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ANTIOXIDANT EFFECT OF FAT-SOLUBLE ROSEMARY AND GREEN TEA EXTRACTS ON STORAGE PERIOD PROLONGATION OF MEAT PASTE

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Abstract

Intensive development of the food market necessitates its research, structuring in order to identify ways to create new products with improved quality characteristics, in particular meat pates, and prolongation of their storage period by adding fat-soluble rosemary and green tea extracts.

Meat pates are manufactured using liver and blanched chicken fillet, pork fat blend and refined deodorized sunflower oil with extracts from Danisco (Denmark) in an amount of 10%. The following blends have been developed: blend (50 : 50) of pork fat and refined deodorized sunflower oil with the rosemary extract addition of 0.1%; blend (50 : 50) of pork fat and refined deodorized sunflower oil with the green tea extract addition of 0.05% and blend (50 : 50) of pork fat and refined deodorized sunflower oil with the green tea and rosemary extract addition of 0.15%. Sensory evaluation research of hydrolytic and oxidative processes during products storage have been carried out for aim achievement, namely, the changes in quality indicators of the test samples were recorded during the storage period until the critical value of the peroxide number (10 mmol½O/kg) was reached. The acid number has been determined as an indicator of hydrolytic putrefaction degree.

Compositions and technologies for the blends production using plant extracts of various concentrations for meat pates have been developed. The oxidative processes dynamics and resistance to damage during products storage have been determined. The storage period of all blends is 50 days, against the generally accepted storage period for fats in an open container - 30 days. The research results have confirmed that the quality indicators of the products are attributable to the specified requirements.

The antioxidant and bactericidal properties of fat-soluble extracts have been established. It has been proven that the use of rosemary and green tea extracts, and especially their combination, helps to increase the meat pates stability, expand the product line and increase their safety for consumers.

***Key words**: Meat pastes, Fat-soluble green tea extract, Fat-soluble rosemary extract, Blend, Oxidation.*

1. Introduction

Researches by many authors indicate the ability of some plants and herbal preparations to stabilize fats of plant and animal origin, showing next to antioxidant properties, also an antibacterial effect [1]. Often used extracts of various plants (spices, sage, cranberries, sea-buckthorn, etc.), which are prepared immediately before adding or use ready-made preparations - phyto extracts. These are ecologically and microbiologically

clear products which are standardized for the active substances content, in contrast to natural spices and plants. The presence of biological and functional-technological action in these substances - not only provides a technological effect, but also provides a protective and general strengthening effect on the consumer health [2]. The use of commercial preparations is more convenient in industrial conditions, because it allows to avoid varying the extracts quality, fluctuations in the active substances content in them, standardize the process and increase industrial efficiency. Addition of extracts also contributes to increasing the biological value of products due to the phytopreparations of essential oils and bioflavonoids content in them [3].

Fat raw materials (vegetable oils and animal melted fats) are a complex multicomponent system, the basis of which are triacylglycerols. Fatty acids with different chain lengths, degrees of unsaturation and isomerism are part of triacylglycerols. The availability of double bonds in fatty acids explains their high reactive capacity, especially with regard to molecular oxygen. The interaction of triacylglycerols with oxygen leads to various destructive changes in triacylglycerols with the formation of a large number of physiologically negative products. To effectively slowing the oxidation process, the necessary step is to investigate its chemistry with various initiation methods [4].

Oxidation stability is an important quality indicator and storage period of fats, whereas the oxidation products are low-molecular substances with bad smell and taste, which make it impossible to use or consume that kind of fats. The process of changing the properties of edible fats is based on radical oxidation reactions, in particular the peroxy radical reactions. Peroxy radicals cause oxidative destruction of organic compounds, which can be prevented using natural and synthetic inhibitors [3].

In recent years, model oxidation reactions of oleic, linoleic, and linolenic acids have been used to test antioxidants. However, the study of natural systems oxidation in some cases seems more appropriate. It has been found that the process of free radical oxidation reactions in model systems depends on the particles size in which the oxidation process is studied. In such systems, the specific tocopherol concentration in lipids is so high that it can take part in chain extension, speeding up the process. The process research of fatty raw materials oxidation is the key importance to develop control methods for their quality, storage conditions and establishing pull-date of the product [5].

