



Morphological characteristics of the digestive tube organs in broiler chickens after feeding coarsely ground mussel shells

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Abstract. The relevance of the study is conditioned by the practical and scientific interest in feeding mineral feed additives from marine aquatic organisms and the effect of their particle

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size on the performance and condition of the digestive system in broiler chickens. In this regard, the purpose of the present study was to determine the effects of a feed additive made from sea mussel shells by coarse grinding on the growth rate and structure of the digestive organs of broiler chickens. To clarify this issue, histological studies of the tube-like digestive organs of chickens of the control and experimental groups were conducted, and their morphometric parameters were determined. Broiler chickens of the experimental group were fed with coarsely ground sea mussel shells in addition to the standard diet by free feeding. According to the findings of the study, the body weight of chickens in the experimental group stayed unchanged. In chickens of the experimental group, a decrease in the weight of the muscular part of the stomach and intestines and an increase in the length of the small intestine were found. An increase in the thickness of the epithelium and the area of the secretory portions of glands was observed in the crop and oesophagus, and an increase in the thickness of the cuticle and the mucosal lamina propria in the muscular part of the stomach. In the mucous membrane of the oesophagus and proventriculus, a larger number and area of lymphoid tissue accumulations in the form of small diffuse fields and lymph nodules were detected. In the duodenum of broiler chickens, the height of villi and the depth of crypts were lower, and in the caecum, the depth of crypts was lower. The morphological features of the crop, oesophagus, stomach, and small intestine, established upon the use of a mineral feed additive from mussel shells indicated the development of protective and adaptive reactions of the oesophagus and stomach to the traumatic effect of coarse particles of mussel shells, and the small intestine – to the increased calcium content in the chyme. The experimentally substantiated feeding of mineral feed additives from mussel shells to broiler chickens proved the need for their thorough grinding and dosed use in poultry feeding

Keywords: poultry; mineral feed additive; marine hydrobionts; oesophagus; stomach; intestine; morphometric parameters

Introduction

Chicken is a valuable poultry worldwide, providing people with meat and eggs. Presently, broiler chicken breeding is the most intensive livestock industry, characterised by a short growth period, high feed to animal protein conversion, and relatively low capital investment. Chicken meat is the most accessible product for multiple segments of the population, cultures, and religions around the world. According to Y. Wang *et al.* (2024), as the world's population and food consumption grows, poultry will be the most common source of meat. However, to be able to sustainably meet the growing demand, it is necessary to optimise all components of poultry production, considering the

use of feed additives. N. Barzkar *et al.* (2024) pointed out that marine aquatic organisms can be one of the sources of unconventional feed additives in poultry feeding. L. Peshuk *et al.* (2023) emphasised that the hydrosphere occupies approximately 70% of the planetary surface, providing a wide range of diverse natural resources. Considering the global shortage of food resources necessary to maintain a healthy and active lifestyle, the study of alternative sources of human food and animal feed, including marine bioresources, is becoming increasingly relevant. According to J. Feng *et al.* (2025), in China, the use of marine biomass, primarily mussels, crabs, and scorpions, has

been identified as key species in the development of ecological “sea farming”. According to O. Golubenko *et al.* (2023), the breeding of marine aquatic organisms plays a prominent role not only in maintaining the aquatic ecosystems of Ukraine but is also an essential component of such a profitable industry as aquaculture. A.A. Menchynska *et al.* (2022) proved that the development of technologies to produce various foods from marine aquatic organisms is promising for the food industry of Ukraine. The researchers experimentally confirmed the effectiveness of combining crustaceans with fish, animal, and plant raw materials. At the same time, hydrobionts are underutilised in the feeding of farm animals, despite their considerable potential.

Y. Zhang *et al.* (2022) found that for wild birds, aquatic organisms such as marine mussels are a regular food. S.A. Siddiqui *et al.* (2025) emphasised the significance of research aimed at the use of aquaculture facilities in animal diets in the context of the problem of shellfish shell utilisation. The latter are by-products of the primary processing of marine mussels, accumulating at processing plants, creating considerable environmental and economic challenges. H.K. Zanu *et al.* (2023) noted the practical and scientific significance of investigating the effects of mineral components and particle size distribution of mineral feed additives on the performance of poultry. Therewith, the researchers noted that the particle size of such additives affects the quantitative and qualitative indicators of productivity and depends on the duration of their presence in certain organs of the digestive tract of animals.

The use of marine mollusc shells in poultry feed is a long-standing practice in agriculture. The availability of significant extensive of marine aquatic organisms primary processing waste at the enterprises of Odesa region, as well as the potential economic effect,

encouraged their use in livestock production as mineral feed additives. At the same time, due to the imperfect method of coarse grinding of sea mussel shells, the particles of this product have varying sizes and sharp edges, which can pose a potential danger to the health of poultry when fed. Considering the lack of information on the effects of the use of a mineral feed additive obtained by coarsely grinding sea mussel shells on the morphological and functional organisation of the poultry digestive system, it is prompt and relevant to explore this issue. The purpose of the present study was to determine the specific features of macro- and microscopic structure of the tube-like digestive organs in broiler chickens when using a mineral feed additive made from coarsely ground sea mussel shells in the diet.

