

## INFLUENCE OF THYME AND JUNIPER ON THE MICROBIOLOGICAL PROPERTIES OF PORK BRISKET

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### Abstract

The aim of the work is to determine an influence of non-traditional mixtures of spicy-aromatic plants of thyme and juniper, their essential oils and alcohol extracts on the course of microbiological processes in pork brisket. Pork brisket was an experimental sample. It was added with black pepper, thyme and juniper, in the dry condition, as essential oils and alcohol extracts. Pork brisket, added with dry plants, essential oils, extracts of black and fragrant pepper, was taken as a control. According to the microbiological studies, it was established, that an exceed of the standard index of the content of mesophilic-aerobic and facultative anaerobic microorganisms (more than  $1,0 \times 10^3$  g/CCU) takes place in brisket samples with dry black pepper. And also in samples with thyme and juniper in ratios 0,9:0,7:0,2 and 0,9:0,6:0,3. An exceed of nMAFAM index is also fixed in experiments using essential oils of these spices. It was established, that after 80 min of the experiment, these mixtures remain resistant to spoilage by 20 min longer comparing with using dry black and fragrant pepper. Most intense antibacterial properties relative to the development of microorganisms were observed in experimental samples. It is noticeable in a sample with using dry comminuted spices thyme and juniper in ratios 0,9:0,8:0,1. The use of spicy-aromatic plants of thyme and juniper allow to increase the storage term of ready products.

**Keywords:** dry mixtures of black pepper, essential oils of thyme, alcohol extracts of juniper.

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### 1. Introduction

Different supplements of the vegetable and animal origin, with a high food value, functional properties and able to improve or stabilize the quality of ready products, are used in modern technologies of sausages production alongside with base raw materials.

Complex spicy-aromatic mixtures became an unalienable part of recipes and technological process of sausages production. It gives a possibility to make products of the stable quality and to widen their assortment. The use of spicy-aromatic plants allows to improve an outlook and sanitary-hygienic quality parameters of meat products. Due to substances, contained in them, spoilage processes of products stop due to their destructive effect on the pathogenic microflora [1].

The study was conducted as to the use of elder, thorn, ashberry extracts in food industry for revealing their braking effect on creation of free radicals. It was established, that plants contain the high level of natural antioxidants – anthocyanins and polyphenols [2].

The studies proved that aromatic herbs of rosemary, thyme and lavender and their essential oils have antioxidant properties and prolong the storage life of products [3]. There were studied antioxidant and antimicrobial properties of savory essential oil. The effectiveness of its use at food products manufacturing was proved [4]. The surface part of savory is used as a bactericidal, spasmolytic, diaphoretic, antiseptic means. It is used in food industry at pickling of cucumber and tomatoes, as an aromatic spice for soups, meat, mushroom, fish dishes and salads. It is used in Bulgaria as a spice in the mixture of salt, paprika, black pepper and fenugreek [5]. Essential oils gained the wide use together with spicy extracts. The results of the conducted studies proved that essential oils of rosemary and sage more intensely inhibit oxidizing spoilage of liver paste than a synthetic antioxidant [6]. Essential oils of balm are also characterized by positive antioxidant properties. Antioxidant properties of infusions of spicy herbs were studied [7].

The traditional recipe of sausage products includes black and fragrant pepper. Black pepper fruits contain essential oil up to 2,5 %, its main part is formed by pinnen, limonene and phellandrene, it also contains choline and acetylcholine, 5–9 % of piperine. Active substances, included in black pepper, stimulate the activity of the thyroid gland and increase oxygen consumption by tissues [8]. Black pepper contains 3,5 % of essential oil, which main component is eugenol, cineol, caryophyllene and phellandrene. Fruits and leaves of black pepper are a source of essential oil, used in medicine, especially in stomatological practice as an antiseptic means. Pepper also improves the stomach work, it is useful at wind [9].