One of the simple and effective methods of stopping oxidative fat damage (especially during storage) is the addition of substances which slow down this process. Such substances are oxidation inhibitors or antioxidants. The identification of their action is increasing the induction period and reducing the oxidation rate. A large amount of antioxidants is widely used in the fat-and-oil industry. Each of them in a certain way affects the oxidation process induction period prolongation in various types of fats. There are no universal antioxidants that are equally effective for any fat, therefore, it is good to determine their effectiveness experimentally. Preference should be given to natural antioxidants, their use is recommended for stabilizing edible fats according to the requirements of the Ministry of Health of Ukraine [6].

Intensive development of the food market determines the need for its research, structuring in order to identify ways to create new products with improved quality characteristics. Accordingly, the quality issue of meat products, in particular pastes produced and sold in Ukraine, is relevant [6]

In paste formulations, one of the important components, which are responsible for the structure and consistency of the product is fatty raw materials (bacon, fatty pork). Reducing calorie pastes by replacing animal fats with vegetable oils, through the use of plant components, milk proteins or raw materials with a low content of adipose tissue remains relevant. Moreover, fats must satisfy the following basic requirements: be characterized by nutritional value, have high organoleptic properties and have a sufficiently high stability during storage and cooking [7].

The aim of this research was development of a formulation and technology of blends and meat pates using plant extracts of different concentrations, namely rosemary extract (0.1%), green tea extract (0.05%) and their mixture (0.15%) in order to increase the products stability during storage period.

2. Materials and Methods

Object of research were meat pates manufactured using liver and blanched chicken fillet, pork fat blend and refined deodorized sunflower oil with extracts from Danisco (Denmark) in an amount of 10%. The following blends have been developed: blend (50 : 50) of pork fat and refined deodorized sunflower oil with the rosemary extract addition of 0.1%; blend (50 : 50) of pork fat and refined deodorized sunflower oil with the green tea extract addition of 0.05% and blend (50 : 50) of pork fat and refined deodorized sunflower oil with the green tea and rosemary extract addition of 0.15%.

Sensory research on meat pates with rosemary extract, green tea extract extract and their mixturewas carried out by a taste panel of 10 people. Physico-chemical quality indicators were determined by standard methods.

Oxidative stability of blends was carried out under various storage conditions:

1) The accelerated method of "active oxygen" was carried out according to the state standard ISO 6886-2003 "Animal fats and vegetable oils: Determination of oxidation resistance (Accelerated oxidation test) (ISO 6886: 1996, IDT)". The method is based on blowing air at a constant speed through a fat layer at constantly elevated temperature and determining the degree of fat oxidation at certain intervals. 100 g of the blend was added to the well and placed in a water bath at a temperature of 75 ± 2 ^{0}C. Air began to be supplied at a rate of 8 L/h after setting the temperature. The control was a sample of oil and pork fat without antioxidants. The datum value of the peroxide, acid, and anisidine numbers of all samples was measured and at certain time intervals (for determining the PN - 1 hour, AN - 2 hours, AnN - 3 hours). At set intervals, 1 mL of the sample was taken from the well into a prepared flask with a ground stopper and, the flask was re-weighed after cooling. The mass of the sample was found by difference, and used to determine oxidation indices.

2) At room temperature with free light and air access (auto-oxidation). Blend samples were stored in glass cups at a temperature of 24 ± 2 ^{0}C. Native fats without adding oils to them were used as a control sample. During storage, samples were taken every 10 days to determine the peroxide and acid numbers.

3) In low-temperature storage conditions without light access. Samples of vegetable oils, pork fat, control and blended samples, were stored at a temperature of minus 6 ± 0.5 ^{0}C. During storage, samples were taken every month for analysis and the peroxide and acid numbers were determined.

Blend oxidation was stopped when the peroxide number reached a value more than 10 mmol½O/kg, and the acid number reached more than 6 mg KOH/h.

