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Morphological changes in the immune system organs of piglets fed with different feed additives

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Nowadays, the interest in studying the impact of feed additives on the organism of piglets during critical periods of weaning remains increasingly relevant. Feed additives containing blends of essential oils with plant extracts, preparations containing dry yeast, organic acids, pro- and prebiotics are suggested to replace growth promoters and antibiotics. This contributes to the increased productivity, survivability, immune response formation, and ensuring normal intestine morphology in piglets. Proper concentrations and timing of feed additive application aimed at maintaining a healthy intestine in piglets after weaning and improving their productivity remain equally important. This study presents the results of an experiment on the use of various feed additives for piglets before weaning and their effect on the morphology of the immune system organs. The research was conducted on piglets of the Large White breed, divided into four groups of ten individuals each. From days 7 to 28 of life, various feed additives were introduced into the main diet for each group: piglets of the control group (I) received a standard nutritionally balanced diet; piglets of group II had a supplement added to the standard diet containing a combination of live yeast of the *Saccharomyces* genus and a complex of 6 enzymes (proteases, cellulases, xylanases, α -amylases, β -glucanases, and phytases) at a rate of 0.5 kg per ton of feed; piglets of group III received a feed additive containing a blend of essential oils with plant extracts (*Thymus vulgaris*, *Rosmarinus officinalis*, *Origanum vulgare*, *Capsium* extract) at a rate of 0.2 kg per ton of feed; piglets of group IV had a feed additive added to the main diet containing dry yeast and egg powder enriched with immunoglobulins at a rate of 2 kg per ton of feed. The main feed used in the experiment was provided in the form of fine granules. Histological examination of the thymus, mesenteric lymph nodes, spleen, and duodenum lymphoid formations revealed that the morphological structure of the organs was preserved in piglets of all experimental groups. The partial structure of the thymus was well defined, with a clear differentiation into cortical and medullary substances. Piglets receiving the feed additive "Activo" with a blend of essential oils from plant extracts and the feed additive "Globigen Jump Start," which includes dry yeast and egg powder enriched with immunoglobulins, demonstrated a slight increase in the thymic cortex relative area, both on days 14 and 28 of the study, and an increase in the number of plasma cells. In the spleen and lymph nodes of piglets in all experimental groups, the lymphoid tissue was predominantly represented by small and medium lymphocytes; lymphoid nodules of large size with reactive centers were observed too. Brachet staining in the lymph nodes of piglets in all experimental groups revealed plasma cells with bright eosinophilia, and large nuclei predominantly with a single nucleus. However, their quantity varied, with the highest number of plasma cells observed in piglets of group IV, which received the feed additive "Globigen Jump Start" with dry yeast and egg powder enriched with immunoglobulins. Histologically, in the piglets' duodenum, the villi were closely packed, a small number of goblet cells were located between enterocytes, plasma cells with distinct eosinophilic cytoplasm and eccentrically placed nuclei were localized in the lymphoid nodules of the mucous membrane and in the crypt areas. Their quantity was higher in piglets of groups III and IV.

Keywords: piglets; feed additives; yeast; plant extracts; egg powder; thymus; spleen; lymph nodes; lymphoid formations; plasma cells.

Introduction

The weaning period for piglets is considered to be the most stressful and typically involves a decline in feed consumption due to the sudden cessation of suckling from the sow and adaptation to plant-based dry feeds containing complex proteins and carbohydrates, including various anti-nutritional factors. This leads to changes in the gastrointestinal tract microflora and the mucous membrane morphology (Hampson, 1986; Pluske et al., 1997; Brooks et al., 2001; Williams, 2003). It has long been established by scientists that the separation of piglets from the sow and the transition to a different feeding scheme result in significant changes in the intestine, including temporary villous atrophy, crypt hyperplasia, cellular barrier weakening, and the development of local inflammatory reactions (Pluske et al., 1997; White et al., 2002; Zhang & Xu, 2003). The diarrhea, frequently observed in piglets after weaning, is commonly associated with

the colonization of the gastrointestinal tract by b-hemolytic enterotoxigenic serotypes of *E. coli*, which proliferate in the small intestine after weaning. In such cases, antimicrobial agents are often employed (Osek, 1999; Fairbrother et al., 2005; Nagy & Fekete, 2005; Schierack et al., 2006). Additionally, research conducted by Snoeck et al. (2004) indicates a decrease in gastric motility in weaned piglets and a reduction in the stomach emptying rate, especially on the 3rd and 14th day of the post-weaning period, compared to suckling piglets.