Literature Review

The positive experience of using both aquatic organisms and their primary processing waste in poultry feeding is evidenced by a series of publications. According to E. Toyas-Vargas *et al.* (2018), the inclusion of 5% of crushed entrails of scallops, squid, shrimp, and blue mussel flour in the diet of laying hens increased the carotene content of egg yolks and was a better alternative to fish meal. W.D. Lee *et al.* (2021) found that the use of oyster shells in chicken feed contributed to higher bone mineral density in the lower leg. According to K.E. Buğdaycı *et al.* (2019), the positive effects of limestone and Mediterranean mussel shells on the productive and interior indicators of Japanese quail: body weight, eggs, feed consumption, feed conversion, yolk index, calcium and phosphorus content in the shell and serum was proved. R. Xing *et al.* (2019) found that a mineral feed additive made from scallop shells was a more effective source of active dicalcium phosphate than chicken eggshell powder. Its addition to the main diet increased the average

daily gain in live weight, muscle yield, and reduced the content of abdominal fat.

S.A. Lee *et al.* (2021) indicated that the particle size of a mineral feed additive affects the condition of the pectoral muscles of broiler chickens. The use of small limestone particles in the diet showed a tendency to improve the quality of pectoral muscles. B.S. Dongare *et al.* (2024) noted that the use of calcium from the diet by birds is influenced by its source, particle size, bioavailability, etc. Insufficient inorganic substances in bone tissue causes skeletal disorders in broilers with high body weight, which leads to significant economic losses. The use of specially coated limestone particles as a source of calcium contributed to an increase in body weight, better feed conversion, and greater bone strength in broiler chickens.

According to D. Joardar *et al.* (2020), a comparative study on the inclusion of limestone particles of varying sizes in the main diet of laying hens showed better mineral utilisation in the digestive tract when coarse particles were fed. At the same time, according to I.J. Bueno *et al.* (2016), the size of limestone particles in the feed did not affect the performance of poultry, both parent flocks and broiler chickens. The researchers did not observe any substantial difference in egg production, egg weight, shape index, and specific weight of eggs, eggshell weight, egg hatchability, body weight, and bone morphology of chicks.

According to W. Awad *et al.* (2008), high calcium content in the diet causes a decrease in the height of the villi of the small intestine in broiler chickens. However, according to Q.J. Wu *et al.* (2013), the intestinal villi height is lower when the mineral feed additive clinoptilolite is used in the diet of broiler chickens. According to R. Xing *et al.* (2019), the use of scallop shell powder in the feeding of broiler chickens contributed to a decrease in crypt

depth and an increase in the ratio of villi height to crypt depth in the intestine.

Thus, as the literature analysis suggests, most researchers noted a positive impact on the state of poultry productivity of the use of marine aquatic organisms as a mineral feed additive. There is no consensus on the effects of the particle size of such additives on the state of the poultry organism. There is also no information on the effects of mineral feed additives made from marine aquatic organisms in the form of coarsely ground mollusc shells on the state of the digestive system, which was determined the purpose of the present study.

Materials and Methods

An experiment on feeding a mineral feed additive was conducted at the Budaki poultry farm in Odesa region on Ross 308 broiler chickens during 2024. Poultry were kept in a poultry house in group cages on the floor with soft bedding. The feed additive was produced by coarse mechanical grinding of Black Sea mussel (*Mytilus galloprovincialis*) shells to obtain particles up to 10 mm in size. The finished feed additive was used by free feeding along with the main diet.

Two groups of clinically healthy poultry of 14 days of age, 20 birds each, were formed according to the principle of pairwise analogues. After a preparatory period of 6 days, chickens of the experimental group from 20 to 42 days of age were additionally fed ground mussel shells as a mineral feed additive to the main diet. The feeder with the mineral feed additive was located next to the feeder containing the main feed. Broiler chickens of the control group received only the basic diet. The feed composition of the main diet included (by weight): maize – 32%, wheat – 25%, sunflower cake – 9%, extruded soybeans – 16%, soybean cake – 18%, and protein and vitamin complex *Biomix®Broiler* 1.5% at 15 g/kg of feed. During the experiment, the broiler chickens were allowed to walk and had

free access to clean and fresh drinking water. The chickens were kept and treated following the international principles of the European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Purposes (European Convention ..., 1986) and the Law of Ukraine No. 3447-IV "On the Protection of Animals from Cruelty" (2006).