Oxidation induction time of the samples was calculated to establish the blends antioxidant activity. Oxidation induction time is the time during which a significant increase in the concentration of oxidation products occurred. Antioxidant activity is the total braking effect, which is due to a combination of elementary initiation reactions, lengthening and breaking of chains. The duration of fat oxidation before the end of induction period was determined by kinetic oxidation curves. The end of induction period is the peroxides content at the level of 2.5 mmol½O/kg (by peroxide number), the beginning of free fatty acid accumulation (by acid number) and an increase concentration of secondary oxidation products (by anisidine number). The effectiveness of the antioxidant effect in vegetable oil, pork fat and blends was determined for a specified oxidation induction time of the test samples.

The studies were repeated three times and results were statistically processed using Microsoft Excel Statistics 2007 to provide accuracy of the obtained results.

3. Results and Discussion

Along with calorie reduction of meat and meat-containing paste, an important problems are their quality and safety. Various chemical preserving agents, antioxidants and biologically active substances of natural origin are used to increase the safety level of meat products and prolong storage period.

It is known that an increase in the consumption of meat products is associated with an increase in saturated fat intake, which negatively affects the physiological functions of the body. According to the FAO/WHO, fat intake per person over 30 years in the world has increased from 53 g/day to 73 g/day, and in Europe from 117 g/day to 148 g/day [8]. The most advanced and effective way to improve the fatty acid composition of meat products is the use of protein-fat emulsions enriched with oils. This method allows to control the addition level of biologically significant components, to regulate their content and the raw material composition of the products, depending on application.

An important direction of scientific research and applied work is the use of vegetable oils in the technology of meat products. The research to improve the quality of meat products by addition vegetable oils into the recipe has been performed in the meat industry for a long time. At the first stage, this process was due to a shortage

of animal fats, then to the low cost of vegetable oils production, and more recently, the desire to create products for a balanced diet [9].

In the last few years, research and practical approbation in the application field of CO_2- spices extracts in the meat industry has expanded significantly [10]. Compared to extracts obtained with other solvents, CO_2- extracts preserve all biologically active substances as much as possible; they are sterile and have bactericidal effect on the product microflora; they convey the taste and flavor of the product from which they had been obtained; contain natural preserving agents and antioxidants.

Great experience in the meat industry is the use of fat-soluble plant extracts containing antioxidant and antimicrobial components. Phenolic diterpenes, carnosol and carnosic acid of a fat-soluble rosemary extract are able to utilize hydroxy- and peroxyl lipids radicals and form chelate complexes with metal ions, for example, with iron, which a significant degree stabilizes the lipid fraction of meat products [11].

The preventive properties of aromatic plants are due to the presence in their composition of many biologically active substances, which entering to the body, exhibit physiologically active properties. Pharmacologists and representatives of alternative medicine are widely studying methods of using essential oil raw materials

and essential oils [12]. Domestic and foreign scientists from commodity research are studying the flavonoids antioxidant effect of: eucalyptus, hypericum, yarrow, dill, parsley, calamus, coriander, caraway, sage and other essential oil raw materials and their effect on the prolongation of food storage. They proved that these additives not only have negative impact on the human body, but also prolong the foods storage period due to their antioxidant properties.

Essential oil and spicy aromatic raw materials not only give a pleasant flavor and taste to consumer goods, but also have a preventive and therapeutic effect on the human body. Namely: it strengthens the nervous system, normalizes coronary circulation, relieves inflammation and hydrops, disinfects, stimulates physical and mental activity, et cetera. Recently, as mentioned above, modern enterprises, in pursuit of profit, this valuable raw material change with synthetic preserving agents, taste and flavor improvers and do not pay enough attention to the useful properties of aromatic raw materials extracts [13].

However, the addition of these antioxidants into meat systems is associated with such difficulty as the impossibility to evenly apportion a small amount of aromatic substances in a large mass of minced meat. In this regard, premixing extracts with other natural spices, sugar or the preparation of aqueous emulsions is recommended.