Feed antibiotics were commonly used for many years to control gastrointestinal disorders in piglets during the weaning period, leading to the development of antibiotic-resistant bacterial strains (Amezcuca et al., 2002). It was precisely due to the emergence of antibiotic resistance that the European Union implemented a complete ban on the use of antibiotics in the feed of productive animals. In the initial law enforcing stages, this led to serious consequences in the industrial pig farming sector, specifical-

ly an increase in the weaning age and a reduction in the number of piglets weaned per year (Hayes et al., 2002).

Presently, the use of feed antibiotics in the nutrition of productive animals is also prohibited in Ukraine. Therefore, the development of alternative feed additives that can enhance piglet productivity, strengthen immunity, and positively influence the intestine microbiota has become a promising direction in animal nutrition. Yeast products have been used as feed additives for piglets for several years. Studies conducted by researchers (Jiang et al., 2000; Shen et al., 2009) indicate their positive impact on stimulating digestion and achieving growth, specifically mannanoligosaccharides and β -glucans, which are part of yeast cell walls. Cho et al. (2006) reported that yeast extracts rich in nucleotides had a positive impact on the intestinal mucosa and its lymphoid formations, contributing to the development of local immunity. In recent years, the use of phytogetic feed additives, which contain a wide range of herbs, spices, and essential oils, has become relevant (Zazharskyi et al., 2019; Boyko et al., 2021; Lieshchova & Brygadyrenko, 2022, 2023; Bilan et al., 2023). Scientists believe the possible mechanisms of action of plant extracts on animals are related to the intestinal microflora correction, increased absorption and nutrient assimilation, as well as pronounced antioxidant and immunomodulatory effects. The rapid metabolism and short half-life of biologically active compounds suggest a minimal risk of accumulation and cross-resistance in tissues. Essential oils are volatile oily liquids that exhibit strong antimicrobial action, selectivity for gram-negative bacteria, and are successfully used as an alternative to antibiotics for weaned piglets (Cho et al., 2006). Therefore, the overall impact of essential oils on the intestine morphology depends on the balance between tissue irritation and their beneficial effects on the intestine microbiota.

The immune system of the gastrointestinal tract is often referred to as the intestine-associated lymphoid tissue, which contains the largest amount of lymphoid tissue and plays a crucial role in the immune defense of pigs. According to some authors, adding essential oils as feed supplements improved the immune status of piglets after weaning. This is evidenced by an increase in the lymphocytes proliferation rate, the phagocytosis rate, as well as levels of IgG, IgA, IgM, C3, and C4 in the blood serum (Zhang et al., 2012; Luo et al., 2022). Walter & Bilkei (2004) reported that pigs receiving feed with the addition of 3 g per kilogram of oregano (60 g carvacrol and 55 g thymol per kilogram) had higher CD4:CD8 ratio and T/B cell ratio in peripheral lymphoid organs compared to pigs fed with conventional diets.

Until today, industrial pig farms use probiotics, i.e. products containing specific microorganisms that alter the small intestine microbiota via its implantation or colonization, positively affecting the overall health of piglets. The results of experiments evaluating the effectiveness of probiotics as an alternative to antibiotics vary significantly. For example, the use of lactic acid bacteria has shown a positive impact on pig performance indices (Lessard & Brisson, 1987; Shu et al., 2001; Taras et al., 2006). The addition of yeast cultures to diets for weaned piglets has also shown positive results in terms of maintaining the physiological condition of the intestine and productivity indices (Mathew et al., 1998; Bontempo et al., 2006), although other authors did not observe such effects (Komegay et al., 1995; Van Heugten et al., 2003). The inconsistency in many experimental studies and their results can be explained by the different dosages and types of probiotic strains, various zootechnical parameters of pig housing, frequent changes in diets, and feed ingredients (Bontempo et al., 2006). Therefore, future research on the effectiveness of probiotics should focus on identifying their components (single or multiple components), doses, and the timing of their application, which should remain constant.

