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Quality of pig meat after supplementation of feed with humic acids

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Abstract: The animals used in this experiment were divided into two age categories: the weaning category at 28 days of age and the pre-fattening category at 2 months of age. In both age groups, the animals were further divided into a control group that did not receive humic acid supplementation and an experimental group that did. Supplementation with humic acids improved the meat's water retention during cooking. Samples from the experimental group showed lower weight loss than those from the control group. The effect of humic acids on meat protein content was positive. In the experimental samples, the average protein content was higher than in the control. Based on the results, we can say that feeding humic acids had a positive effect on prolonging the shelf life of pig meat by reducing the rate of fat oxidation.

I. Introduction

Compared to other livestock farming, pig farming offers many advantages, such as high fertility, relatively short generation intervals, efficient feed utilization, rapid growth, and the good nutritional and culinary properties of pork from modern hybrids. Every breeder therefore strives to maximize productivity in terms of both reproduction and production indicators [6].

Nowadays, we can observe a growing trend towards healthier animal products. This has resulted in increased demand for animal husbandry, creating a need for more natural and non-residual feed additives. Animal welfare and consumer needs are among the primary reasons for the high demand today for alternatives to the feed additives used in the past. The ban on the use of antibiotic growth promoters, cost-effectiveness, and the harmful effects of residues have also contributed to this [8].

To increase production indicators in pig farming, we can use humic substances, which can improve weight gain, feed intake, feed conversion, and fecal consistency. Replacing antibiotics with humic acids eliminates the risk of antibiotic residues in products and reduces the development of microbial resistance to these antibiotics.





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Humic substances are formed by the association of various components involved in humification processes (pectin, lignin, amino acids, carbohydrates) through intermolecular forces (hydrophobic, hydrophilic, ionic, donor-acceptor). The mechanism of humic substance formation is also strongly influenced by biological, geographical, and physical conditions [13]. The organic matter of humic substances consists mainly of mixtures of microbial and plant components in various stages of decomposition [2].

Humic substances are divided into three components based on their solubility in different media: humic acids, fulvic acids, and humic acids. Due to this multifunctionality, humic substances are one of the strongest chelating agents among organic natural substances [9].

The addition of natural humic substances (Humacid 60 preparation) at a concentration of 0.5% in the feed ration had a positive effect on weight gain in weaned piglets aged 25 to 70 days, reduced mortality rates, and at a concentration of 0.7% in complete mixtures for broiler chickens, it increased growth intensity, reduced feed consumption, and also reduced the percentage of dead individuals [14].

Slavik [12] demonstrated an increase in weight and a positive effect on feed conversion in pigs when observing a preparation containing humic acids at a concentration of 0.5-1%.

After the use of humic acids, which covered the exposed nerve endings in the mucosa, a reduction in intestinal irritability and in reactions to stressful conditions was observed in piglets with intestinal mucosal lesions [12].

Conversely, excessive addition of humic substances can also adversely affect breeding parameters. Písařiková and Herzig [10] found that when 3% sodium humate was added to pig feed, fattening pigs had lower weight gain, worse feed conversion, and lower nutrient digestibility in fattening pigs with comparable feed intake, which can be attributed to the ability of humate to create various types of binding interactions that can negatively affect nutrient resorption at the given concentration.

Meat quality is influenced by the components contained in the feed. The process of converting muscle tissue into meat is very complex and involves both physical and biochemical changes. It is a change in which muscle tissue, as a metabolically active system capable of stretching, is converted into meat, and thus into an inactive system incapable of stretching. The extent and speed of this change directly affect the properties of meat and its subsequent use for food [3].

Ji et al. [4] observed a redder color of meat in pigs fed humic substances containing 54.6% humic acids. However, when pigs were fed humic substances with a lower humic acid content (12.2%), this change was not observed. This means that humic acids can influence some of the qualitative properties of pig meat.

II. Material and Methods

The experiment used samples from six weaned piglets aged 28 days and six piglets from the pre-fattening age category aged 2 months.