Essential oil and spicy aromatic raw materials are a valuable addition to perfumes and cosmetics and food products and can not only improve the taste and prolong the storage period of the latter, but also have a preventive effect on the human body.

However, a more extensive study of its possibilities to use is necessary. To selecting most effective extracts for meat products, the total content of lipophilic (fat-soluble) and hydrophilic (water-soluble) constituents of antioxidant extracts from various plants (Table 1) has been determined by amperometric method (Yashin *et al.*, [3]).

The highest antioxidants content, both fat-soluble and water-soluble, has been determined in samples of rosemary extracts (producer - Kalsec, Danisco, and Kemin), and green tea (Danisco).

The addition level of the studied biologically active substances, with which their effective use as antioxidants for fat systems is ensured, has been determined by preliminary experimental research. It has been established that effective concentrations are 0.1% for rosemary fat-soluble extract and 0.05% fat-soluble green tea extract.

In consideration of the foregoing premises, it was interesting to study the antioxidant stability of pork fat, refined deodorized sunflower oil and their blends with antioxidant additives of rosemary and green tea, from the point of view of free radical oxidation theory. Rose-

Table 1. The total content of fat-soluble and water-soluble antioxidants in plant extracts

Extracts name, manufacturing companies	Total antioxidant content	
	fat-soluble (TCFA), mg/g, standard - gallic acid	water-soluble (TCWA); mg/mL, standard - quercetin
Sage extract (Aquaresin: Sage; Dalmatian; NS), Kalsec	1.4 ± 0.08	1.2 ± 0.06
Rosemary extract (Herbalox® TYPE XT-W), Kalsec	9.9 ± 0.1	7.6 ± 0.1
Rosemary extract (Aquaresin Rosemary, NS); Kalsec	4.3 ± 0.08	0.12 ± 0.02
Rosemary extract (Guardian™ Rosemary Extract); Danisco	7.3 ± 0.11	21.3 ± 0.09
Green Tea Extract (GUARDIAN™ Green Tea Extract); Danisco	82.3 ± 0.42	159.663 ± 25
Rosemary extract (EORTIUM™ RG20 liquid); Kemin	48.5 ± 0.17	864 ± 0.1
Rosemary and green tea extract (Natur FORT™ 12 liquid); Kemin	12 ± 0.12	0.93 ± 0.05

Table 2. Formulations of blended fats with fat-soluble rosemary and green tea extracts

№	Vegetable oil blends	Name of oil extract	The amount of oil extract, %
1	**Pork fat (50%) + refined deodorized sunflower oil (50%)**	Fat-soluble rosemary extract	0.1
2	**Pork fat (50%) + refined deodorized sunflower oil (50%)**	Fat-soluble green tea extract	0.05
3	**Pork fat (50%) + refined deodorized sunflower oil (50%)**	Fat-soluble rosemary and green tea extract	0.15

mary and green tea extracts from company "Danisco" (Denmark) with the highest water- and fat-soluble antioxidants content were selected based on the characteristics of selected components from different manufacturers. The body's daily maintenance in vitamins increases under oxidative stress conditions. Therefore, vitamins and especially fat-soluble antioxidant vitamins must be included in the formulation.

The developed blend formulations with fat-soluble extracts are shown in Table 2 and blend production technology is in Figure 1.

Quality indicator changes of the studied samples have been recorded during the storage period until the critical value of the peroxide number (10 mmol½O/kg) had reached. The oxidation induction period was determined as the time to reach the peroxide number of 2.5 mmol½O/kg, when the accumulation rate of peroxidic compounds is negligible. The results, according to the research, are shown in Table 3. The value of the acid number was determined as a degree indicator of hydrolytic damage. The control sample were pork fat and refined deodorized sunflower oil without extracts.