At 42 days of age, the live weight of chickens of the control and experimental groups was determined. After slaughtering the chickens, the weight of the glandular and muscular parts of the stomach, as well as the weight and linear indicators of individual intestines were determined. During the selection of internal organs, their topography, shape, size, and colour were determined. The boundary between the jejunum and ileum was determined by the tops of the caeca. The body weight of chickens and their digestive organs was measured using electronic scales A-250P (Poland), and the length of individual intestines was measured using a ruler. The relative weight of organs (%) was calculated as the ratio of their absolute weight to body weight, and the relative length of individual intestines (%) was calculated as the ratio of their absolute length to the length of the entire intestine.

Morphological studies were performed in the histological laboratory of the Department of Normal and Pathological Morphology of the State Biotechnological University (Kharkiv). For histological studies, pieces of the oesophagus, thoracic oesophagus, glandular and muscular parts of the stomach, duodenum, and jejunum were taken from their middle sections – 5 samples from each group of broiler chickens of different sexes. After fixation in a 10% solution of neutral formalin, the pieces of material were washed in running water, dehydrated and sealed in alcohols of increasing concentration, kept in intermediate media using chloroform and embedded in paraffin. Histological sections

from paraffin blocks were made using a sled microtome MS-2 (Ukraine). Histological sections were stained with eosin and haematoxylin. Examination of histological specimens and micrographs were performed using a Jenamed 2 microscope (Carl Zeiss Jena, Germany). Microphotographs were taken using a Sigeta US-MOS 5100 SMP camera (China). The morphometric parameters of the microstructures of the digestive organs were determined on histological specimens using an ocular square grid (N=256) and Adobe Photoshop CS5 software on the obtained microphotographs.

The obtained digital data were processed statistically to determine the arithmetic mean (M) and statistical error (m). The reliability of the difference between the indicators was established according to the criterion of reliability (td) and Student's tables at three levels of probability $P < 0.05$, $P < 0.01$, and $P < 0.001$.

Results and Discussion

During the experiment, the feeding activity of broiler chickens was monitored. It was recorded that the birds of the experimental group showed a high attraction to the mineral feed additive, which was characterised by a specific "marine" scent reminiscent of fish. Additionally, an increase in the volume of drinking water consumed in this group was noted. At the initial stage of the study, the average body weight of 14-day-old chicks was 212.1 ± 13.3 g. At the end of the experimental period (42 days), the average body weight of broilers in the control group reached $1,284.3 \pm 57.3$ g, while in the experimental group this figure was $1,273.8 \pm 60.3$ g, which was 0.8% less than in the control group. The obtained findings were somewhat consistent with the data of N.R. Abdulla *et al.* (2016), who found a decrease in body weight of chickens with excessive calcium intake, which caused a decrease in the intensity of absorption of organic matter, crude protein, ether extract, and phosphorus.

According to V. De Gregorio *et al.* (2018), the intensity of animal growth is limited by the ability to absorb and digest feed, i.e., depends on the morphological and functional state of the digestive system. On the other hand, according to G. Lamprecht & P. Bodammer (2016), the composition of the feed itself, its physicochemical properties, determine the state of the digestive tract, which is essential

in the prevention and treatment of its pathologies and is a vital element of adaptation. The results of determining the absolute and relative weight of the glandular and muscular parts of the stomach and intestines, as well as the absolute and relative length of individual intestines of broiler chickens when using a mineral feed additive made from coarsely ground sea mussel shells in the diet are presented in Table 1.

Table 1. Macroscopic parameters of digestive organs in experimental broiler chickens ($M \pm m$, $n = 5$)

Indicator	Group	
	control	experimental
Weight of the glandular part of the stomach, g	5.9 ± 0.2	5.3 ± 0.2
Relative weight of the glandular part of the stomach, %	0.5 ± 0.02	0.4 ± 0.02
Weight of the muscular part of the stomach, g	23.1 ± 1.0	18.4 ± 1.0*
Relative weight of the muscular part of the stomach, %	1.8 ± 0.1	1.4 ± 0.1**
Intestinal weight, g	73.6 ± 4.5	55.5 ± 2.8*
Relative intestinal weight, %	5.7 ± 0.2	4.4 ± 0.2**
Absolute intestinal length, cm	150.3 ± 8.3	180.8 ± 10.4
Absolute length of the small intestine, cm	120.8 ± 6.0	152.0 ± 7.3*
Relative length of the small intestine, %	80.4 ± 2.2	84.1 ± 2.1
Absolute length of the large intestine, cm	29.5 ± 0.9	28.8 ± 1.3
Relative length of the large intestine, %	19.6 ± 1.3	15.9 ± 1.0
Absolute length of the duodenum, cm	27.1 ± 1.5	32.1 ± 2.0
Relative length of the duodenum, %	18.0 ± 0.2	17.8 ± 0.4
Absolute length of the jejunum, cm	82.3 ± 4.0	107.7 ± 6.8*
Relative length of the jejunum, %	54.8 ± 1.9	59.6 ± 1.7
Absolute length of the ileum, cm	11.4 ± 0.2	12.2 ± 0.5
Relative length of the ileum, %	7.6 ± 0.3	6.7 ± 0.4
Absolute length of the caecum, cm	23.3 ± 1.0	23.2 ± 0.5
Relative length of the caecum, %	15.5 ± 1.0	12.8 ± 0.7
Absolute length of the rectum, cm	6.2 ± 0.2	5.6 ± 0.2
Relative length of the rectum, %	4.1 ± 0.3	3.1 ± 0.4

