THE FORMATION OF FLAVORING CHARACTERISTICS OF MEAT PRODUCTS BY CHANGING THE CHEMICAL COMPOSITION OF FOOD COMPOSITIONS

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Abstract
The article presents the results of the study of changes in flavour characteristics when using corrective additives. Monosodium glutamate, ribotide, yeast extract and hydrolysate of vegetable soy protein were used as flavoring additives (FA). To assess the effect of composition of meat product recipe, as well as the method of FA-introduction on taste intensity, the recipes of model meat systems with partial replacement of meat raw materials were used. Pork fat, soy protein and potato starch were used as meat substitutes. The effect of recipe composition on the content of non-volatile substances of aroma was accessed. It is shown that replacement of pork by pork fat in the recipe by 20–40% led to a sharp decrease in the concentration of aromatic substances and a decrease in intensity of taste of the finished product several times. The ways for taste correction using FA was studied. For this, a chopped semi-finished product — minced meat was prepared from chilled whole-muscle pork and 0.05% of each FA was added. It is shown that the dynamics of changes in the content of free amino acids is the most pronounced when using monosodium glutamate not as a mono-additive, but in compositions: monosodium glutamate with yeast extract and monosodium glutamate with ribotide. A pool of chemical compounds involved in the formation of taste and aroma of products was detected. The main components were derivatives of C6–C24 fatty acids, as well as a significant number of other biochemical compounds, mainly substituted amines, amides, alcohols and ketones, with a content ranging from 0.001 to 0.2 mg/kg. The results of organoleptic analysis showed that the most delicious and attractive samples were those containing monosodium glutamate with yeast extract and monosodium glutamate with ribotide.

Introduction
The quality of meat products depends on the composition and properties of raw materials, as well as the conditions of its technological processing. The influence of natural factors, conditions of livestock rearing, transportation, pre-slaughter animal treatment, slaughtering conditions, the state of primary meat processing, parameters of autolysis of the obtained raw materials and their further cold storage is significant [1,2].

The main consumer indicators of raw material quality, including tenderness, pH level, and the degree of development of muscle tissue elements, are largely inherited and can be corrected by various food additives [3].

Feeding diets have a decisive influence on the quality of raw materials of animal origin and, ultimately, on its chemical composition. Feeding diets largely determine the composition and ratios of biochemical substances formed in the flesh of the animal, which later form flavour characteristics [4,5].

The lack of essential components in the feeding diets leads to an increase in water content and causes a decrease in the mass fraction of protein and fat, as well as an increase in the coarseness of fiber structures [6].

As a result of disorders in the type of feeding diets, as well as the presence of stress factors, animal raw materials may have parameters of lower quality with a predominance of a specific smell and taste. Microbiological additives in feed, as well as additives of processed seafood waste, also contribute to the appearance of an undesirable oil or fishy off-flavor [7].

Disorders in the type of feeding diets, increased susceptibility of animals at mass management, stress during the slaughter result in production of raw materials, primarily of lower morphological quality. In this case, it is significant that meat with non-traditional quality characteristics is formed: the so-called PSE (Pale, Soft, Exudative) watery, flabby and exudative meat with a pH < 5.4 and coarse, dark DFD (Dark, Firm, Dry) meat with a pH > 6.2. Recently, red non-standard watery meat RSE (Red, Soft, Exudative) is also distinguished. Meat with abnormal characteristics has technological properties that are not typical of NOR (normal) raw materials and, most important, different taste, aroma, consistency, i. e. other organoleptic properties. The amount of PSE, DFD, and RSE can be from a quarter to half of all processed volumes. This significantly complicates the production of “delicious” meat products

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The role of glutamic acid in the flavour spectrum is very significant. It and its sodium salt, even in a small amount, about 0.03%, impart the product a meat taste. The presence of free amino acids such as valine, methionine and glycine also contributes to the unique aroma of meat products. The content of inosinic, cytidylic, uridine and guanylic acids in beef and pork in the process of heat treatment is changed almost in the same manner. Their decrease may be 25–40% of the initial value [15,18].