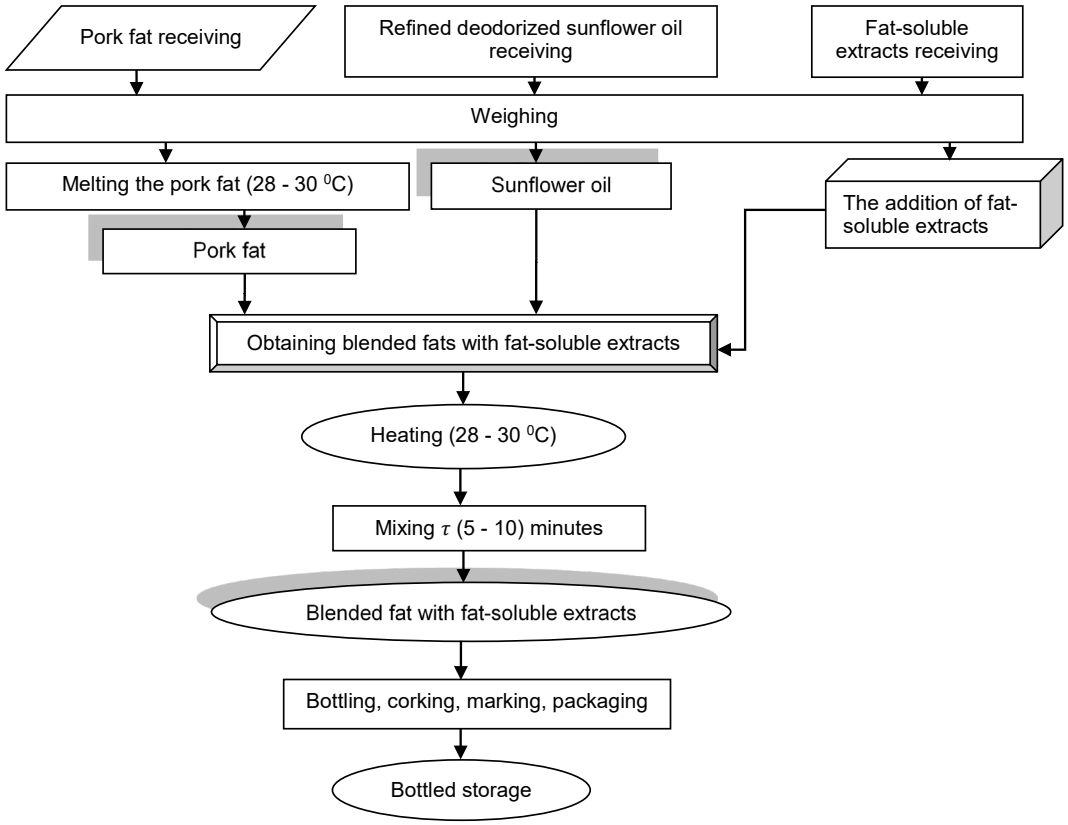

Figure 1. The technological scheme of blended fats with fat-soluble rosemary and green tea extracts

Table 3. Changes dynamics of peroxide and acid numbers of blends with fat-soluble rosemary and green tea extracts during storage

Sample	Oxidation time, days											
	AN, mg KOH/g						PN, mmol½O/kg					
	0	10	20	30	40	50	0	10	20	30	40	50
Pork fat - control sample	0.31	0.5	0.6	0.7	0.9	1.1	1.36	3.2	6.5	12	1.36	-
Refined deodorized sunflower oil - control sample	1.2	1.3	1.4	1.57	2	2.6	1.6	4	13	8.58	-	-
Blend (50 : 50) of pork fat and refined deodorized sunflower oil with the rosemary extract addition	0.83	0.96	1.14	1.42	1.72	2	1.2	1.3	3	5.9	-	-
Blend (50 : 50) of pork fat and refined deodorized sunflower oil with the green tea extract addition	0.84	1.1	1.33	1.6	1.89	2.2	1.1	1.2	3.3	10.3	-	-
Blend (50 : 50) of pork fat and refined deodorized sunflower oil with the green tea and rosemary extract addition	0.82	0.93	1.1	1.3	1.54	1.8	1.2	1.3	2	5	10	14.9

Analysis of the results in Table 3 showed that all samples undergo changes of various depth during the experiment. At the initial moment of oxidation, the values of AN and PN of all the studied samples were close in their values. However, after leaving the induction period, changes in fats become more signified and are characterized by a length of storage period. The chance in peroxide and acid numbers during storage of fatty raw materials under conditions of autooxidation are shown in Figure 2, and Figure 3.