Note: * $P < 0.05$; ** $P < 0.01$, statistically significant in relation to the control

Source: developed by the authors

According to the data obtained, the use of the feed additive caused a decrease in the absolute and relative weights of both the stomach and intestines (see Table 1). Compared with the control group, the absolute weight of the glandular and muscular parts of the stomach of the experimental group chickens decreased by 10.2% and 20.3% ($P < 0.05$), respectively, while the intestinal weight – by 24.6% ($P < 0.05$). Their relative indices also decreased accordingly: the relative weight of the glandular part of the stomach – by 0.1%, the muscular part of the stomach – by 0.4% ($P < 0.01$), and the intestine – by 1.3% ($P < 0.01$).

In parallel with the decrease in weight characteristics, the broilers of the experimental group showed an increase in the linear length of the intestine by 20.6% compared to the control. Therewith, the increase in the total length of the digestive tract was mainly due to the small intestine, the absolute length of which increased by 25.8% ($P < 0.05$), while the large intestine showed a decrease of 2.4%. Such transformations led to an increase in the relative length of the small intestine by 3.7% and, accordingly, to a decrease in the relative length of the colon by a corresponding amount. Analysis of the morphometric parameters of individual intestinal sections revealed that in the experimental group, the absolute length of the duodenum increased by 18.5%, the jejunum – by 30.9% ($P < 0.05$), and the ileum – by 7.0%. The relative length of the duodenum underwent a slight decrease of 0.2%, the jejunum showed an increase of 4.8%, while the ileum – a decrease of 0.9%. Therewith, the absolute length of the large intestine, the relative length of the caecum and rectum did not change significantly. Thus, the use of a mineral feed additive caused the greatest macroscopic changes in the jejunum.

Thus, according to the findings of the study on the use of coarsely ground mussel shell

particles in the diet of broiler chickens, there were significant changes in the morphometric parameters of the glandular and muscular parts of the stomach and jejunum. As reported by H. Zhang *et al.* (2019), the small intestine plays a key role in digestion and nutrient adsorption. According to N.A. Abd El-Azeem *et al.* (2023), the development of the gastrointestinal tract is crucial for maximising nutrient utilisation and productive growth of chickens. According to M.M. Kushch *et al.* (2019), the relative weight of the small intestine and its length were greater in heavy geese compared to light geese.

The experimental data obtained during the study demonstrate congruence with the observations of Q.J. Wu *et al.* (2013) on the lengthening of the small intestine and reduction of its weight when natural and synthetic clinoptilolite, a mineral with a high calcium content, is introduced into the broiler diet. An analogous trend can be observed in the report by R. Xing *et al.* (2019), who recorded an increase in the linear dimensions of the duodenum, small intestine, and ileum in broilers when using ground scallop shells as a source of active dicalcium phosphate.

Overall, the histological structure of the tube-like digestive organs of broiler chickens corresponded to the general patterns of their structure in other poultry species and was consistent with the information of R.R. Beheiry (2018) on their structure in domestic turkeys and D.S. Makhotina *et al.* (2020) on their structure in domestic ducks. Microscopically, the wall of both the oesophagus and thoracic oesophagus of broiler chickens consisted of three membranes: mucosa, muscularis, and adventitia. The submucosa was not found. The characteristic structure of the oesophageal wall was longitudinal folds. They were formed not only by the mucosa but also by the inner layer of the muscularis (Fig. 1).

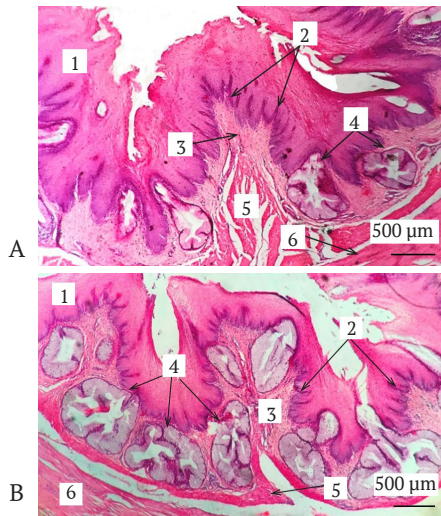


Figure 1. Secretory glands in the wall of the thoracic part of the oesophagus of chickens of control (A) and experimental (B) groups