The weaned piglet group consisted of three control group animals (not supplemented with humic acids) and three experimental group animals (supplemented with humic acids). In the pre-fattening category, 3 animals from the control group (not supplemented with humic acids) and 3 animals from the experimental group (supplemented with humic acids) were used. The experimental piglets received humic acid in HUMALAC Natur AFM Liquid per os at a dose of 2 ml/piglet/day until 10 days of age. Later, the experimental pigs received HUMALAC Natur AFM at a concentration of 0.5% of the feed mixture.

Determination of water and dry matter content: the principle of this method is to measure the weight loss of the sample at 170 ± 2 °C [11]. An aluminum weighing pan was weighed with an accuracy of 0.01 g, and then 10 g of ground sample was measured into it. The drying process continued until a constant weight was reached.

Determination of protein content: the homogenized sample was transferred to a Kjeldahl tube, poured with 15 ml of concentrated sulfuric acid, and then a catalyst (copper sulfate) was added. The prepared sample was titrated with 0.2 M HCl using a Kjeltec Auto 1030 Analyzer (Tecator, Sweden).

Determination of fat content by indirect extraction: after drying, the sample was quantitatively transferred to weighed filter paper using cotton wool and benzyl alcohol, then placed in a Soxhlet extraction device, and extraction was started. After cooling and drying in a desiccator, the sample was weighed.





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Determination of decomposition changes – Fat decomposition changes were evaluated by analyzing the thiobarbituric acid (TBA) value. This expresses the amount of malondialdehyde (MDA), a decomposition product of secondary fat damage caused by the oxidation of unsaturated fatty acids. The method was performed according to Marcinčák et al. [5].

III. Results and Discussion

A comparison of the differences in weight loss of shoulder, loin, and thigh samples from the control and experimental groups in the pre-fattening category in the cooking test showed lower meat weight loss in the experimental pigs. When comparing shoulder weight loss, we found that the experimental sample showed a slightly lower weight loss, specifically 0.57% less compared to the control group. The loin sample showed a significant difference in weight loss between the groups. The control group lost 19.15% more than the experimental group. The thigh sample from the control group lost 6.65% more weight during cooking compared to the experimental group. The samples from the experimental group lost less weight after cooking than the samples from the control group. The biggest difference was in the thigh sample, where the difference in water loss was 19.15% at a disadvantage of the control group.

Table 1. Chemical composition and pH of samples from loin in weaning category of pigs

Sample – Loin (weaning)	Control group	Experimental group	
Water	r 71.17 ± 1.8 72.3		
Fat	5.86 ± 2.9	4.82 ± 1.82	
Proteins	19.61 ± 1.5	20.74 ± 0.18	
Ash	1.39 ± 0.3	1.53 ± 0.08	
Collagen	1.36 ± 0.1	1.43 ± 0.05	
Pure muscle protein	15.67 ± 1.7	16.78 ± 0.42	
NaCl	0.15 ± 0.1	0.18 ± 0.11	
рН	5.79 ± 0.1	5.75 ± 0.07	

The water content in the loin samples taken from the control group in the pre-fattening category was on average 0.6% higher than in the experimental group (Table 4).

In the shoulder samples from the control group's weaning category, the average water content was 1.16% higher. Conversely, in the pre-fattening category, we can see that the water content in the shoulder samples is on average higher in the experimental sample, by 2.1% (Table 2, Table 5).

Fat content – In loin samples from the weaning category, the average fat content was higher in the control group, specifically by 1.04%. In the pre-fattening category, the average fat content was also higher in the control samples by 1.7% (Table 1, Table 4).

In the pre-fattening category, the difference in fat content in the shoulder between the experimental and control samples was 2.2%, with a higher fat content recorded in the control samples (Table 5).

In the pre-fattening category, the control group again had a higher average fat content in the thigh, specifically by 1.4% (Table 6).

Protein content – Loin samples from the weaning category contained 1.13% more protein in the experimental group. Samples from the experimental group in the pre-fattening category also contained 2.5% more protein (Table 1, Table 4). In samples from the shoulder of the weaning category in the control group, the protein content was 5.72% lower than in the experimental group (Table 2). The average protein content in samples from the thighs of the control group in the weaning category was 1.63% lower than in the experimental group. In the pre-fattening





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category, the experimental group also contained 0.9% more protein than the control group (Table 3, Table 6). In general, we found a higher protein content in meat samples from the experimental group.

