Original Research Article: full paper

(2022), «EUREKA: Life Sciences»
Number 1

STUDY OF EFFICIENCY OF BERRY EXTRACTS IN THE TECHNOLOGY OF SEMI-SMOKED SAUSAGES

Vasyl Pasichnyi
Department of Technology of Meat and Meat Products[1]

Nataliia Bozhko
Department of Biophysics, Biochemistry, Pharmacology and Biomolecular Engineering
Medical Institute of Sumy State University
31 Sanatorna str., Sumy, Ukraine, 40018

Vasyl Tischenko
Department of Technology and Food Safety
Sumy National Agrarian University
160 Herasyma Kondratieva str., Sumy, Ukraine, 40021

Andriy Marynin✉
Problem Scientific and Research Laboratory[1]
andrii_marynin@ukr.net

Yevgenia Shubina
Department of Technology of Meat and Meat Products[1]

Roman Svyatnenko
Problem Scientific and Research Laboratory[1]

Oleksandra Haschuk
Department of Technology of Meat and Meat Products[1]

Olena Moroz
Technological Department
Lviv Professional College of Food Technology and Business
30/32 Bortnianskoho str., Lviv, Ukraine, 79039

[1]National University of Food Technologies
68 Volodumurska str., Kyiv, Ukraine, 01601

✉**Corresponding author**

Abstract

The work is devoted to the study of the effectiveness of the use of berry extracts (*Aronia melanocarpa Elliot and Ribes nigrum L.)* in the technology of semi-smoked sausages. The aim of the study was to study the effectiveness of the use of berry extracts (*Aronia melanocarpa Elliot and Ribes nigrum L.)* in the technology of semi-smoked sausages. The raw minced semi-smoked sausages were supplemented with the above-mentioned extracts in concentrations of 0.2–0.5 % of the weight of raw materials in order to slow down the oxidative processes in the lipid fraction of the product. Studies of the antioxidant efficiency of berry extracts and the effect on microbiological stability were performed during the shelf life of the finished product.

It has been found, that the introduction of the chokeberry extract in a concentration of 0.2 to 0.5 % of the weight of minced meat significantly inhibits the hydrolytic oxidation of lipids in the finished product, resulting in reduced intensity of lipid peroxidation. It has been confirmed, that the stabilization of lipid peroxidation in semi-smoked sausages leads to inhibition of the formation of primary oxidation products. At the end of the shelf life of the PV (peroxide value) of the test samples was at least 0.017 mg/KOH (potassium hydroxide), which was 63.04 % less than in the control.

The study of the content of secondary oxidation products allowed to estimate the depth of oxidation processes, occurring in the samples of semi-smoked sausages when stored for 25 days at a temperature of 0–6 °C. It has been proved, that the amount of secondary oxidation products, reacting with thiobarbituric acid, was the lowest at the end of the shelf life of the finished product with a concentration of the chokeberry extract of 0.5 %. TBV (thiobarbituric value) of this sample was 0.197±0.001 mg MA/kg, which was 3.74 times lower than in the control.

The introduction of extracts can reduce microbiological contamination and achieve a bacteriostatic effect. The lowest total contamination was recorded in a sample with a concentration of the chokeberry extract of 0.5 %. The tendency to decrease MAFAnM (number of mesophilic aerobic and facultative anaerobic microorganisms) was noted in all experimental samples. And in direct proportion to the concentration of extracts. Comparative evaluation of the effectiveness of the preparations showed that the extract of chokeberry in a concentration of 0.5 % more effectively inhibits oxidative processes than the extract of black currant.

Keywords: chokeberry extract, black currant extract, semi-smoked sausages, natural antioxidants.

DOI: 10.21303/2504-5695.2022.002286

1. Introduction

One of the main problems of the quality control system of meat products is oxidative spoilage. The high concentration of the fat fraction in meat products, especially unsaturated lipids, leads to the risk of oxidation and accumulation of its products. The consequence of this process is a violation of taste, aroma, appearance and other sensory characteristics of the finished product during storage and sale [1, 2]. On the other hand, producers face with the task of satisfying the consumer's desire to eat healthily and safely. In the conditions of modern production, the technological receptions, allowing to prevent or to slow down undesirable physical and chemical changes and at the same time to be economically effective and safe, acquire special value [3].

An urgent task in the meat industry is the search for natural antioxidants, which help not only achieve the optimal effect of inhibiting hydrolytic and oxidative spoilage of products with a long shelf life, but also to produce safe healthy foods. The use of biologically active compounds of plant origin in the technology of meat products is an alternative to synthetic antioxidants that have a toxic effect on the human body [4, 5].