Note: 1 – epithelial layer of the mucous membrane; 2 – basal membrane of the epithelium; 3 – lamina propria of the mucous membrane; 4 – secretory divisions of glands; 5 – inner layer of the muscular membrane; 6 – outer layer of the muscular membrane

Source: developed by the authors

The mucous membrane of the crop and oesophagus consists of two layers: the epithelial layer and the lamina propria. As can Figure 1 (A, B) shows, the epithelial layer (1) is formed by multilayered squamous non-squamous epithelium, while the lamina propria (3) is formed by loose fibrous connective tissue. The epithelial layer is separated from the lamina propria by a convoluted basement membrane (2). The lamina propria of the thoracic part of the oesophagus contains the secretory divisions of the alveolar tubular glands (4), which were round or oval in shape and formed by prismatic glandular epithelium. The glands were adjacent to the basal membrane of the epithelial layer with one edge, and to the inner layer of the muscularis membrane with the other (5). Due to the direct adhesion of the glands to the basal membrane, their secretory compartments opened into the

epithelial layer, and no excretory ducts were found. The muscularis membrane consisted of two layers: the inner longitudinal and the outer circumferential (6), formed by bundles of smooth muscle tissue and separated by a thin layer of loose fibrous connective tissue.

The stomach of chickens consisted of two parts: glandular, or proventriculus, and muscular, or gizzard, which were connected by a short isthmus, which is consistent with the data of R. Pachauri *et al.* (2024) on its structure in other bird species. The surface of the mucous membrane of the stomach parts was uneven: in the proventriculus, it contained papillae, which opened the glandular ducts, and in the gizzard – low folds formed by the cuticle. In the wall of both the glandular (Fig. 2) and muscular parts of the stomach (Fig. 3) of broiler chickens, four membranes were distinguished: mucosa, submucosa, muscular, and serous.

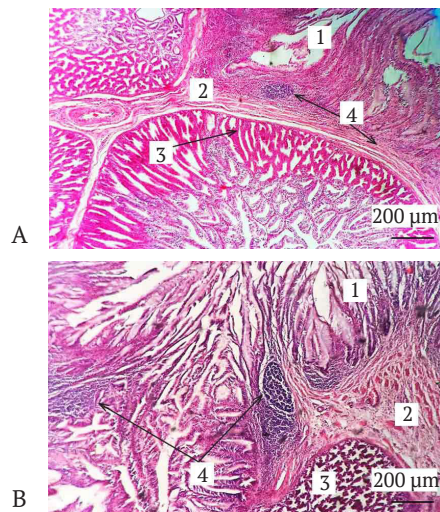


Figure 2. Mucous membrane of the proventriculus of chickens of control (A) and experimental (B) groups

Note: histological preparation, stained with haematoxylin and eosin; 1 – epithelial layer of the mucous membrane; 2 – lamina propria of the mucous membrane; 3 – lobule tubes; 4 – lymphoid tissue

Source: developed by the authors

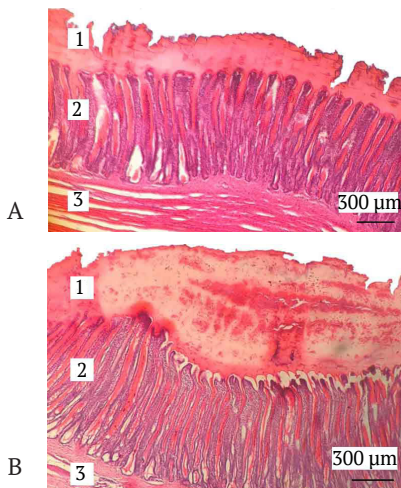


Figure 3. Cuticle and glands in the lamina propria of the gizzard of chickens of control (A) and experimental (B) groups

Note: histological preparation, stained with haematoxylin and eosin; 1 – cuticle; 2 – gastric glands; 3 – muscular membrane

Source: developed by the authors

The mucous membrane (Fig. 2) consisted of two layers: epithelial and lamina propria. The lamina muscularis mucosae was not detected. In the mucosa of the glandular part of the stomach, a glandular layer or superficial glands were noted, which were formed by protrusions of the lamina propria (2) covered with a single-layer prismatic epithelium (1), as well as deep glands located in the submucosa. Due to the significant content of such glands in the entire proventriculus wall, the submucosa was the thickest. The glands in the submucosa were densely arranged lobules of predominantly conical shape with a wider base and narrower apex, separated by thin layers of loose fibrous connective tissue. The lobules consisted of numerous tubes that radially converged to its central outflow duct (3).