Ash content – The ash content in samples from the control group of the weaning category was slightly lower than in the experimental group (Table 1). The difference between the control and experimental samples from the shoulders of the weaning category in ash content was 0.38%, with a higher content in the control sample. In the pre-fattening category, the ash content in the shoulder was higher in the experimental group (Table 2, Table 5). The percentage of ash contained in samples obtained from the legs of the control group in the weaning category was 0.28% higher than in the experimental group (Table 3).

Table 2. Chemical composition and pH from shoulders samples in weaning pig category

Sample – Shoulder (weaning)	Control group	Experimental group
Water	73 ± 1.6	71.84 ± 2.81
Fat	5.0 ± 1.1	4.46 ± 1.27
Proteins	19.6 ± 0.7	25.32 ± 9.74
Ash	1.3 ± 0.1	0.92 ± 0.48
Collagen	1.4 ± 0.1	1.61 ± 0.28
Pure muscle protein	16.1 ± 0.4	18.54 ± 4.91
NaCl	0.3 ± 0.1	0.26 ± 0.20
pН	5.9 ± 0.2	5.85 ± 0.17

Table 3. Chemical composition and pH of samples from thighs in weaning pig category

Sample – Thighs (weaning)	Control group	Experimental group
Water	73.9 ± 0.3	73.64 ± 2.52
Fat	3.1 ± 0.5	3.88 ± 1.15
Proteins	20.8 ± 0.7	22.43 ± 7.37
Ash	1.6 ± 0.1	1.32 ± 0.49
Collagen	1.4 ± 0.1	1.51 ± 0.23
Pure muscle protein	17.1 ± 0.7	18.19 ± 4.08
NaCl	0.3 ± 0.1	0.42 ± 0.36
рН	5.8 ± 0.1	5.78 ± 0.06

Table 4. Chemical composition and pH of samples from loin in pre-fattening pig category

Sample – Loin (pre-fattening)	Control group	Experimental group
Water	72.5 ± 0.4	71.9 ± 1.1
Fat	6.1 ± 1.5	4.4 ± 0.7
Proteins	23.0 ± 1.7	25.5 ± 5.3
Ash	1.7 ± 0.2	1.7 ± 0.1
Collagen	1.5 ± 0.1	1.6 ± 0.1
Pure muscle protein	16.8 ± 0.2	17.9 ± 0.7
NaCl	0.2 ± 0.1	0.1 ± 0.1
рН	5.8 ± 0.2	6.1 ± 0.2





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Collagen content – After taking loin samples from the control group of the pre-fattening category, a difference in collagen content of 0.1% was found in favor of the experimental group (Table 4). The collagen content in samples taken from the shoulders of the experimental group in the weaning category was 0.21% higher than in the control group (Table 5). The average percentage of collagen in control samples taken from the thighs of the weaning category was higher in the experimental group. The difference between the two groups in collagen content was 0.11% (Table 3).

Pure muscle protein content – Loin samples from the experimental weaned pigs contained 16.7% pure muscle protein. The difference between the experimental and control groups was 1.1% in favor of the experimental group. In loins samples of pre-fattening category, a difference of 1.1% was also found between the groups, in favor of the experimental group (Table 1, Table 4). The pure muscle protein represented in shoulder samples from the control group of the weaning category was 2.44% lower than in the experimental group. A higher content of pure muscle protein was recorded by 0.4% in the experimental group also in the samples of the pre-fattening category (Table 2, Table 5).

The samples from the control group in the weaning category had 1.1% less pure muscle protein than the experimental group. Even in the pre-fattening stage, thigh samples from the experimental group had a 0.6% higher pure muscle protein content than the control (Table 3, Table 6).