The high content of polyphenols, flavonoids and other antioxidants in leaves, fruits, seeds of many plants cause their high efficiency in inhibiting the oxidative cleavage of lipids [6, 7]. Berry extracts can be one of the sources of bioflavonoids of multicomponent composition.

Therefore, the aim of the research was to study the effectiveness of the use of berry extracts in the technology of semi-smoked sausages. The task was to evaluate the effect of different concentrations of chokeberry and black currant extracts on hydrolytic, lipid peroxidation, accumulation of secondary oxidation products and microbiological safety of semi-smoked sausages during storage.

2. Materials and Methods

A recipe for semi-smoked sausages with the following ratio of components was developed: semi-fat pork – 30 %, lean pork – 10 %, duck meat (*Anas platyrhynchos*) – 30 %, lard – 25 %, hydrated bamboo fibers. Spices and additional materials were used in the recipe. In order to effectively use regional raw materials in the recipe, beef was replaced by duck meat, which increased the content of the lipid fraction. Hydrated bamboo fiber was added to prevent negative effects on the functional and technological properties of the system.

To prepare the sausages, the meat was minced in a Philips laboratory meat grinder (Germany). Lard was cut by hand into 4×4 mm cubes. The crushed ingredients were stirred for 8 minutes. Minced sausages were stuffed into the shells of lamb intestines. The sausages were precipitated at a temperature of 4–8 °C for 2 hours, then dried in an oven at t=90±10 °C for 30–40 minutes.

Smoking was carried out in a smoking chamber at an initial temperature of 43 °C, every 30 min the temperature was raised by 8–10 °C until the temperature in the center of the sausage was 70±2 °C. After smoking, the sausages were cooled to a temperature, not exceeding 8 °C.

Berry extracts (manufacturer – "Food Ingredients Mega Trade", USA) were added to the experimental samples of minced meat in the following concentrations:

– sample No. 1 – control, without antioxidants;

– samples No. 2 – 0,2 %, No. 3 – 0,3 %, No. 4 – 0,5 % of the chokeberry extract (*Aronia melanocarpa Elliot*) to the weight of raw minced meat;

– samples No. 5 – 0.2 %, No. 6 – 0.3 %, No. 7 – 0.5 % of the black currant extract (*Ribes nigrum L.).*

Recommended concentrations of antioxidants, ranging from 0.01 to 1.0 %, were used to determine the dose for the meat products technology [8, 9].

Cooked sausages were stored for 25 days at a temperature of +4 °C and a relative humidity of 75–78 %. During storage of the sausages, the controlled parameters were acid value (AV), peroxide value (PV), thiobarbituric value (TBV), microbiological parameters [10–12].

The acid value was measured by titration with sodium hydroxide in the presence of an alcohol solution of phenolphthalein [10]. 3–5 g of the test sample were weighed into a conical flask with a volume of 150–200 cm^3. The portion was heated in a water bath. 50 cm^3 of neutralized ether-alcohol mixture were added and shaken. Then 3–5 drops of the alcohol solution of phenolphthalein with a mass fraction of 1 % were added. The solution was rapidly titrated with a solution of potassium hydroxide with a concentration of 0.1 mol/ dm^3 with constant shaking until a pink color that does not disappear within 1 min. The acid value was calculated as the volume of sodium hydroxide solution with a molar concentration of 0.1 mol/dm^3, spent on titration of a test sample weight.

Determination of the peroxide value is based on extraction with a mixture of chloroform and glacial acetic acid and subsequent titration with a solution of sodium hyposulfite with a pre-added starch solution [10]. 0.8–1 g of the sample was placed in a flask with a stopper, heated in a water bath and 10 cm^3 of chloroform and 10 cm^3 of glacial acetic acid were added. 0.5 cm^3 of a saturated freshly prepared potassium iodide solution was quickly added. The flask was capped, the contents were stirred, 1 cm^3 of 1 % starch solution and 100 cm^3 of distilled water were added. Then the flask was placed in a dark place for 3 minutes. The contents were titrated with 0.01 mol/dm^3 sodium hyposulfite solution to remove a blue color.

To check the transparency of the reagents, a control determination was performed without a test sample. The peroxide value was calculated as the difference between the volume of the sodium hyposulfite solution with a concentration of 0.01 mol/dm,^{3} spent on titration of the experimental sample, and the volume of the sodium hyposulfite solution (0.01 mol/dm^3), spent on control titration based on the weight of the sample.