Thus, considering all the aforementioned, we can assert an alternative to antibiotics exists. It is the use of probiotics, prebiotics, organic acids, yeast, and preparations based on essential oils. These alternatives are capable of improving the structure and function of the pig's gastrointestinal tract, strengthening the immune system, and promoting post-weaning growth. An important question that still remains to be answered is determining the concentrations and duration for which these preparations should be used to maintain normal gastrointestinal function and enhance pig productivity after weaning.

The aim of our study was to investigate, in a comparative aspect, the influence of different feed additives on the histological structure of the immune system organs of piglets during the pre-weaning period.

Materials and methods

All experimental studies were conducted in compliance with the international principles of the European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes (Strasbourg, 1986; Kyiv, 2002). The research protocol was approved by the Bioethical Commission of the Institute of Animal Biology at National Academy of Agrarian Sciences of Ukraine (protocol No. 93-01 dated June 3, 2021).

The experiments were carried out on piglets of the Large White breed. To study the influence of feed additives on the immune system organs of piglets, four groups of animals were formed, with ten individuals in each group. From days 7 to 28 of life, the following feed additives were added to the basic diet: 1) piglets in the control group (Group I) received a standard balanced diet throughout the research. 2) piglets in Group II received a standard diet with the addition of the "EnzActive mix" preparation: a combination of live *Saccharomyces* yeast and a complex of 6 enzymes (proteases, cellulases, xylanases, α -amylases, β -glucanases, and phytases) at a dose of 0.5 kg per ton of feed. 3) piglets in Group III received the feed additive "Activo", which contains a mixture of essential oils from plant extracts (*Thymus vulgaris*, *Rosmarinus officinalis*, *Origanum vulgare*, *Capsicum extract*). The amount of the feed additive was 0.2 kg per ton. 4) piglets in Group IV received the feed additive "Globigen Jump Start", which includes dry yeast and egg powder enriched with immunoglobulins at a dose of 2 kg per ton.

The main feed used in the experiment was provided in the form of finely granulated pellets to meet the nutritional needs of piglets. Throughout the experiment, the piglets were kept under identical conditions with free access to feed and water. Weaning was performed at 28 days of age.

On days 14 and 28, three piglets from each group were euthanized, and samples were collected for histological examination. Fragments of the thymus, bronchial lymph nodes, spleen, and duodenum were fixed in a 10% neutral aqueous solution of formalin and Bouin's fluid, dehydrated in a series of ascending alcohols, and then enclosed in paraffin blocks after I and II chloroforms. From the obtained blocks, 7 μ m thick sections were prepared using a microtome MS-2, mounted on glass slides, and stained using the Brachet method after drying. On histological preparations, the area of the cortical and medullary substance of the thymus was measured using Aperio Image Scope software. The prepared histological samples were processed and photographed using a Leica DM-2500 microscope (Switzerland) with a Leica DFC450C camera and Leica Application Suite Version 4.4 software.

Results

Microscopic examination of the thymus of experimental and control group of piglets revealed its typical morphological structure. Polygonal particles were separated by narrow connective tissue; cortical and medullary substances were clearly differentiated. Thymocytes of different sizes were densely located in the cortical substance, and Hassall's corpuscles of large size and rounded shape were located singly in the medulla (Fig. 1). Morphometric studies showed the cortical substance occupied a larger area of the thymus, accounting for 63.5% in the control group of piglets on day 14, while the medullary substance accounted for 30.2%. Similar correlations were observed in piglets of experimental groups II, III, and IV, where the cortical layer area was 63.7%, 63.9%, and 64.1%, respectively, and the medullary substance was 30%, 32%, and 31%. On day 28 of the study, no significant changes in the organ's morphological structure were observed. A slight thickening of the thymus capsule was found in the control group of piglets, accompanied by an increase in the number of Hassall's corpuscles and macrophages against the background of an enlarged area of the medullary substance (Fig. 2). In piglets of the control group, the cortical substance area was 65%, and the medullary substance was 35%. In contrast, piglets of experimental groups II, III, and IV showed a slight cortical layer expansion, occupying an area of 68%, 69%, and 71%, respectively, with the medullary layer accounting for 32.0%, 30.3%, and 30.2% (Table 1). Clear differentiation of organ structures was maintained. On day 28 of the study, a correlation was noted in the ratio of cortical substance to medullary substance, indicating a decrease in the cortical sub-