Table 5. Chemical composition and pH from shoulders in pre-fattening pig category

Sample – Shoulder (pre-	Control group	Experimental group	
fattening)			
Water	72.2 ± 0.3	74.3 ± 0.9	
Fat	6.3 ± 0.7	4.5 ± 0.7	
Proteins	20.8 ± 0.4	20.7 ± 0.2	
Ash	1.3 ± 0.3	1.5 ± 0.3	
Collagen	1.5 ± 0.1	1.4 ± 0.1	
Pure muscle protein	15.9 ± 0.4	16.3 ± 0.2	
NaCl	0.2 ± 0.2	0.2 ± 0.1	
pН	5.9 ± 0.1	6.1 ± 0.2	

Table 6. Chemical composition and pH from thighs samples in pre-fattening pig category

Sample – Thighs (pre-fattening)	Control group	Experimental group	
Water	72.4 ± 1.0	72.6 ± 0.7	
Fat	6.0 ± 1.3	4.6 ± 1.2	
Proteins	21.5 ± 0.5	22.4 ± 1.5	
Ash	1.2 ± 0.1	1.3 ± 0.2	
Collagen	1.5 ± 0.1	1.5 ± 0.1	
Pure muscle protein	16.3 ± 0.2	16.9 ± 0.9	
NaCl	0.1 ± 0.1	0.1 ± 0.1	
pH	5.9 ± 0.2	6.0 ± 0.2	

pH value – Samples taken from the weaning category had a meat pH range of 5.75 to 5.9 in both groups. In the pre-fattening category, the pH in both groups ranged from 5.8 to 6.1.

The technological quality of pork is strongly influenced by physicochemical properties such as water retention, tenderness, yield, shelf life, intensity, and homogeneity of meat color. Many of these properties are strongly influenced by postmortem changes in muscle pH. It follows that pH is an important factor in differences in meat



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quality [1]. If the decrease in meat pH is limited, a condition called DFD (Dark, Firm, Dry) occurs, specifically dark, firm, and dry meat. If the decrease in meat pH is too great, the meat becomes pale with lower yield, soft, and watery, resulting in PSE (pale, soft, exudative) [7].

It should be noted that the muscle does not stop functioning at the exact moment the animal is slaughtered. Pyruvate generated during glycolysis begins to convert to lactic acid, but without the blood system, this acid is not excreted from the muscles and therefore accumulates in them.

During the first few hours, the concentration of ATP is relatively stable and then begins to decline gradually. The onset of this decline is associated with the depletion of creatine phosphate. This is usually depleted before the pH reaches 6 [3]. Many post-mortem changes related to proteins have also been reported. During the first week after the animal's death, total or partial degradation of myofibrillar proteins such as nebulin, desmin, vinculin, and troponin T is observed. However, the enzymes responsible for this degradation have not yet been identified; it is assumed that proteolytic enzymes known as calpains are responsible. Calpains are a group of proteases that are activated by calcium. The current hypothesis is that after ATP depletion, calcium from the sarcoplasmic reticulum flows into the cytosol, where it activates calpain proteases, which begin degradation. An early decrease in pH is often accompanied by an increase in intracellular calcium, which leads to an increase in the activity of the proteases [3].

Table 7. MDA(Malondialdehyde) content after storage in the pre-fattening category

MDA (mg/kg)	Loin	Shoulder	Thigh
Control	0.3554	0.3762	0.3426
Experiment	0.3168	0.3184	0.3428

Table 7 shows the average MDA (malondialdehyde) values recorded in meat samples from the loin, shoulder, and thigh after storage. These values provide us with important information about the degree of fat oxidation in meat, which directly affects its sensory properties, such as aroma, taste, and overall quality. The degree of fat oxidation and thus the concentration of malondialdehyde were lower in the experimental group than in the control group in the loin and shoulder samples. The table allows us to evaluate the extent to which humic acid supplementation could affect fat stability compared to the control group.

IV. Conclusion

Differences in water and weight loss during cooking between the control and experimental groups suggest that humic acid supplementation may enhance meat's ability to retain water during cooking. Samples taken from the experimental group showed lower weight loss compared to the control group. Supplementation with humic acids in the weaning category may lead to lower fat content in some parts of the meat, such as loin and shoulder. The effect of humic acids on the protein content of meat is positive; in most experimental samples, the protein content was higher than in samples from the control groups. The results of this study suggest that humic acid supplementation increases collagen content in pork, potentially improving its structure, texture, and technological properties. The results in both categories clearly indicate that humic acid supplementation has a positive effect on the pure muscle protein content in pork. This effect may influence the nutritional value of meat as well as its technological properties and overall quality. Based on the results obtained, we can say that feeding humic acids has a positive effect on extending the shelf life of pork by reducing the rate of fat oxidation, thereby potentially improving its stability and quality during storage.





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