Thyme is used in food industry for aromatization of sausages, cheeses, sauces, vinegar, confectionary products, for making liqueurs, at pickling vegetables, as a spice for salads, meat and fish dishes. New sprouts contain vitamin C (54,5 mg %), acids, mineral salts, protein substances. Dry thyme is used in many cookeries of the world as a spice for meat, vegetable and fish dishes [10]. Juniper fruits have a sweet and tart smack. Juniper fruits are used first of all for preparing meat of wild animals. This spice is perfectly combined with onion, garlic, mint, marjoram and is often used for producing marinades, in the technology of sausage products. Juniper cone-berries contain carbohydrates (40 %), wax, resins, tanning and dyeing substances, essential oil (2 %), organic acids (apple, vinegar, formic), macro- and microelements; aluminium, copper, iron, manganese and so on [11].

The aim of the work is to determine the influence of mixtures of spicy-aromatic plants of thyme and juniper on the course of microbiological processes in pork brisket. It allows to fix the presence and development of microorganisms at storage that influences the storage life increase.

## 2. Materials and Methods

Black and fragrant comminuted peppers, thyme and juniper of TM “ECO”, PE “Edel”, Ukraine, were used for the study. Essential oils of black and fragrant pepper, thyme and juniper were produced by the method of distillation with water steam. Vegetable raw materials were placed in a distiller. Then steam is removed from a stand of the distiller, and essential oils are mixed with steam. This volatile substance is transformed in water by condensation. Essential oil is extracted on its surface, then is separated from it. Alcohol extracts of black and fragrant pepper, thyme and juniper were produced by extracting. Ethanol was used as an extragent. The process includes preparation of raw materials and extragent, getting of an extract, purification of an extract. Dry comminuted aforesaid plants, their essential oils and alcohol extracts are added to pork brisket. Pork brisket, containing black and fragrant pepper in ratios 1:0,9 mg/1000 g was taken as a control. Experimental samples of pork brisket were added with dry black pepper : thyme : juniper in ratios

(mg/1000 g): 0,9:0,8:0,1 0,9:0,7:0,2 and 0,9:0,6:0,3. The recipes of control and experimental samples are presented in **Table 1**.

**Table 1**  
Recipes of control and experimental samples

Name of the recipe component	Name of the experimental sample											
	Control sample No. 1	Control sample No. 1.1.	Control sample No. 1.2	Sample No. 2	Sample No. 3	Sample No. 4	Sample No. 2.1	Sample No. 3.1	Sample No. 4.1	Sample No. 2.2	Sample No. 3.2	Sample No. 4.2
Pork brisket, g	1000	1000	1000	1000	1000	1000	1000	1000	–	–	–	–
Black pepper, mg/ 1000 g	1	–	–	0,9	0,9	0,9	–	–	–	–	–	–
Fragrant pepper mg /1000 g	0,9	–	–	–	–	–	–	–	–	–	–	–
Thyme mg/1000 g	–	–	–	0,8	0,7	0,6	–	–	–	–	–	–
Juniper mg /1000 g	–	–	–	0,1	0,2	0,3	–	–	–	–	–	–
Essential oils of black pepper, ml/ 1000 g	–	1	–	–	–	–	0,9	0,9	0,9	–	–	–
Essential oils of fragrant pepper ml /1000 g	–	0,9	–	–	–	–	–	–	–	–	–	–
Essential oils of thyme ml /1000 g	–	–	–	–	–	–	0,8	0,7	0,6	–	–	–
Essential oils of juniper ml /1000 g	–	–	–	–	–	–	0,1	0,2	0,3	–	–	–
Alcohol extracts of black pepper, ml/ /1000 g	–	–	1	–	–	–	–	–	–	0,9	0,9	0,9
Alcohol extracts essential oils of fragrant pepper ml/1000 g	–	–	0,9	–	–	–	–	–	–	–	–	–
Alcohol extracts essential oils of thyme ml/1000 g	–	–	–	–	–	–	–	–	–	0,8	0,7	0,6
Alcohol extracts essential oils of juniper ml/1000 g	–	–	–	–	–	–	–	–	–	0,1	0,2	0,3

The method of determination of the number of mesophilic aerobic and facultative-anaerobic microorganisms (nMAFAM) is realized by inoculation in a nutritive medium, incubation of inoculations, calculation of all grown colonies. A tenfold solution is produced from a batch of the product. Inoculation is made from correspondent solutions in two parallel Petri dishes and poured by one of agarized mediums. Inoculations are incubated at temperature 30 °C for 72 hours, then colonies are calculated and the correspondence for each type of products by this parameter is determined.