Thiobarbituric value (TBV) was determined by measuring the color intensity of a mixture of distillate of the test sample with a solution of thiobarbituric acid (1:1) after standing in a water bath for 35 minutes on a spectrophotocolorimeter "Spekol-11" (Germany), **Fig. 1**, at a wavelength of 535 nm [11].

Fig. 1. Spectrophotocolorimeter «Spекol-11» (Germany)

50 g of chopped sausage were mixed with 50 cm^3 of distilled water to the homonymous state. The prepared mass was quantitatively transferred to a Kjeldahl flask, the residue was washed from a mortar by 47.5 cm^3 of distilled water and 2.5 cm^3 of hydrochloric acid were added. Distillation in a Kjeldahl apparatus was performed by collecting 50 cm^3 of distillate in a volumetric flask. 5 cm^3 of distillate were taken, 5 cm^3 of thiobarbituric acid were added and the flask was placed on a boiling water bath for 35 minutes. The control test was performed using 5 cm^3 of distilled water instead of distillate. After cooling the samples for 10 min, the optical density was measured at a wavelength (535±10) nm relative to the control solution.

TBV, mg MA (malonic aldehyde)/kg of product, calculated the optical density of the solution, multiplied by 7.8 – the coefficient of proportional dependence of the density of MA on its concentration in the solution

The number of mesophilic aerobic and facultative anaerobic microorganisms was determined by method [12]. 10 g of each sample under sterile conditions were homogenized with 90 ml of peptone-salt broth using a mixer for 60 s at 20 °C.

10 cm^3 of the homogenized solution were placed in a sterile tube. The tube with the product at a given temperature was kept in a water bath with a temperature of (95±1) °C for 20 minutes

The number of MAFAnM (number of mesophilic aerobic and facultative anaerobic microorganisms) in 1 g (cm^3) was determined by inoculating successive solutions in Petri dishes by the deep method. Dilutions were selected for 15,300 colonies, grown in inoculations on the Petri dishes. Inoculations were thermostated at (30±1) °C for 72 hours.

After thermostating, Petri dishes were selected from 15–300 grown colonies.

The recalculation of the number of MAFAnM per 1 g (cm^3) was performed depending on the type of test product according to the formula:

$$X=a\cdot 10_n\cdot(V_{pr}+V_{water})/V_{pr}\cdot g, \quad (1)$$

where X – number of colonies in 1 g (cm^3); a – number of colonies, grown in the vessel; n – degree of tenfold dilutions; V_{water} – mass (volume) of added water; V_{pr} – mass (volume) of the product, cm^3; g – mass (volume) of inoculation material, g. The test for the detection of coliform bacteria was performed on Kessler's medium according to method [12].

Statistical analysis data were processed using Microsoft Excel software (USA). All experiments were performed in triplicate. The results presented are the results of these redefinitions with standard deviations. Student's t-test was used for the statistical analysis of the obtained results. The data are presented as mean±standard deviation of the mean ($M\pm m$). The smallest acceptable difference for samples from one sample was indicated at 5 %.

3. Results

The results of the study of the acid value dynamics (AV), which indicates the degree of hydrolytic oxidation of fat, showed that in the first half of the shelf life the amount of free fatty acids increased sharply. By the end of the storage period, their accumulation continued and almost doubled. However, during the whole period this indicator was lower in the experimental samples compared to the control. On the 15th day of storage, AV in the control was 0.701±0.03 mg KOH (potassium hydroxide), which is on average 50.5 % higher compared to samples, containing the chokeberry extract, and 26.82 % higher than in samples with the black currant extract. It can be stated, that already in the first stages of oxidative spoilage the extracts used effectively inhibit the hydrolytic decomposition of triacylglycerides of semi-smoked sausages. A similar difference persisted until the end of the standard shelf life of sausages. The AV in the experimental samples fluctuated at the level of 0.391–0.813 mg KOH (potassium hydroxide), which is 18.78–60.94 % lower than in sausages without the addition of antioxidants. This effect can be explained by the fact that berry extracts, especially dark in color, contain polyphenolic compounds that have high antioxidant activity [13]. They inhibit the oxidation of meat ingredients (lipids and proteins) and prevent darkening of meat as a result of oxidation of heme pigments [14–16].