By analyzing the oxidation values at the end of the experiment, it can be argued about the overwhelming activity of green tea and rosemary extract, added together. Specifically, for the sample with the named additive, the PN value on the fortieth day is 10 mmol½O/kg, and the AN is 1.54 mgKOH/g, which is objectively lower than other samples. It should be noted the expressed antioxidant effect of all additives, which is inferior to their complex effect.

The oxidation kinetics research of pork fat, refined deodorized sunflower oil and their blends allowed to quantitatively describe the oxidation stages of fatty raw materials (Table 4). The extracts addition increases the induction period and allows to prolong the storage period of fatty raw materials. Moreover, the slowdown effect of the peroxidation processes is most pronounced with the simultaneous additive adding of green tea extract and rosemary extract. The obtained data correlate well with the results published in scientific sources, when the interaction between antioxidants can be synergistic.

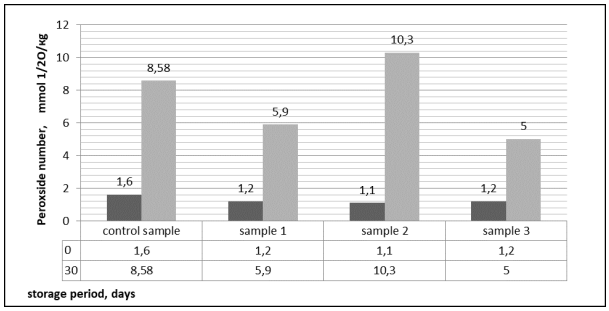

Figure 2. Change in PN during autooxidation of blends with fat-soluble rosemary and green tea extracts

Control sample - refined deodorized sunflower oil; Sample1 - Blend (50 : 50) of pork fat and refined deodorized sunflower oil with the rosemary extract addition; Sample2 - Blend (50 : 50) of pork fat and refined deodorized sunflower oil with the green tea extract addition; Sample3 - Blend (50 : 50) of pork fat and refined deodorized sunflower oil with the green tea and rosemary extract addition.

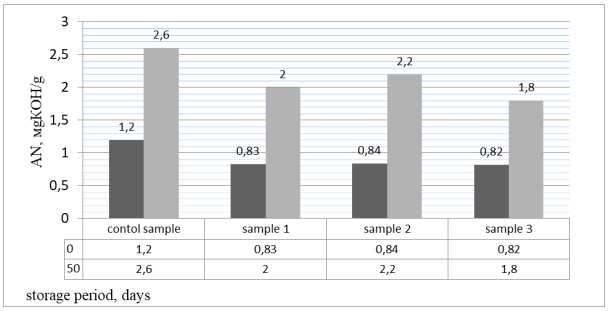

Figure 3. Change in AN during autooxidation of blends with fat-soluble rosemary and green tea extracts

Control sample - refined deodorized sunflower oil; Sample1 - Blend (50 : 50) of pork fat and refined deodorized sunflower oil with the rosemary extract addition; Sample2 - Blend (50 : 50) of pork fat and refined deodorized sunflower oil with the green tea extract addition; Sample3 - Blend (50 : 50) of pork fat and refined deodorized sunflower oil with the green tea and rosemary extract addition.