Substances that are flavor precursors are of great importance. Substances that determine the taste and aroma of meat have a molecular weight of less than 200 Da and are extractive. Native meat extracts contain natural amino acids and short peptides, as well as glucose, glucosamine, fructose and ribose. After heat treatment, a greater amount of lactic acid appears, amines, inosine monophosphate, inosines, carbonyl and sulfur-containing compounds are formed, as well as significant amounts of free and derived amino acids. An important role here is played by the flavor precursors contained in raw meat. These include: carnitine, lysine, carnosine, methionine, creatinine, methylhistidine, cysteine, isoleucine, cystine, leucine, glucose-6-phosphate, glutamic acid, glutamine, nicotinamide adenine dinucleotide (NAD), glutathione, ornithine, glycophosphothanolamine, glycine, phenylalanine, glycoproteins, phosphoethanolamine, histidine, phosphoserine, hydroxyproline, fructose, hypoxanthine, fructose-6-phosphate, inosine-5-monophosphate, proline, nucleotides, uridine, inosine tetralide, uridine nucleosides, ribose, ribose-5-phosphate, serine, methylhistidine, taurine, tyrosine, threonine, isoleucine. Each of these substances has a certain influence on the variety of flavour characteristics [19].

According to information sources, the aromatic composition of meat can contain up to 0.5% of ketones and esters, 1.5% of hydrocarbons, 1.5–2% of sulfur-containing compounds, 4–5% of alcohols, 12–15% of aldehydes, 28–30% of furans and up to 50% of pyrazines. Pyrazines were detected in heat-treated meat, for example, 2,3-diethyl-5-methylpyrazine, 2-methylpyrazine, 2,3-diethyl-2-methylpyrazine, 2,5-dimethylpyrazine, 2,3,5-triethylpyrazine, 2-acetyl-3-methylpyrazine, 1-pyrazinyl-2-propanone, etc., in total a few tens of substituted pyrazides [20,21].

Pyrazines have a sensory effect in very low concentrations and play a significant role in creating the aroma of fried food. Methoxyalkylpyrazines convey the aroma of greenery. The presence of a substituent in the third position of the ring causes the aroma of green beans, substitution of hydrogen atoms in the ring causes the characteristic aroma of green bell pepper. Threshold concentrations of sensory properties of these compounds are, ppb: 2-ethylpyrazine (nutty, burnt) — 400.0; 2-ethyl-3,6-dimethylpyrazine (aroma of fried, spicy boiled potato) — 0.002; 2-ethyl-6-vinylpyrazine (buttered, baked) — 0.002; dimethylhexahydroxycyclopentapyrazine (fried beans) — 0.002; 2-methylamino-3-methylpyrazine (roasted, greenery) — 0.002; 2-methyl-5-thiomethylpyrazine (meat,
The mechanisms of forming taste and aroma of meat are determined by a complex of organic components, which can be contained in small amounts, up to 0.001%. However, a fundamental role in the formation of the aromatic “bouquet” of meat is played by a small group of key substances that form the four main tastes — salty, sweet, sour and bitter. In meat, the sour taste is formed mainly by lactic, phosphoric and pyruvic acids; salty — by salts of the same acids and chlorides, bitter — by nitrogen-containing carboxylic acid creatine, some free amino acids, such as L-tryptophan and L-isoleucine, and nitrogenous extracts. The sweet taste is determined by glucose, ribose, trioses, and some amino acids, such as a mixture of D — and L-tryptophan, as well as L-glycine, L-alanine, L-serine, L-proline, D-valine, D-leucine, D-threonine, D-methionine, and D-histidine [23, 24].

Recently, some Asian countries (Japan, China) have distinguished the fifth taste, the so-called Umami, which means meat, spicy and delicious taste with a long aftertaste. Umami taste receptors are proteins with a high content of glutamine and glutamic acid, as well as glutamates. This hydrolysate of vegetable soy protein “HVP 2M-P1” produced by “Ajinomoto do Brasil Industriae Comercio de Alimentos Ltda” (Brazil) with formula C₉H₁₈NO₅Na and basic substance content > 95%, an average particle size of 150 μm, pH of 1% solution — 7.0;