stance in the control group. The relative area of the thymus' cortical substance in the control group was 59.7%, the medullary substance was 32.5%, and the ratio of cortical to medullary substance was 1:1.83 on day 28, compared to 1:2.11 on day 14. The area of the connective tissue stroma increased, accounting for 6.3% on day 14 and 7.8% on day 28.

In contrast, in piglets of groups III and IV, which received the supplement "Activo" with a mixture of essential oils and plant extracts and "Globigen Jump Start" with dried yeast and egg powder enriched with immunoglobulins, the ratio of cortical to medullary substance was 1:2.10 and 1:2.12 on day 14 and 1:1.99 and 1:2.13 on day 28, respectively.

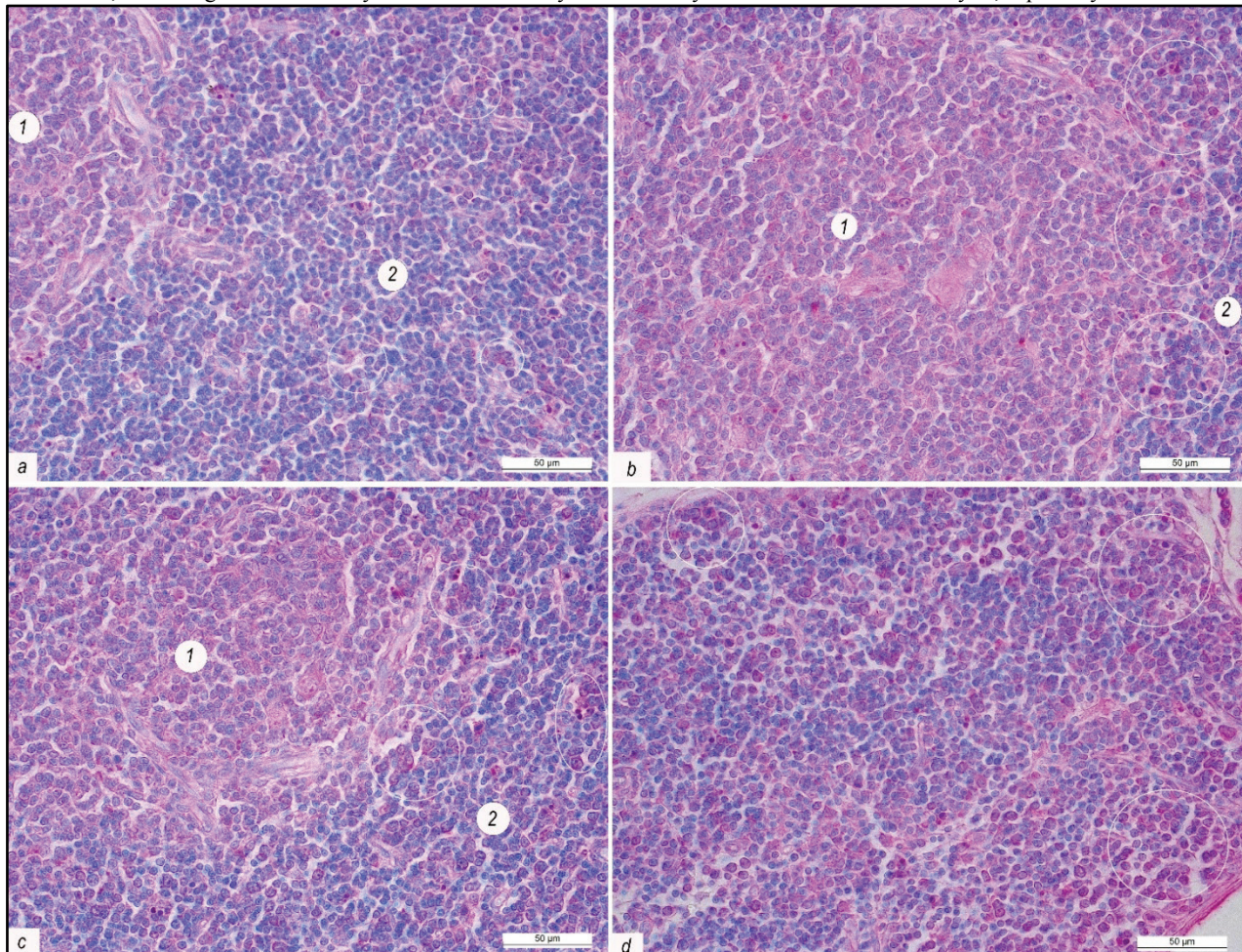


Fig. 1. Thymus of piglets on the 28th day of the experiment: *a* – control group, *b* – II group, *c* – III group, *d* – IV group; 1 – medulla, 2 – cortical substance, plasma cells with bright pyroninophilic cytoplasm (marked in a circle); Brachet staining