When studying the effect of berry extracts on lipid peroxidation in semi-smoked sausages, it has been found, that the dynamics of the peroxide value in all samples of semi-smoked sausages was positive, i.e. increased throughout the control period. After two weeks, storage PV in the control sample was 0.037±0.001 % J_2, which is on average 2.46–1.95 times higher compared to samples, containing berry extracts. It has been found, that after 25 days of storage, semi-smoked sausages with 0.5 % chokeberry extract had a concentration of peroxides 0.017±0.003 % J_2, which is three times less than in sausages without added antioxidants. The concentration of peroxides in the samples with the blackcurrant extract ranged from 0.018 to 0.027 % J_2, which is 2.55 times less than in the control. This is due to the fact that the active compounds (phenols, proanthocyanidins, anthocyanins, phenolic acids) of the extracts block the attachment of active oxygen to free fatty acid radicals and break the chain of free radical oxidation [17–19].

A similar trend was found in the study of the degree of accumulation of malonic aldehyde – the main product of secondary oxidation of lipids. The results of the study of TBV (thiobarbituric value) in samples of semi-smoked sausage showed that as a consequence of inhibition of the first and second stages of lipid oxidation with berry extracts, the amount of malonic aldehyde in the experimental samples decreased. TBV in experimental semi-smoked sausages was 0.197–0.507 mg MA/kg, while in the control this figure reached 0.736. When comparing the effectiveness of different concentrations and preparations, it should be noted, that the concentration of secondary compounds, found in sausage samples with the addition of the chokeberry extract, was 0.197–0.318 mg, which is three times less than in the control. It has been proved, that when adding the black currant extract to semi-smoked sausages, there is also a decrease in TBV compared to the control, but not so intensely: the difference is 31.52–45.24 %.

Antimicrobial activity of chokeberry and black currant extracts was confirmed during microbiological studies. The number of mesophilic aerobic and facultative anaerobic microorganisms in the test samples was less than in the control and ranged from 0.96 to $1.59 \cdot 10^3$ CFU (colony-forming unit) in 1 g against $1.98 \cdot 10^3$. It should be noted, that this indicator did not exceed the normative value for this group of meat products: $2.5 \cdot 10^3$ CFU in 1 g. The presence of phenolic compounds in plant extracts is determined by the antimicrobial properties of plant raw materials in meat products [20, 21]. The addition of berry extracts prevents oxidative spoilage and ensures the microbiological stability of perishable products, and, as a consequence, extends the shelf life.

In practical use, the concentration of preparations should be taken into account, which should be 0.2–0.5 % of the weight of raw minced meat. Extracts are introduced during the technological operation of mixing the ingredients of the minced meat system. The addition of extracts provides suppression of oxidative and microbiological processes within normal limits in the finished semi-smoked sausages during the shelf life of 25 days at a temperature of +4 °C and a relative humidity of 75–78 %. Further research is planned to extend the shelf life of semi-smoked sausages with berry extracts at different temperatures.

4. Conclusions

It has been confirmed, that the addition of extracts of chokeberry and black currant to semi-smoked sausages can inhibit the oxidative damage of lipids during all stages of this process. It has been shown, that the efficiency of extracts in inhibiting the hydrolytic cleavage of di- and triacylglycerides in the product is 126.82–150.5 %.

It has been established, that the use of extracts of chokeberry and black currant reduces the level of formation and accumulation of peroxidation products of free fatty acids. The degree of hydrolysis of fat in sausages during the entire shelf life was 0.17 mg/KOH, which is twice less than in sausages without antioxidants.

It has been proved, that both extracts are effective at the stage of secondary oxidation of peroxides with the formation of malonic aldehyde. The accumulation of compounds that react with thiobarbituric acid in sausages with the chokeberry extract is 0.197–0.301 mg MA/kg. The thiobarbituric value in sausages with the black currant extract was 0.403–0.507 mg MA/kg, which is three times lower than in sausages without added antioxidants.

The use of extracts allows to stabilize the number of mesophilic aerobic and facultative anaerobic microorganisms in the product during storage and to achieve a bacteriostatic effect.

The proposed technology can be used in the production of semi-smoked sausages to prevent oxidative spoilage of products during storage in order to obtain products of standard quality.

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Received date 10.12.2021
Accepted date 16.01.2022
Published date 31.01.2022

How to cite: *Pasichnyi, V., Bozhko, N., Tischenko, V., Marynin, A., Shubina, Y., Svyatnenko, R., Haschuk, O., Moroz, O. (2022). Study of efficiency of berry extracts in the technology of semi-smoked sausages. EUREKA: Life Sciences, 1, 25–31. doi: https://doi.org/10.21303/2504-5695.2022.002286*