— ribotide Ajidite, Ribotide (I+G) produced by Ajinomoto (Japan), which is a 1:1 mixture of sodium inosinate (E627) and sodium guanylate (E631), containing > 98% of basic substance, pH of 1% solution — 7.8;

— yeast extract containing, %: dry matter > 95; ash — 9.5; nitrogen of free amino groups — 5.5; total nitrogen — 10.7; potassium — 5.7; calcium — 0.1; magnesium — 0.12; sodium — 0.3; amino acids: alanine — 8.7; histidine — 2, proline — 4, arginine — 5, isoleucine — 5.6; serine — 4.7; aspartic acid — 9.7; leucine — 7.6; threonine — 4.4; cystine — 0.8; lysine — 8, tryptophan — 1.2, glutamic acid — 16.1; methionine — 1.3; tyrosine — 2.3; glycine — 4.9; phenylalanine — 3.8; valine — 5.8; pH of 2% solution — 6.8.

— hydrolysate of vegetable soy protein “HVP 2M-PI” produced by Vitana (Czech Republic), containing > 95.0% of dry matter, 26% of protein, 2.0% of fat, 7% of natural glutamate, pH of 10% solution — 5.6.

Determination of the mass fraction of moisture, fat, protein, ash, carbohydrates, and pH was performed using standard methods [25].

The content of amino -, fatty acids, free monosodium glutamate, inosine and carnosine was carried out by chromatographic method [26].

Model systems were prepared according to the following scheme. At the first stage, semifat pork pre-minced in a meat grinder was added to the stirrer, and the ingredients listed above were added one by one. The finished minced meat was shaped in the form of cutlets. Heat treatment was carried out without adding oil in the convection steamer until the temperature 72–75°C inside was reached. 4 hours after the heat treatment, 100 g of the sample of each model system was minced using a blender. Non-volatile substances (NVS) were isolated by extraction 1:4 with hexane, and the composition was analyzed by the method of HPLC in the conditions: the volume of introduced sample 20 μl, gradient of eluent at the start 98% of A and 2% of B (A — 0.1% aqueous solution of formic acid, B — acetonitrile), 10 min — 80% of A and 20% of B, 20–30 min 10% of A and 90% of B, flow rate 1 ml/min, pressure 1500 psi, UV detector 355 nm. Identification was performed in automatic mode using mathematical statistics methods, estimating the sum of one hundred most significant peaks on the HPLC chromatogram.

The chemical composition of aroma components was analyzed by gas chromatography on a 7890A chromatograph.
The effect of the recipe composition on the content of non-volatile components of NVS aroma was assessed. As it can be seen from Table 2, replacement of pork with pork fat in the recipe by 20% and 40% (model systems 1 and 2) led to a sharp decrease in the concentration of NVS, i.e., to a decrease in the taste intensity of the finished product 6.25 and 14.2 times, respectively. At the same time, replacing pork with soy protein or starch only slightly reduced the taste intensity of the product: when replacing pork with 20 and 30% of soy protein (model systems 3 and 4), the taste intensity of the product decreased by 5% and 9%, respectively. A similar replacement with starch (model systems 5 and 6) resulted in a decrease in the taste intensity of the product, respectively, by 3 and 5%.

The obtained results can be explained by the fact that with a significant increase in the fat content in the recipe, there is a deficiency of protein for binding fat drops and, as a result, there is an increase in the proportion of fat drops without protein shells. These drops actively absorb fat-soluble NVS, and that leads to a noticeable decrease in the taste of the finished product. In addition, protein deficiency can lead to reduced stability of meat emulsions and formation of broth-fat pockets.

It is also necessary to note the deterioration of the quality of pork fat sold on the Russian market. So, its fatty-acid composition included (%): C4:0—0.02; C6:0—0.11; C8:0—0.1; C10:0—0.17; C12:0—0.43; C14:0—0.61; C15:0—0.08; C16:0—19.8; C17:0—1.4; C18:0—17.4; C19:0—1.3; C20:0—0.1; C22:0—1.47; C14:1—0.12; C15:1—0.1; C16:1—4.6; C17:1—0.5; C18:1n9c — 27.3; C20:1—0.6; C22:1n9—0.35; C18:2n6c — 4.1; C18:3n6—0.48; C18:3n3—0.29; C20:2—0.35; C20:3n6—0.62; C20:4n6—1.35; C22:2—0.48; C20:5n3 — 0.22; C22:5n3—0.11; C22:6n3—0.02.