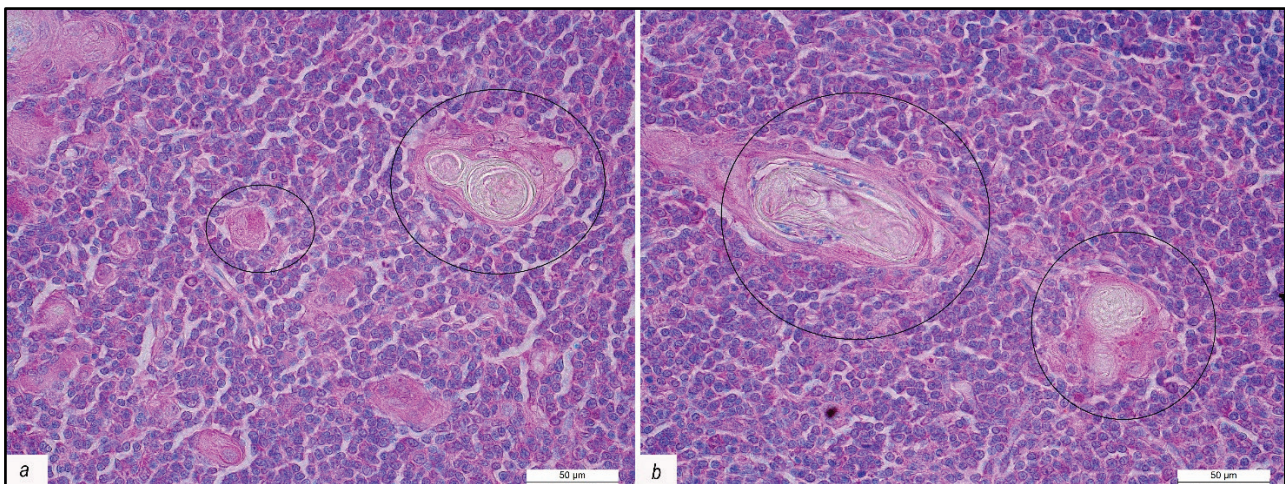


Fig. 2. Thymus of piglets on the 28th day of the experiment: *a* – control group, *b* – II group; an increase in the number of thymic Hassall's corpuscles; Brachet staining

Thus, based on the morphometric data, it can be concluded that the use of the feed supplements "Activo" and "Globigen Jump Start" for piglets throughout the research stimulated the functional activity of the organ both on day 14 and day 28, compared to other groups. This was indicated by an increase in the relative area of the thymus cortical substance and an increase in number of plasma cells. The quantity and sizes of thymic

Hassall's corpuscles are also indicators of the organ's functional activity. It is known that an increase in their quantity and absolute size may indicate previous stress on the thymus and gradual degradation of the particles' epithelioreticular stroma, which directly regulate the antigen-independent differentiation, proliferation of T-lymphocytes, and their positive and negative selection.