In the muscular part of the stomach (Fig. 3), the epithelial layer of the mucosa was covered by a cuticle (1). The mucosa located under the cuticle formed the glandular layer (2), which

consisted of densely arranged tube-shaped glands, representing the immersion of the epithelium in the lamina propria. The lower part of the glands was formed by secretory compartments, while the upper part – by excretory ducts that brought the secretion from which the cuticle was formed to the surface of the epithelial layer. The submucosa in the form of a wide strip separated the mucous membrane from the muscular membrane and was formed by a dense interweaving of connective tissue fibres. The muscular membrane (3) is formed by bundles of smooth muscle cells, mostly with a circular orientation.

In the small intestine of broiler chickens, the duodenum, jejunum, and ileum were identified. The duodenum formed a characteristic loop that contained the pancreas and was separated from the jejunum by the duodenal papilla. The jejunum formed 10-12 loops, the border with the ileum was set at the level of the tops of the caeca. The intestinal wall consisted of three membranes: mucosa, muscular membrane, and serous membrane (Fig. 4).

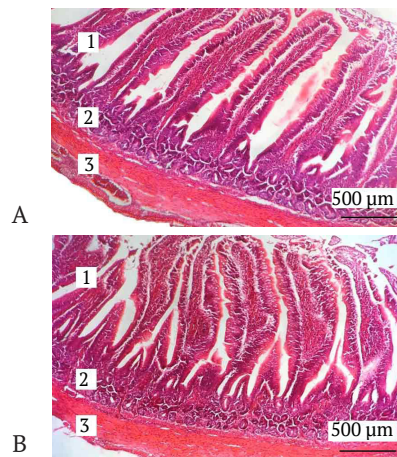


Figure 4. Duodenum of chickens of control (A) and experimental (B) groups

Note: Histological preparation, stained with haematoxylin and eosin, Sigeta; 1 – villi; 2 – crypts; 3 – muscular membrane

Source: developed by the authors of this study

The mucous membrane presented in Figure 4 consisted of three layers: epithelial, lamina propria, and muscularis mucosae. A feature of the mucous membrane relief was villi and crypts. Villi (1) were finger-shaped protrusions of the lamina propria covered with an epithelial layer. Crypts (2) were tube-shaped immersions of the epithelium into the lamina propria. The muscular lamina, in the form of a narrow chain of smooth muscle cells, was located directly under the bottom of the crypts and had a longitudinal direction of their location. The muscular membrane (3) of the intestinal wall consisted of two layers of smooth muscle tissue – a thicker inner circular layer and an outer longitudinal layer.

According to A.S. Davis *et al.* (2021), quantitative analysis of biological structures is a powerful tool that provides objective data that is challenging to obtain by other methods. It is morphometric studies that enable an unbiased morphological assessment of the state of the body's structures. The results of determining the morphometric parameters of the

microstructures of the crop, thoracic part of the oesophagus, glandular and muscular parts of the stomach, duodenum, and jejunum of broiler chickens in the control group and with the use of a feed additive are presented in Table 2.

According to the findings obtained (Table 2), feeding broiler chickens with coarsely ground sea mussel shells caused changes in the microscopic parameters of the digestive tract organs. Thus, in the oesophagus and thoracic oesophagus, the thickening of the epithelial layer of the mucous membrane formed by multilayered non-squamous immature epithelium was noted by 39.0% ($P < 0.001$) and 11.8%, respectively. Furthermore, in broiler chickens of the experimental group, the area of the secretory glands in the thoracic part of the oesophagus (Fig. 1, note 4) increased by 38.5% ($P < 0.05$) compared to the control group. In contrast to the control group, in broiler chickens of the experimental group, some of the secretory compartments contained a much larger cavity filled with mucous secretion.

Table 2. Microscopic parameters of digestive organs in experimental broiler chickens ($M \pm m$, $n = 5$)

Indicator	Group	
	control	experimental
Height of the epithelial layer of the crop mucosa, μm	457.5 \pm 12.5	775.0 \pm 15.0***
Area of the secretory glands of the thoracic oesophagus, μm^2	395.4 \pm 35.5	547.5 \pm 50.7*
Submucosal thickness of the glandular part of the stomach, μm	1,682.5 \pm 17.5	1,478.3 \pm 108.5
Area of lymphoid tissue accumulation in the submucosa of the glandular part of the stomach, μm^2	6,240.3 \pm 341.5	17,026.3 \pm 908.6***
Thickness of the cuticle of the mucous membrane of the muscular part of the stomach, μm	435.2 \pm 27.5	621.3 \pm 34.5**
Thickness of the glandular layer of the mucous membrane of the muscular part of the stomach, μm	465.3 \pm 32.5	686.0 \pm 38.3**
Thickness of the mucous membrane of the muscular part of the stomach, μm	902.4 \pm 16.4	1,305.5 \pm 79.8**
Height of duodenal villi, μm	1,534.9 \pm 102.4	1,243.1 \pm 56.0*
Depth of duodenal crypts, μm	344.7 \pm 24.7	243.5 \pm 64.7**
Ratio of villi height to duodenal crypt depth	4.5 \pm 0.02	5.1 \pm 0.2**
Height of the villi of the jejunum, μm	778.5 \pm 27.5	735.4 \pm 6.0
Depth of crypts of the jejunum, μm	274.8 \pm 23.8	184.9 \pm 19.1*
Ratio of villi height to depth of crypts of the jejunum	2.8 \pm 0.1	4.0 \pm 0.2*

