.28^{A}$	$^{B}4.49^{A}$	$^{CBD}4.6^{A}$	**NS**
12d	$^{A}7.78^{A}$	$^{A}7.67^{A}$	$^{A}6.5^{A}$	$^{CB}4.7^{B}$	**1.783**
15d	$^{B}6.33^{A}$	$^{B}5.77^{B}$	$^{CB}4.18^{C}$	$^{A}6.58^{A}$	**0.456**
18d	$^{C}4.62^{A}$	$^{D}3.64^{B}$	$^{ED}2.2^{C}$	$^{B}4.99^{A}$	**0.427**
21d	$^{D}3.25^{B}$	$^{ED}3.3^{AB}$	$^{EDF}1.9^{C}$	$^{FED}3.69^{A}$	**0.404**
24d	$^{ED}2.99^{AB}$	$^{EF}2.9^{B}$	$^{EF}1.62^{C}$	$^{FE}3.37^{A}$	**0.467**
27d	$^{ED}2.9^{A}$	$^{F}2.84^{A}$	$^{EF}1.48^{B}$	$^{FE}3.1^{A}$	**0.304**
30d	$^{E}2.43^{A}$	$^{F}2.66^{A}$	$^{F}0.96^{B}$	$^{F}2.75^{A}$	**0.523**
LSD 5%	**0.622**	**0.410**	**1.236**	**1.001**	

T1: control sample, T2: nitrite 200ppm, T3: thyme 0.5%, T4: smoke liquid 1%.
Data within the same raw with different letters on the right side denote significant differences (P≥ 0.05) according to LSD.
Data within the same column with different letters on the left side denote significant differences (P≥ 0.05) according to LSD.

Changes in nitrate and nitrite contents of pastrami samples treated with different preservatives during storage at 4±1°C for 30 days:

The obtained results illustrated in Table (6) show residual nitrate levels in pastrami samples with various alternatives after 30 days of storage at 4±1°C, and it states that residual nitrate levels in all samples rose over time. Table (6) indicated that there were significant differences among all treatments and within the storage period. For example, the amounts of residual nitrate in the control sample increased from 53.36 to 121.05 ppm after 30 days of storage. The initial residual nitrate content in the untreated sample (control) was 53.36 ppm. Meanwhile, 60.4; 52.15 and 36.27 ppm were detected for samples treated with nitrite 200ppm, thyme 0.5%, and smoke liquid 1%, respectively. At the end of the storage period, the initial nitrate concentrations for untreated sample (control), nitrite 200ppm, thyme 0.5% and smoke liquid 1% were 121.05; 124.95; 127.26 and 121.91 ppm; respectively. The highest residual nitrate content found at the end of the storage period was in thyme 0.5% with about (127.26 ppm; respectively). It's possible that the residual amount of nitrate detected after the technical process is free nitrite that hasn't been bound to meat components, oxidized to nitrate (Fernandez-Lopez *et al.,* 2008; Eisinaitė *et al.,* 2020). This high content of residual nitrate may also be because the product is sold as a raw salt dry meat product that has not been exposed to heat, and the salt used in the curing process could be contaminated with nitrate salts (Elgazzar *et al.,* 2017). The initial nitrate concentration of the control sample increased