### 2. 1. Experimental procedures

Pork brisket is comminuted in pieces, added with dry mixtures of spicy-aromatic plants, extracts and essential oils. Experimental samples were selected for the further studies. Processes of fat oxidation were realized by subjecting experimental samples to the effect of high temperatures ( $t 100 \pm 2$  °C) with the air access during 120 min. For revealing the influence of spicy-aromatic plants, their essential oils and extracts on the development of microorganisms, there were selected samples for the microbiological studies. Namely: fresh brisket and spoiled samples. Control ones – with using dry comminuted spices, their extracts at 60 min of the experiment, essential oils – at 60 and 80 min of it. Experimental ones – with using dry spices, their essential oils after 60–120 min

of the experiment, extracts – after 60–80 min of it. Samples were inoculated for determining the number of mesophilic aerobic and facultative-anaerobic microorganisms, CCU, in g of the product. nMAFAM must not exceed  $1,0 \times 10^3$ , according to SSU 4668:2006 “Boiled, smoked-boiled, smoked-baked, baked, fried, raw-smoked pork products”.

### 3. Results

nMAFAM in the control sample No.1 with using dry comminuted thyme and juniper grew twice after 60 min of the experiment, comparing with the initial value that was  $2,5 \times 10^2$  CCU/g, control sample No.1.1 with using their essential oils at 60 min –  $1,1 \times 10^3$ , and at 80 min –  $9,8 \times 10^3$ , that demonstrates the intense development of microorganisms not only after spoiling samples, but also when the value of their peroxide and benzidine numbers are at the stage of doubtful freshness, control sample No.1.2, in which black and fragrant pepper extracts were used – reached maximal value  $4,7 \times 10^4$  CCU/g after 60 min that exceeds the previous samples essentially (Table 2).

**Table 2**

Comparative characteristics of nMAFAM CCU/g in experimental samples of brisket with using spicy-aromatic plants

Name of sample	nMAFAM, CCU/g			
	0 min	60 min	80 min	120 min
Control Sample No. 1 – dry comminuted BP:FP1:0,9 mg/1000 g	$2,5 \times 10^2$	$3,0 \times 10^4$	–	–
Control Sample No. 1.1. – essential oils BP:FP 1:0,9	$2,0 \times 10^2$	$1,1 \times 10^3$	$9,8 \times 10^3$	–
Control Sample No. 1.2 – alcohol extract BP:FP 1:0,9	$2,1 \times 10^2$	$4,7 \times 10^4$	–	–
Sample No. 2 – dry comminuted BP:T:J 0,9:0,8:0,1	$2,3 \times 10^2$	$6,0 \times 10^2$	$9,9 \times 10^2$	$2,0 \times 10^4$
Sample No. 3 – dry comminuted BP:T:J 0,9:0,7:0,2	$1,9 \times 10^2$	$6,2 \times 10^2$	$1,2 \times 10^3$	$1,5 \times 10^4$
Sample No. 4 – dry comminuted BP:T:J 0,9:0,6:0,3	$2,8 \times 10^2$	$7,2 \times 10^2$	$1,1 \times 10^3$	$2,8 \times 10^4$
Sample No. 2.1 – essential oils BP:T:J 0,9:0,8:0,1	$1,8 \times 10^2$	$5,4 \times 10^2$	$2,5 \times 10^3$	$9,5 \times 10^3$
Sample No. 3.1 – essential oils BP:T:J 0,9:0,7:0,2	$3,0 \times 10^2$	$6,9 \times 10^2$	$2,7 \times 10^3$	$1,1 \times 10^4$
Sample No. 4.1 – essential oils BP:T:J 0,9:0,6:0,3	$1,6 \times 10^2$	$4,6 \times 10^2$	$1,9 \times 10^3$	$8,1 \times 10^3$
Sample No. 2.2 – Alcohol extract BP:T:J 0,9:0,8:0,1	$2,6 \times 10^2$	$1,9 \times 10^3$	$3,2 \times 10^4$	–
Sample No. 3.2 – Alcohol extract BP:T:J 0,9:0,7:0,2	$3,2 \times 10^2$	$2,8 \times 10^3$	$4,5 \times 10^4$	–
Sample No. 4.2 – Alcohol extract BP:T:J 0,9:0,6:0,3	$2,9 \times 10^2$	$2,3 \times 10^3$	$3,7 \times 10^4$	–