Note: * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$ statistically significant in relation to the control

Source: developed by the authors

A feature of the structure of the glandular part of the stomach in broiler chickens of the experimental group was a smaller thickness of the submucosa containing glandular lobules. Compared to the control group, its thickness was 12.1% smaller ($P < 0.05$). Lymphoid tissue in the form of primary lymph nodules and diffuse fields was found between the secretory glandular compartments, as well as in the mucosal lamina propria (Fig. 2, note 4). Compared to the control group, their area increased by 172.9% ($P < 0.001$).

The muscular part of the stomach of broiler chickens in the experimental group was characterised by a greater thickness of both the cuticle and the mucous membrane lamina propria, which contained the secretory portions of the tubular glands that synthesise its components (Fig. 3). Compared to chickens of the control group, the secretory compartments of such glands were wider and contained wide strands of secretion in the form of vertical columns that passed into the cuticle. The use of a mineral feed additive in the diet increased the thickness of the cuticle of the mucous membrane of the muscular part of the stomach of broiler chickens (Table 2) by 42.8% ($P < 0.01$), the thickness of the mucous membrane lamina propria by 47.4% ($P < 0.01$), and the thickness of the entire mucous membrane by 44.7% ($P < 0.01$) compared to the control group.

Considering the form of the mineral feed additive – coarsely ground fragments of sea mussel shells – it can be assumed that changes in the micro- and macrostructural state of the thoracic part of the oesophagus, as well as the glandular and muscular parts of the stomach of broiler chickens, occurred due to their mechanical effect. The fact that sea mussels are a regular food for wild birds can be explained by the use of defence and adaptive mechanisms that have evolved over a lengthy period of adaptation to this type of food specialisation (Khomych *et al.*, 2021).

In broiler chickens of the experimental group, the study observed a decrease in the height of villi and the depth of duodenal crypts (Fig. 4) by 19.0% ($P < 0.05$) and 29.4% ($P < 0.01$), and in the jejunum – by 5.5% and 32.7% ($P < 0.05$), respectively. The findings of the study revealed that such a significant indicator of the morphological and functional state of the intestine of broiler chickens as the ratio of intestinal villi height to crypt length increased in the experimental group. Thus, in the duodenum, the ratio of villi height to crypt depth increased by 14.8%, in the jejunum – by 39.2%, which was, respectively, 5.1 ± 0.2 and 4.0 ± 0.2 against 4.5 ± 0.1 and 2.8 ± 0.2 in the control group.

T.D. Hinnant *et al.* (2025) reported that the absorption surface area of the small intestinal mucosa determines the ability of the entire intestine to absorb nutrients. Changes in villus height and crypt depth are prominent indicators of intestinal function. The intestinal crypts are the site of formation of enterocytes, the epithelial cells of the small intestine that ensure the performance of its functions. C.F. Marchini *et al.* (2011) found that a decrease in the depth of crypts and the height of duodenal villi of broiler chickens against the background of body weight loss is characteristic of such an unfavourable factor as heat stress. S. Gotoh *et al.* (2023) proved that the villi of the small intestine actively absorb calcium, and this process is more active at their tops.

The obtained findings of the study on the reduction of the height of the villi of broiler chickens when using mussel shells in the diet were consistent with the data of W. Awad *et al.* (2008) on the reduction of the height of the villi of the small intestine with a high calcium content in the diet of broiler chickens and contradicted the information of Q.J. Wu *et al.* (2013), according to which the use of clinoptilolite, which is a source of calcium, increased their height. The findings obtained in this study on the decrease in crypt depth and the increase

in the ratio of villi height to crypt depth of the intestine of broiler chickens using mussel shells were consistent with analogous data from R. Xing *et al.* (2019) on the use of scallop shell powder in broiler feed.

Thus, the presented studies of the morphological and functional state of the oesophagus and stomach of broiler chickens when fed mussel shells subjected to rough mechanical grinding indicated the need to use a mineral feed additive from mussel shells in the form of small particles or flour that would not have a traumatic effect on the mucous membrane of the digestive tract. Changes in the morphological parameters of the small intestine indicate a high calcium content in the diet, which requires the dosed use of a mineral feed additive, considering the needs of the poultry body and the total calcium content in the diet.