gradually throughout cold storage at 4±1 °C, peaking at 121.05 ppm at the end of storage period. From the results in the same Table pastrami manufactured with 1% smoke liquid and the control sample (no additives) had the lowest initial residual nitrate concentration (121.91 and 121.05 ppm; respectively) at the end of storage period, while other samples treated with other preservatives had a higher nitrate concentration. This low content of residual nitrate may be due to the presence of nitrate in the water and spices used during the manufacturing process (Sebranek and Bacus, 2007; Honikel, 2008; Gürbüz and Güngör, 2020). All samples treated with different preservatives were less than the maximum permissible limit of residual nitrate in cured beef products (150 ppm) after the storage period, according to Directive (2006) the European Parliament and Council Directive (EU) (2006). To compare the change in residual nitrite in pastrami samples over time of storage, nitrite 200 ppm, thyme 0.5%, and smoke liquid 1% were added to the formula of pastrami and pastrami coating to manufacture pastrami product. Table (7) shows the residual nitrite concentrations in pastrami with various alternatives after 30 days of storage at 4±1°C. The residual nitrite levels in all samples decreased over time. These results were in line with (Oz *et al.,* 2021). At the completion of the storage periods, the nitrite content was decreased, and this may be due to nitrite being converted to nitrite oxide, which forms chemicals that impact meat color, nitrosomyoglobin, and aroma (Sanchez Mainar and Leroy, 2015; Bosse *et al.,* 2016). The results in Table (7) demonstrated a substantial difference between all treatments and storage periods. Although residual nitrite in the control sample decreased from 67.02 to 53.88 ppm after storage. Residual nitrite levels in control and all treatments decreased gradually during storage. These findings are in the line with (Sindelar *et al.,* 2007; Aksu *et al.,* 2020), who discovered that the amount of residual nitrite in storage decreases over time. Similar findings were reported by (Ha *et al.,* 2015). The conversion of nitrite into nitric oxide and nitrous oxide in the mixing step and nitrite oxidation to nitrate over time were reported to be responsible for the reduction in residual nitrite levels during processing. Nitrite levels were said to be affected by storage duration and temperature. Because the storage temperature was kept constant (4±1°C) in this investigation, storage duration probably significantly lowered residual nitrite levels. This was consisted with (Ko *et al.,* 2017). The results indicated that, the initial residual nitrite content measured in the control sample (with only salt added) was 68.26 ppm. Meanwhile, for pastrami samples manufactured with nitrite 200 ppm, thyme 0.5%, and smoke liquid 1% 67.02; 67.05 and 63.07 ppm were recorded; respectively. The initial residual nitrite concentrations for samples prepared without additives (control), nitrite 200ppm, thyme 0.5% and smoke liquid 1% were 54.06; 53.88; 48.67; 55.45 ppm; respectively, at the end of the storage periods. The initial residual nitrite concentration of the control sample decreased gradually during cold storage at 4±1 °C, reaching its lowest value (54.06 ppm) at the end of the cold storage periods. In terms of these findings, pastrami samples made with 0.5% thyme EOs had the lowest initial residual nitrite concentration (48.67 ppm) at the end of cold storage. In contrast, those with other preservatives had greater initial residual nitrite concentration. This decrease in residual nitrite content may be due to its conversion to other compounds such as nitric oxide or nitric acid (Govari and Pexara, 2015). The antioxidant properties of thyme occur because of its high content of phenols (Sedki *et al.,* 2020). The highest content of nitrite found at the end of the storage period was in pastrami sample preserved with 1% smoke liquid with nitrite content of 55.45 ppm. The pH of the medium, the time and temperature of processing, the microbial load present in the raw material, and the addition of antioxidants all have a role in reducing nitrite to nitric oxide at the end of the production process (Ras *et al.,* 2018). The residual amounts of nitrite and nitrate found in the current study were comparable to those seen in commercial items (Lee, 2018). The higher residual nitrite content in traditionally coated pastrami could be attributed to the higher weight loss, resulting in less available nitrite water to form nitrous acid

and nitric oxide (Abdallah *et al.*, 2018). All samples treated with various preservatives at the completion of the storage period had residual nitrite levels below the EOS-mandated (2010) limit of 100 ppm for cured beef products.

Table (6): Changes in Nitrate content (ppm) of pastrami treated with different preservatives during storage at 4±1 °C for 30 days.

Treatments / Storage period	T1	T2	T3	T4	LSD 5%
3d	$^{J}53.36^{BC}$	$^{J}60.4^{A}$	$^{H}52.15^{C}$	$^{J}36.27^{D}$	**1.911**
6d	$^{I}65.41^{A}$	$^{I}68.28^{A}$	$^{G}65.24^{A}$	$^{I}55.85^{B}$	**4.485**
9d	$^{H}76.35^{B}$	$^{H}90.1^{A}$	$^{F}72.63^{C}$	$^{H}65.51^{D}$	**2.273**
12d	$^{G}90.96^{B}$	$^{G}101.34^{A}$	$^{E}86.87^{C}$	$^{G}83.2^{D}$	**2.668**
15d	$^{F}96.63^{B}$	$^{F}108.74^{A}$	$^{E}90.52^{C}$	$^{F}86.88^{D}$	**2.063**
18d	$^{E}103.1^{B}$	$^{E}112.46^{A}$	$^{D}101.31^{C}$	$^{E}92.91^{D}$	**1.575**
21d	$^{D}108.54^{C}$	$^{D}116.14^{A}$	$^{C}110.35^{B}$	$^{D}101.51^{D}$	**2.0057**
24d	$^{C}112.62^{C}$	$^{C}120^{A}$	$^{B}118.27^{B}$	$^{C}106.13^{D}$	**1.447**
27d	$^{B}117.62^{C}$	$^{B}122.2^{B}$	$^{A}126.34^{A}$	$^{B}113.39^{D}$	**1.797**
30d	$^{A}121.05^{D}$	$^{A}124.95^{B}$	$^{A}127.26^{A}$	$^{A}121.91^{CD}$	**1.668**
LSD 5%	**1.588**	**2.1075**	**3.8851**	**2.1758**	