Table 4. Oxidation indices of blends with fat-soluble rosemary and green tea extracts and without extracts

Sample	Oxidation stage	
	Induction period, days, to PN 2.5 mmol½O/kg	Storage period, days, to PN 10 mmol½O/kg
	Without extracts	
Pork fat	12.5	24.9
Refined deodorized sunflower oil	7.4	10.9
Blend (50 : 50) of pork fat and refined deodorized sunflower oil	13	17.8
	With rosemary extract	
Pork fat	23.9	45.3
Refined deodorized sunflower oil	18.9	27.9
Blend (50 : 50) of pork fat and refined deodorized sunflower oil	24.1	33
	With green tea extract	
Pork fat	15	30.6
Refined deodorized sunflower oil	15	22.3
Blend (50 : 50) of pork fat and refined deodorized sunflower oil	23.6	27.5
	With rosemary and green tea extracts	
Pork fat	28.4	49.9
Refined deodorized sunflower oil	26.8	38.9
Blend (50 : 50) of pork fat and refined deodorized sunflower oil	29.7	44.1

Table 5. Formulations of model samples of meat pastes

Name of raw materials	Content, %			
	Control sample	Samples		
		№ 1	№ 2	№ 3
Chicken fillet	15	22	22	22
Chicken liver	30	30	30	30
Pork fat	15	10	10	10
Chicken eggs	4	4	4	4
Onions	10	10	10	10
Carrots	7	7	7	7
Refined deodorized sunflower oil	5	-	-	-
Blend (50 : 50) of pork fat and refined deodorized sunflower oil with the rosemary extract addition	-	10	-	-
Blend (50 : 50) of pork fat and refined deodorized sunflower oil with the green tea extract addition	-	-	10	-
Blend (50 : 50) of pork fat and refined deodorized sunflower oil with the green tea and rosemary extract addition	-	-	-	10
Milk powder	2	2	2	2
Wheat fiber	12	5	5	5
Total amount	100	100	100	100

It can be seen from the research, that the oxidation flow data have been obtained quite consistent. The oxidation induction period is 26 - 29 days for all the test samples, which indicates the same effectiveness of the added extracts for various substrates. The storage period for all fats reaches 50 days, against the generally accepted storage period for fats in an open container is 30 days.

The oxidative stability of the fat component for meat products which we have studied, is an important quality indicator and storage period of finished products. In oxidation time, the low molecular weight decomposition products with an unpleasant flavor and taste are formed, which negatively affect the product consumer properties. It has been established that the addition of rosemary and green tea extracts have an active effect on the fat stability to oxidative damage processes under conditions of autooxidation. Therefore, it has been chosen for future introduction in the formulation of meat products, namely meat pastes.

The formulation for the control sample of meat paste was formulated with liver and scalded chicken fillet, pork fat, onions, fried carrots, skimmed milk powder, chicken eggs, wheat fiber, and refined sunflower oil. Paste model samples were made according to similar formulations, but rosemary and green tea extracts, their mixture were added to the fat base, which consisted of a blend of pork fat and refined deodorized sunflower oil. Formulations of model samples of meat pastes are shown in Table 5.

The technological scheme for the meat pastes production using rosemary and green tea extracts is given below in Figure 4.

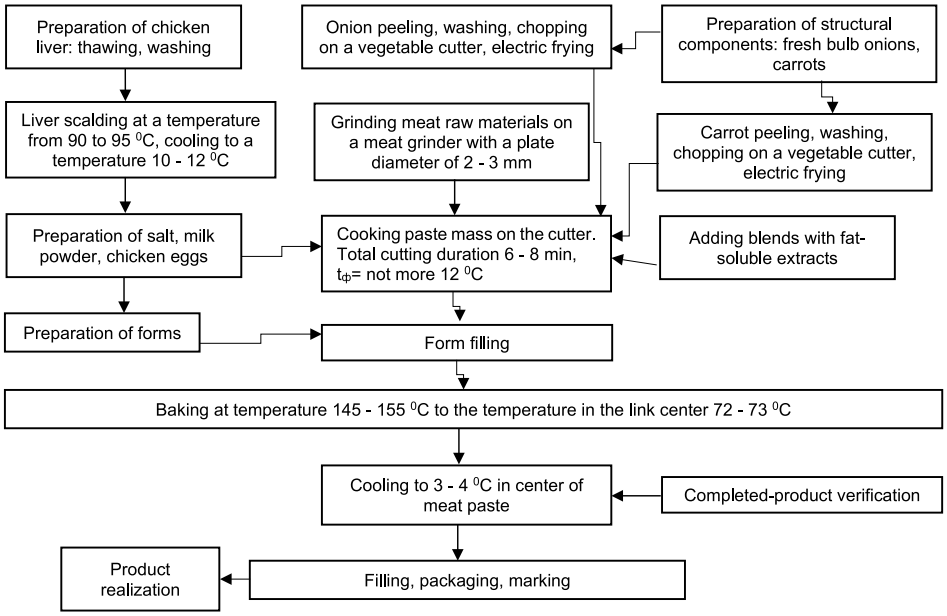

Figure 4. The technological scheme for the meat pastes production using rosemary and green tea extracts