The use of black pepper, thyme and juniper, used in different ratios, manifests the antibacterial effect in brisket and stops the development of microorganisms more intensely than black and fragrant pepper. The difference is noticeable after 60 min of oxidizing the product. Whereas the growth of microorganisms exceeded the norm in the control sample, nMAFAM of the experimental ones remained within  $6,0 \times 10^2$ – $7,2 \times 10^2$  CCU/g that is didn't exceed the standard requirements. The increase of the number of microorganisms that exceed the norm  $1,0 \times 10^3$  CCU/g was revealed at 80 min (up to  $1,2 \times 10^3$  CCU/g, and at 120 min, when accumulations of oxidation products in goods reached the maximal value – the number of microorganisms grew twice. A bit better microbiological indices are inherent to the brisket samples with using essential oils of black pepper and spicy-aromatic plants comparing with dry comminuted ones, but the nMAFAM value don't essentially differ from the previous ones – at 60 min it is within the norm (maximally to  $6,9 \times 10^2$  CCU/g), at 80 min – from  $1,9 \times 10^3$  (Sample No. 4.1) to  $2,7 \times 10^3$  (Sample No. 3.1), CCU/g. It must be noted that nMAFAM of the control sample with using essential oils of black and fragrant pepper exceeded the norm at 60 min –  $1,1 \times 10^3$  CCU/g, at 80 min reached maximal value  $9,8 \times 10^3$  CCU/g, whereas when nMAFAM of the experimental samples was within  $8,1 \times 10^3$  –  $1,1 \times 10^4$  CCU/g.

Alcohol extracts are ineffective in the fight against the development of microorganisms, because they manifest the weakest antibacterial effect. At using the alcohol extract of black and fragrant pepper there was observed the maximal value of nMAFAM  $4,7 \times 10^4$  comparing with all studied samples. The development of microorganisms is noticeable in samples with using alcohol extracts of spicy-aromatic plants after 60 min of the experiment from  $1,9 \times 10^3$  to  $2,8 \times 10^3$  CCU/g, the maximal value of the parameter was fixed after 80 min –from  $3,2 \times 10^4$  (sample No. 2.2) to  $4,5 \times 10^4$  (sample No. 4.2) CCU/g, that is they have an advantage comparing with using alcohol extracts of black and fragrant peppers.

It must be noted, that at studying the total value of inoculation of the product by microorganism, it is impossible to determine what ratio of the used spicy-aromatic plants manifests maximal properties as to preserving the product quality, because initial indices of nMAFAM differ from each other. It may be caused by contamination of experimental samples from air, hands auxiliary materials at their selection for studies and preparation for inoculations. But it is proved, that black pepper, combined with thyme and juniper stops the development of microorganisms essentially better than the combination of black and fragrant pepper in any form (dried, comminuted, essential oils, extracts). The use of spicy-aromatic plants in the dry comminuted condition is not inferior to their essential oils as to the influence on the development of microorganisms, so their use in the offered condition is possible.

#### 4. Conclusions

The use of both dry comminuted spicy-aromatic plants, based on black pepper, thyme and juniper, and their essential oils manifests the antibacterial activity much more intensely than black and fragrant pepper and stop the development of microorganisms twice. It is an advantage of the offered spices. A shortcoming is the fact that alcohol extracts of the experimental spices are ineffective, because it was established, that accumulation of oxidizing compounds and the development of microorganisms in experimental samples with their use is much faster comparing with other ones.

The studies are useful because the use of mixtures of the experimental spices in the dry comminuted condition guarantees the effective braking of animal fats spoilage processes. The use of mixtures of black pepper, thyme and juniper as new natural supplements is possible in the technology of sausage products for prolonging the storage term.

Further studies will be directed on investigating the influence of these spices on preservation of quality parameters of pastes of semi-smoked sausages.

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## **DETERMINATION METHODS OF DEFROSTED PROTEIN-VEGETABLE MIXTURES PARAMETERS DEVELOPMENT**

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