T1: control sample, T2: nitrite 200ppm, T3: thyme 0.5%, T4: smoke liquid 1%.
Data within the same raw with different letters on the right side denote significant differences (P≥ 0.05) according to LSD.
Data within the same column with different letters on the left side denote significant differences (P≥ 0.05) according to LSD.

Table (7): Changes in nitrite content (ppm) of pastrami sample treated with different preservatives during storage at 4±1°C for 30 days.

Treatments / Storage period	T1	T2	T3	T4	LSD 5%
3d	$^{A}68.26^{A}$	$^{A}67.02^{B}$	$^{A}67.05^{B}$	$^{A}63.07^{C}$	**0.44**
6d	$^{B}64.58^{A}$	$^{B}65.8^{B}$	$^{B}62.77^{C}$	$^{B}62.06^{D}$	**0.44**
9d	$^{C}62.73^{A}$	$^{C}62C^{AB}$	$^{C}60.25^{C}$	$^{B}61.81^{B}$	**0.76**
12d	$^{D}60.78^{C}$	$^{C}62.46^{A}$	$^{D}59.16^{D}$	$^{C}60.85^{BC}$	**1.16**
15d	$^{E}58.87^{BC}$	$^{D}60.05^{A}$	$^{E}57.23^{D}$	$^{ED}58.46^{C}$	**1.05**
18d	$^{E}58.97^{BC}$	$^{D}60.55^{A}$	$^{E}56.83^{D}$	$^{D}58.64^{C}$	**0.87**
21d	$^{FE}58.6^{BC}$	$^{ED}59.93^{A}$	$^{E}57.04^{D}$	$^{EF}57.77^{CD}$	**1.07**
24d	$^{FG}57.74^{BC}$	$^{EF}59.18^{A}$	$^{FE}56.65^{D}$	$^{GF}56.96^{CD}$	**0.98**
27d	$^{G}57.57^{AB}$	$^{F}58.42^{A}$	$^{F}55.97^{C}$	$^{G}56.49^{BC}$	**1.15**
30d	$^{H}54.06^{BC}$	$^{G}53.88^{C}$	$^{G}48.67^{D}$	$^{H}55.45^{A}$	**0.70**
LSD 5%	**0.86**	**0.77**	**0.86**	**0.86**	

T1: control sample, T2: nitrite 200ppm, T3: thyme 0.5%, T4: smoke liquid 1%.
Data within the same raw with different letters on the right side denote significant differences (P≥ 0.05) according to LSD.
Data within the same column with different letters on the left side denote significant differences (P≥ 0.05) according to LSD.

Changes in N-nitrosamine content of pastrami treated with different preservatives during storage at 4±1°C for 30 days:

In terms of health, it's considered that when residual nitrite levels are low, nitrosamine production in meat products is also low. According to Manar *et al.,* (2014), the nitrosamine content of beef sausage (0.29 g / 100 g) with 125 ppm nitrite was lower than that of beef sausage (0.11 g / 100 g) supplemented with 10% beetroot juice. Because the presence of nitrosamine in meat products is primarily associated with high cooking temperatures (> 200 °C) rather than nitrite concentration (Drabik-Markiewicz *et al.,* 2009), N-nitrosamine molecules did not appear in all treatments. This outcome was in accordance with (Jin *et al.,* 2018; IARC, 2010). These results may also be due to antioxidants like ascorbic acid added to all samples having an oxidation-reduction characteristic that aids in reducing nitrosating substances to NO (Lidder and Webb, 2013; Milkowski *et al.,* 2010).