Its fatty acid profile is mainly represented by unsaturated fatty acids, which are quickly oxidized during storage, and that, in turn, negatively affects the meat taste. Due to the ongoing reduction in the fattening period of pigs from 12 to 5 months, i.e. 2.5 times, changes occur in the fatty acid profile, in particular, during this period there is no sufficient accumulation of saturated fatty acids, which are not capable of rapid oxidation.

From the obtained data, it can be concluded that it is expedient to replace high-quality meat raw materials with non-fat-containing ones in the recipes of meat products, and use hydrated soy protein or starch. In this case, there is a less significant decrease in the flavour characteristics of the finished product.

To study the effect of the method of introduction of food additives that affect the taste of finished products, the following experiment was conducted. In order to simulate standard conditions of introduction, monosodium glutamate was added to the meat system at the first stage of mixing in the model meat system (variant 1, Table 3). For comparison, sodium glutamate was added to the model system (variant 2, Table 3) at the second stage of the minced meat mixing.
As the results show, the introduction of monosodium glutamate at the second stage was more effective, since the amount of substances responsible for the taste of meat and meat products increases by more than 18%.

Ways of correction of taste of meat and meat products in order to improve them were studied. To do this, a chopped semi-finished product — minced meat was made from chilled whole-muscle pork. From the total mass of minced meat, samples of minced meat weighing 100 g each were selected and flavouring additives were added one by one according their compositions:
— sample № 1 (control);
— sample № 2 sodium glutamate (dosage of 0.05%);
— sample № 3 sodium glutamate (dosage of 0.05%) + yeast extract (dosage 0.05%);
— sample № 4 sodium glutamate (dosage of 0.05%) + ribotide (dosage of 0.05%);
— sample № 5 sodium glutamate (dosage of 0.05%) + vegetable protein hydrolysate (dosage 0.05%).

The results of the analysis of the chemical composition of the samples with the added FA are presented in Table 4.

The results presented in Table 4 indicate the identity of the chemical composition of the studied samples. The indicator “carbohydrate content” draws attention. In samples № 3 and № 4, their amount increased almost 3 and 4.3 times, respectively, compared to the control sample. This fact can be explained by the presence of increased carbohydrate content in sample № 3 (yeast extract) and sample № 4 (ribotide).

From the above data, it can be seen that the addition of sodium glutamate leads to its adequate increase almost 4 times in the minced meat, while the content of inosine and carnosine remains almost unchanged compared to the original sample.

Addition of the composition consisting of monosodium glutamate and yeast extract (sample № 3) contributes to the increase in the content of aroma precursors — inosine and carnosine, respectively, 2.2 and 1.7 times, which positively affects the taste of meat products. Besides, it is known that there is a synergistic effect between monosodium glutamate and the nucleotides contained in the yeast extract.

In sample № 4, containing a composition of monosodium glutamate and ribotide (inosinic acid: guanoic acid = 50:50), a significant increase in all analyzed indicators was noted: monosodium glutamate — 5.8 times; inosine — 4.7 times; carnosine — 1.6 times.

It can be assumed that the above composition will be the most impactful on improving the taste of the finished meat product.

Sample № 5, containing monosodium glutamate and vegetable protein hydrolysate also provides an increase in the level of all the studied indicators: monosodium glutamate 5.6 times; inosine — 1.2 times; carnosine — 1.5 times.

The obtained results confirm the fact of significant influence of the used NVS on the characteristics of the food composition.

The composition of samples containing various types of flavor enhancers after heat treatment in convection steamer was studied. Table 5 shows the characteristics of heat-treated products.
The results presented in Table 5, show that due to the heat treatment there is a change in the balance of moisture, fat and protein because of thermal loss during steam-boiling. High carbohydrate content of the samples after heat treatment should be noted; its level increased more than 6 times, that, apparently, can be explained by the intensive interaction of amino acids contained in the flavouring compositions with the proteins of the muscle tissue of the studied samples.