In order to identify the full "flavor" profile, sensory evaluation on a five-point scale of meat pastes has been carried out by employees of the Department of meat and meat products technology - National University of Food Technologies. The following indicators: taste, colour, flavour, appearance and texture were taken into account. The profilogram is shown in Figure 5.

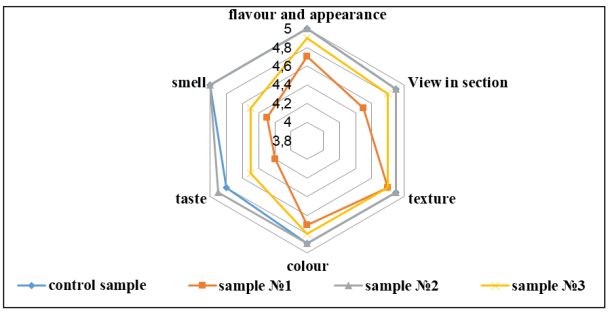

Figure 5. Sensory profile of developed meat pastes with rosemary and green tea extracts

Appearance assessment of the meat paste samples did not identify visible defects. The meat pastes had a crushed, moderately uniform structure and clean, dry surface. Sensory evaluation results of samples of a new type paste meat showed a positive change in the meat pastes consistency with the green tea and rosemary oil extracts addition in the formulations. Also, all members of the taste panel noted the spicy taste and smell of new meat pastes.

The next step was the dynamics assessment of oxidative processes occurring in the fat fraction of the researched meat paste with the addition of rosemary extract, a mixture of rosemary and green tea extracts and control sample during 15 storage days at a temperature 0 - 4 ^{0}C. The research data indicate that the accumulation rate of oxidation products in the developed meat pastes is less than in the control sample, which is characterized by a more intense peroxides accumulation in the first 6 storage days.

The presence of an induction period in the fat fraction oxidation is characteristic for meat pastes with rosemary extract and a mixture of rosemary and green tea extracts in the first 6 storage days.

The peroxide accumulation in the research meat paste samples is significantly accelerated after 6 storage days. However, their accumulation rate is more significant for the control sample. Up to 10 days, the peroxides number in it increases in 2.7 times, while in meat pastes with plant extracts of rosemary and extract mixture of rosemary and green tea in 1.8 times relative to the initial period.

4. Conclusions

- The feasibility of using plant extracts to prolong the storage period of meat pastes has been considered based on literary sources analysis. The presence of antioxidant and bactericidal properties in fat-soluble rosemary and green tea extracts has been established. A synergistic effect towards the accumulation of primary oxidation products has been found. The effect of fat-soluble rosemary and green tea extracts on the lipids stabilization of refined deodorized sunflower oil and meat pastes has been studied.

- Different extract characteristics have been studied and their properties have been studied, with the view to use them in meat paste formulations to increase the product stability during storage.

- Formulations and manufacturing techniques of blends and meat pastes using plant extracts of various concentrations have been developed, namely rosemary extract (0.1%), green tea extract (0.05%) and their mixture (0.15%).

- It has proved that the use of rosemary and green tea extracts and their combination helps to increase the biological value of products, expands the meat paste range and increases their stability during storage. They have good sensory characteristics.

- The dynamics of oxidative processes and spoilage resistance during meat paste storage have been determined. The limiting storage period for all fats reaches 50 days against the generally accepted storage period for fats in an open container, which is 30 days. Based on research, it has found that all indicators are within the marks.

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