Table 1Relative area of tissue components of the thymus thoracic lobe after the use of different feed additives ($\bar{x} \pm SD$, $n = 5$)

Day	Indicators	Control group	Group II	Group III	Group IV
14 th day of experiment	Lymphoid tissue, %	93.66 ± 3.72	93.72 ± 2.34	94.17 ± 3.08	94.26 ± 4.12
	Cortical substance, %	63.51 ± 4.11	63.72 ± 3.97	63.86 ± 3.27	64.05 ± 3.22
	Medullary substance, %	30.15 ± 3.23	30.00 ± 3.12	30.31 ± 2.98	30.21 ± 2.24
	Ratio of cortical to medullary substance	1 : 2.11	1 : 2.12	1 : 2.10	1 : 2.12
	Connective tissue stroma, %	6.34 ± 1.17	6.28 ± 1.21	5.83 ± 1.32	5.74 ± 1.12
28 th day of experiment	Lymphoid tissue, %	92.21 ± 3.07	93.11 ± 3.04	93.48 ± 3.23	94.13 ± 3.16
	Cortical substance, %	59.74 ± 2.67	60.94 ± 2.84	62.23 ± 2.72	64.12 ± 3.42
	Medullary substance, %	32.47 ± 3.11	32.17 ± 2.35	31.25 ± 2.43	30.01 ± 2.28
	Ratio of cortical to medullary substance	1 : 1.83	1 : 1.89	1 : 1.99	1 : 2.13
	Connective tissue stroma, %	7.79 ± 2.13	6.89 ± 1.73	6.52 ± 1.37	5.87 ± 1.12

Histological examination identified the typical morphological structure of the bronchial lymph nodes in piglets. Similar to the thymus, lymph nodes exhibit clear differentiation into cortical and medullary zones, as well as capsules and trabeculae, which branch and sometimes anastomose in the central part of the organ. Broad marginal sinuses are located beneath the capsule, dividing primary lymphoid nodules, medullary cords, and medullary substance. The space between sinuses is filled with a dense reticular tissue network, within the loops of which lymphocytes are located, forming the organ's parenchyma. The parenchyma of lymph nodes is topographically divided into two zones: the cortical layer, located beneath the capsule and consisting of lymphoid nodules, and the medullary layer, composed of medullary cords of lymphoid tissue. In the cortical layer, round and oval distinct nodules separated by trabeculae and intermediate sinuses are clearly visualized. Secondary nodules with germinal centers are

also well-defined, surrounded by a marginal zone densely populated with lymphocytes. B-lymphocytes available in the primary lymphoid nodules of the lymph node cortical layer quickly proliferate and differentiate into plasma cells which are producers of immunoglobulins. Using the Brachet staining, plasma cells with bright eosinophilic staining, large nuclei, and predominantly one nucleolus were identified. However, the quantity of these cells varied slightly among different experimental groups of piglets (Fig. 3). The highest concentration of plasma cells was observed in the primary lymphoid nodules, diffusely in the cortical layer of the lymph node in piglets of the group IV, which received the feed supplement "Globigen Jump Start" with dried yeast and egg powder enriched with immunoglobulins. It is known that plasma cells are essential participants in humoral immunity, developing from antigen-activated B-lymphocytes.