It is known that glutamic and aspartic acids, histidine, serine, cystine and methionine have the most significant influence on the taste of meat and meat products, which have the ability to correct the taste of the product according the sweet — bitter scale [27,28].

Amino acids are contained in food in a free and bound state, and free amino acids influence the taste and aroma more significantly [29].

A comparative analysis of the content of free amino acids before and after heat treatment in the studied samples is given in Table 6.

From the results shown in Table 6 it can be seen that the addition of monosodium glutamate, as a flavor enhancer, allows increasing the content of glutamic acid in sample № 2 almost 2 times after heat treatment, compared with its content in the control sample (semifat pork), in samples № 3, 4, 5 — almost 4.0 times. The addition of flavor enhancers allowed increasing the content of aspartic acid after heat treatment in sample № 2, almost 1.6 times, in sample № 3–2.85 times, and in sample № 4.5 — almost 2 times.

The content of serine in sample № 2 increased 1.35 times, in samples № 3, 4–1.6 times, in sample № 5–3 times. The amount of histidine in sample № 2 increased about 3.2 times compared to its content in the control sample, in samples № 3, 4, 5 — almost 6 times.

The content of cystine in sample № 2 increased approximately 3.5 times, in sample № 3— almost 5 times, in sample № 4–2 times, in sample № 5 remained almost unchanged.

According to the expert assessment, the "enhanced taste" of the meat product most clearly appeared in the case of using a combined mixture of monosodium glutamate and soy protein hydrolysate.

Mass spectrometric analysis of aromatic substances revealed a pool of free fatty acids involved in the formation of taste and aroma of products. The basic components were derivatives of fatty acids. Their composition in products with monosodium glutamate and soy protein hydrolysate, before and after heat treatment, in the form of methyl esters is shown in Figure 1, in concentrations, mg/kg:
Most of the major compounds were methyl esters of myristic (methyl tetradecanoate), erucic (methyl cis-13-ecisenoate), palmetoleic (methyl hexadec-9-enoate), palmitic (methyl hexadecanoate) and oleic acids (methyl (Z)-9-octadecenoate), which are mainly monounsaturated fatty acids.

In the composition of aromatic substances of the product with the addition of monosodium glutamate and soy protein hydrolysate were found more than 200 components, including those with the highest content is presented in Table 7.

A significant proportion of the aroma components was present in quantities from 0.001 to 0.2 mg/kg. And only 38 components were present in higher concentrations.

It is obvious that for a more complete assessment of the influence of these substances on the aroma profile, more in-depth research is needed, which was not the purpose of this work.