Conclusions

Free feeding of the mineral feed additive from mussel shells in addition to the main diet from 20 to 42 days of age did not affect the growth rate of broiler chickens, as indicated by the absence of a statistically significant difference in body weight of the control and experimental groups at the end of the experiment. At the same time, the findings of morphological studies revealed the effects of a mineral feed additive made from coarsely ground sea mussel shells on the anatomical and histological parameters of the structure of the tube-like digestive organs of broiler chickens: the oesophagus, glandular and muscular parts of the stomach, and intestines. At the end of the experiment, the absolute and relative weight of the muscular part of the stomach decreased by 20.3% and 0.4%, respectively, while the absolute and relative weight of the intestine decreased by 24.6% and 1.3%, respectively. Upon the use of a mineral feed additive, an increase in the length of the intestine of chickens by 20.6% was observed, which was caused by an

increase in its small section by 25.8%. Moreover, such changes were associated with a 30.9% increase in the length of the jejunum. The length of both the large intestine and its individual intestines did not undergo statistically significant changes. In the crop of the chickens of the experimental group, an increase in the thickness of the protective epithelial layer of the mucous membrane by 39.0% was observed, in the thoracic part of the oesophagus – an increase in the area of the secretory glands by 38.5%, the mucous secretion of which facilitated the passage of feed and performed a protective function. Between the secretory portions of glands in the submucosa of the glandular part of the stomach, an increase in the area of such protective structures as lymphoid tissue accumulation by 172.9% was observed. In the muscular part of the stomach, the cuticle thickness increased by 42.8% and the thickness of the secretory layer of the mucosa – by 47.4%. Feeding a mineral feed additive contributed to a decrease in the height of villi and the depth of the duodenal crypts by 19.0% and 29.4%, respectively, and the depth of the crypts of the jejunum by 32.7%. Such changes in the parameters of the main microstructures of the intestine caused an increase in the ratio of villus height to crypt depth in the duodenum by 14.8% and in the jejunum by 39.2%.

The detected modifications of the morphological and functional organisation of the digestive tract suggested the formation of protective and adaptive reactions in response to mechanical stimulation by mussel shell particles. The recorded structural characteristics of the digestive organs correlated with the generally accepted principles of adaptation of biological systems as a dynamically labile structure to environmental factors. It is promising to investigate the degree of assimilation of nutrients in the diet by the body of broiler chickens using feed additives that differ in the size of mussel shell particles. Such an investigation will

enable the production of feed additives that would ensure their most efficient use.

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Conflict of Interest

None.

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Морфологічна характеристика органів травної трубки в курчат-бройлерів за згодовування грубо мелених стулок мідій

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Анотація. Актуальність дослідження зумовлена практичним і науковим інтересом щодо згодовування мінеральних кормових добавок із морських гідробіонтів і впливу розміру їх частинок на продуктивність і стан органів травлення в курчат-бройлерів. У зв'язку з цим, мета роботи полягала у визначенні впливу кормової добавки, виготовленої зі стулок морських мідій шляхом грубого подрібнення, на інтенсивність росту та будову органів травлення курчат-бройлерів. Для з'ясування цього питання проведені гістологічні дослідження трубоподібних органів травлення у курчат контрольної і дослідної груп, визначено їх морфометричні показники. Курчатам-бройлерам дослідної групи додатково до стандартного раціону шляхом вільного згодовування використовували грубо подрібнені стулки морських мідій. За результатами дослідження маса тіла в курчат дослідної групи залишалася без змін. У курчат дослідної групи встановлено зменшення маси м'язової частини шлунка, кишечника і збільшення довжини тонкого відділу кишечника. У волі і стравоході відмічали збільшення товщини епітелію, площі секреторних відділів залоз, а у м'язовій частині шлунку – зростання товщини кутикули і власної пластинки слизової оболонки. У слизовій оболонці стравоходу і шлунку виявлено більшу кількість і площу

скупчень лімфоїдної тканини у вигляді дрібних дифузних полів і лімфатичних вузликів. У дванадцятипалій кишці курчат-бройлерів були меншими висота ворсинок і глибина крипт, а в порожній кишці – глибина крипт. Встановлені морфологічні особливості вола, стравоходу, шлунку і тонкого відділу кишечника за використання мінеральної кормової добавки зі стулок мідій свідчать про розвиток захисно-приспосувальних реакцій стравоходу і шлунку до травмуючої дії грубих частинок стулок мідій, а тонкого відділу кишечника – до збільшеного вмісту кальцію в хімусі. За експериментально обґрунтованого згодовування мінеральних кормових добавок зі стулок мідій курчатам-бройлерам доведено необхідність ретельного їх подрібнення і дозованого використання в годівлі птиці

Ключові слова: сільськогосподарська птиця; мінеральна кормова добавка; морські гідробіонти; стравохід; шлунок; кишечник; морфометричні показники