Organoleptic evaluation of pastrami treatments:

External appearance, color, taste, flavor, and overall acceptability of pastrami containing different preservatives (nitrite 200 ppm, thyme 0.5%, and smoke liquid 1%) were evaluated in comparison to control during refrigerated storage at 4±1°C. Table (8) shows the average scores for organoleptic properties during cold storage. The effects of various preservatives on the appearance of pastrami are shown in Table (8). The data showed no discernible variance in the external appearance scores following processing across all samples, with the control sample scoring 7.79. Meanwhile, for samples treated with nitrite 200 ppm, thyme 0.5%, and smoke liquid 1% recorded 8.14; 6.79 and 7.32; respectively. The color score for all prepared pastrami treatments was convergent at zero time, as shown in Table (8) by the maximum color score of 8.14 for nitrite 200 ppm, which was the highest color score. Meanwhile, the results for pastrami treated with just salt (control), thyme 0.5%, and smoke liquid 1% recorded 7.89; 7.14 and 7.04; respectively. The color score of pastrami made with 1% smoke liquid had the lowest color score of all the treatments. However, there were no significant variations ($P \geq 0.05$) in color scores between all pastrami samples and the control sample (7.89). Table (8) demonstrated that the taste scores for all prepared pastrami treatments were convergent at zero time, with the highest taste score being 7.82 for pastrami sample treated with nitrite 200 ppm. Meanwhile, for pastrami treated with only salt (control), thyme 0.5%, and smoke liquid 1% it recorded 7.64, 6.00 and 7.21; respectively. The pastrami prepared with 0.5% thyme had the lowest taste score of all the treatments. However, no significant differences ($P \leq 0.05$) were found in taste scores of all pastrami samples except for the sample mixed 0.5% thyme essential oil (EOs) with the control sample (7.64). Table (8) shows the influence of various preservatives on the flavor of pastrami. The data show that there was a discernible difference in Flavor scores across all samples in the following processing, with the control sample (with only salt added) scoring 7.93. Likewise, for samples treated with nitrite 200ppm, thyme 0.5%, and smoke liquid 1% it recorded 7.71; 6.39 and 7.00; respectively. Moreover, the control sample received the best grades, while those containing 0.5% thyme essential oils (EOs) received the lowest. Table (8) shows that there were no significant differences ($P \geq 0.05$) in overall acceptability scores of pastrami samples mixed with nitrite 200 ppm, thyme 0.5% and smoke liquid 1% when compared to the control sample, which scored (8.21). Furthermore, the highest overall acceptability score in all treatments was (8.07) for nitrite 200ppm. Meanwhile, for pastrami formulated with thyme 0.5%, and smoke liquid 1%, it recorded 6.64 and 7.61; respectively. The overall acceptance score of pastrami treated with thyme 0.5% was the lowest of all treatments, with no significant difference between them.

Table (8): Organoleptic evaluation of pastrami treated with different preservatives during storage at 4±1°C for 30 days.

Treatments	External appearance	Color	Taste	Flavor	Overall acceptability
T1	7.79	7.89	7.64 ^{a}	7.93 ^{ab}	8.21 ^{a}
T2	8.14	8.14	7.82 ^{a}	7.71 ^{a}	8.07 ^{ba}
T3	6.79	7.14	6.00 ^{b}	6.39 ^{b}	6.64 ^{c}
T4	7.32	7.04	7.21 ^{ba}	7.00 ^{ba}	7.61 ^{bac}
LSD 5%	**Ns**	**Ns**	**1.24**	**1.27**	**1.23**

T1: control sample, T2: nitrite 200ppm, T3: thyme 0.5%, T4: smoke liquid 1%.
The values with different letters denote significant differences ($p<0.05$) according to LSD.

CONCLUSION

Because of meat and meat products' complex makeup of saturated and unsaturated fats, carbohydrates, proteins, vitamins, and pigments, they are sensitive to microbiological and biochemical deterioration, especially during storage. The use of sodium nitrite in the meat and poultry business has considerably helped the sector by creating products with distinct colors, textures, and flavors and enhanced food safety, and a longer shelf life with superior storage stability. However, nitrite for curing was nearly outlawed in the 1970s due to a heated public controversy about the possibility of producing carcinogenic nitrosamines. As a result, both industry and government have made many efforts to limit the danger of nitrosamine development and alleviate potential human health issues. So, they saw that natural preservatives are far better for both humans and the environment than synthetic preservatives, which can cause health problems. To boost functional properties, various ingredients such as EOs, and natural extracts with antioxidant properties have been added to raw meat products to reduce nitrite residual and its risks on human health. Finally, the use of 1% smoke liquid for preserving meat products made the best-preserving properties (TBA value and peroxide value), lowering the residual nitrite and nitrate content with significant inhibition for pathogenic bacteria, and all the studied characteristics were within the permissible limits stated by the Egyptian organization for standardization and quality control at the end of the storage period.

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