### Table 7. Aromatic substances of the product with the addition of monosodium glutamate and soy protein hydrolysate

| №  | Compound name                          | Content, mg/kg, without heat treatment/ with heat treatment | Compound name                          | Content, mg/kg, without heat treatment/ with heat treatment |
|-----|---------------------------------------|-----------------------------------------------------------|---------------------------------------|-----------------------------------------------------------|
| 1   | cyclopentyl ester                      | 2.82 (ND)                                                | 5,6-dimethyl-phenanthridinium         | 2.22 (ND)                                                |
| 2   | 10-pentadecen-5-yn-1-ol                | 0.85 (ND)                                                | 1-methyl-4-(1-methylethyl)-1,3-cyclohexadiene | 0.94 (ND)                                                |
| 3   | 6-tetra-O-methyl-octanoic acid ethyl ester | 0.89 (0.33)                                            | 1-undecene                           | 1.71 (ND)                                                |
| 4   | trenbolone                             | 0.93 (005)                                              | 1-phenyl-4-(2-cyano-2-phenylethenyl) benzene | 0.54 (ND)                                                |
| 5   | 2-ethylacridine                        | 2.31 (1.56)                                             | heneicosane                          | 3.62 (3.3)                                              |
| 6   | eicosane                               | 2.14 (1.45)                                             | tetratriacontane                      | 2.47 (ND)                                                |
| 7   | pyridine                               | 2.26 (0.76)                                             | 2-hexen-1-ol                          | 0.31 (ND)                                                |
| 8   | 10-methyleneadecane                    | 4.01 (2.5)                                              | 7-methoxy-3,7-dimethyl-octanal       | 0.56 (0.1)                                              |
| 9   | n-nonadecanol-1                        | 7.53 (ND)                                               | 3-methyl-tridecane                    | 0.94 (0.45)                                              |
| 10  | octadecane                             | 2.16 (ND)                                               | 2-naphthyl-p-tolyl sulfone            | 0.40 (ND)                                                |
| 11  | trans-2,3-methylenedioxy-b-methyl-b-nitro styrene | 0.29 (ND)                                                 | nhexahydro-2H-pyrido(1,2-a)pyrazin-3(4H)-one | 0.69 (ND)                                                |
| 12  | (3,4-dimethoxy-benzyl)-(4-morpholin-4-y) l-amine | 0.46 (ND)                                         | 5-(4-ethoxyphenyl)-3-(4-pyrol-1-ylphenyl)-[1,2,4]oxadiazole | 0.41 (0.16)                                              |
| 13  | N-(2-chloroethoxy carbonyl)-l-methionine, propyl ester | 0.45 (ND)                                               | 2,3-dihydro-2,8-dimethyl-benz[b]-1,4 oxazepine-4(5H)-thione | 0.55 (ND)                                                |
| 14  | ethanethioic acid                      | 0.21 (ND)                                               | paroxetine                           | 0.41 (ND)                                                |
| 15  | ethyl ester decanoic acid              | 3.86 (ND)                                               | 1-acetyl-4-[1-piperidyl]-2-buty none  | 0.35 (ND)                                                |
| 16  | 5-ethyl-2-methyl-octane                | 7.13 (ND)                                               | 2-methylaminomethyl-1,3-dioxolane     | 0.36 (ND)                                                |
| 17  | triacetone                             | 3.37 (2.55)                                             | 1-tetracontane                       | 0.67 (ND)                                                |
| 18  | (3S)-pentanol                          | 1.02 (ND)                                               | 4-methyl-2-hexanone                   | 0.34 (ND)                                                |
| 19  | docosane                               | 1.13 (1.25)                                             | tetratriacontane                     | 2.61 (0.95)                                              |
Some components found in the composition of aromatic substances of the starting raw material, after heat treatment together with the used NVS, were found to be in significantly smaller quantities or were present in the form of other chemical derivatives of these components. Generally, the overall aroma seems to be due to the synergistic effect of all the detected substances.

The results of organoleptic analysis obtained by ten tasters: appearance, color index of the product (in the section), taste, aroma and texture of the studied products showed that the most delicious samples were those containing monosodium glutamate with yeast extract (sample № 3), and monosodium glutamate with ribotide (sample № 4). The results of a positive tasting assessment correlate with the dynamics of changes in the amino acid composition of samples after heat treatment, confirming the significance of the influence of free amino acids on the flavour characteristics of food products.

It can be concluded that the above-mentioned compositions are effective correctors for enhancing the flavour of meat and meat products.

Conclusion
The results of the study of the mechanism of formation of flavour properties of meat products show the effectiveness of introducing FA containing monosodium glutamate at the final stage of mixing/cutting, after the introduction of fat-containing raw materials.

The results of chromatographic analysis of nonvolatile substances responsible for the taste of semi-finished meat indicate that when replacing a part of raw meat, the dominant factor, deteriorating the taste of the product when introducing additives, is the quality and quantity of pork fat containing a substantial amount of unsaturated fatty acids that undergo rapid oxidation and, as a consequence, deteriorate the taste of meat product. Carbohydrate-containing components (soy protein and starch) have almost no negative impact on taste when replacing part of the meat, as evidenced by almost no decrease in quality indicators of the studied meat products based on pork raw material.

Using more fat-containing raw materials leads to absorption by fat droplets of non-volatile substances responsible for taste that can cause deterioration of the flavour profile of the product. Noticeable effect of known meat taste precursors — mono-additives inosine and carnosine, when they were added together with monosodium glutamate, was not detected.

The most effective compositions of food additives that enhance the taste of meat products were mixtures of sodium glutamate with yeast extract and sodium glutamate with ribotide in dosages of 0.05% of each component by weight of raw materials.

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