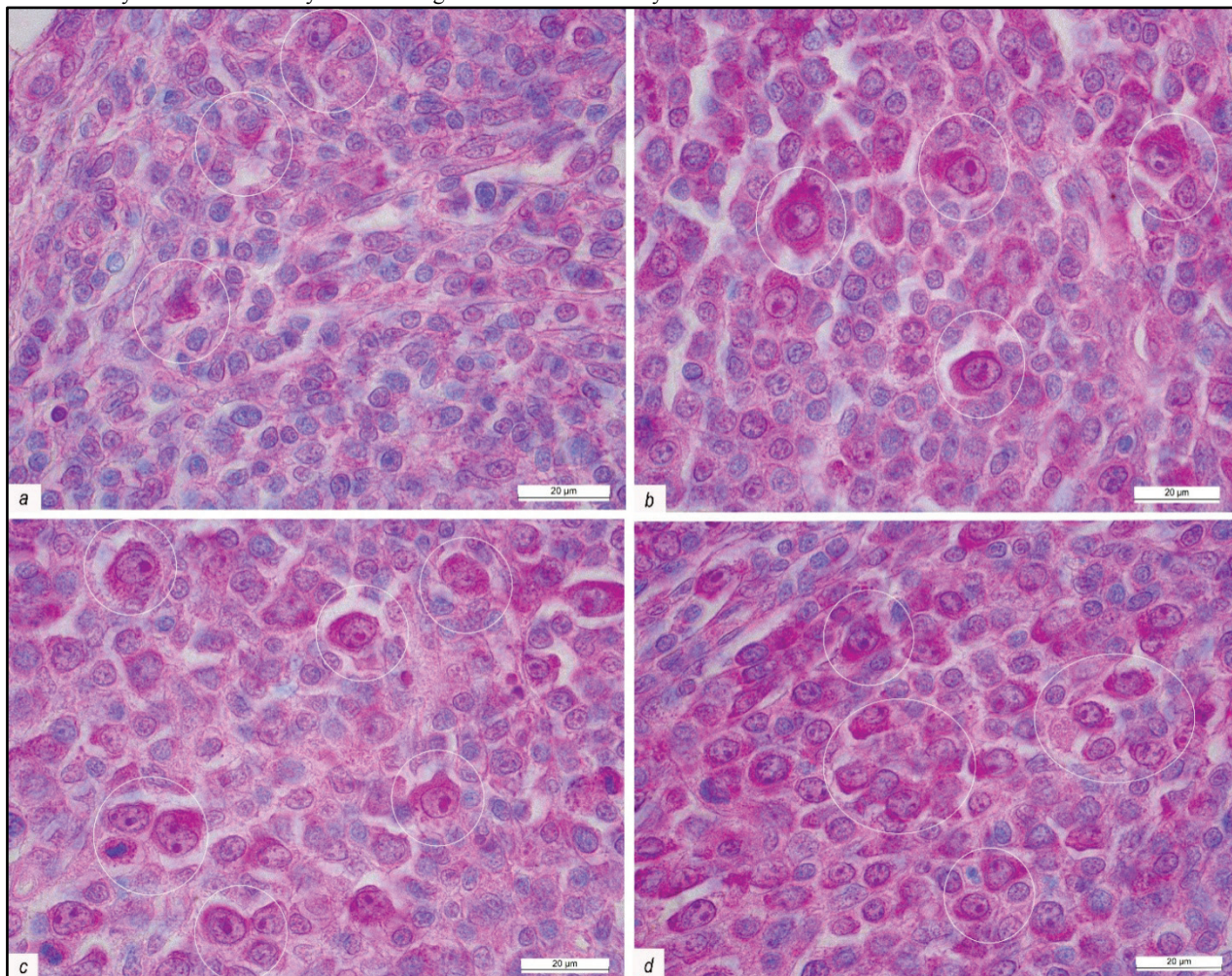


Fig. 3. Piglet lymph node on day 28 of the experiment: *a* – control group, *b* – Group II, *c* – Group III, *d* – Group IV; an increase in the number of plasma cells in the paracortical zone of the lymph node in piglets of Groups III and IV; Brachet staining

Histological examination of the spleen in piglets revealed its typical morphological structure. Microscopically, a clear division into red and

white pulp was noted, with the formation of round lymphoid nodules, predominantly small to medium in size, localized around the arteries. It is

worth noting that lymphoid nodules with germinal centers were more frequently observed in piglets that were fed with the supplements "Activo" and "Globigen Jump Start." The lymphoid nodules themselves were mainly composed of small and medium-sized lymphocytes, lymphoblasts, and plasma cells. The cell population density was noticeably higher in piglets of Groups III and IV compared to the control group, especially

on day 28 of the study. A slight increase in the number of plasma cells localized in the subcapsular zone was also observed. Additionally, the localization of plasma cells was found near trabeculae and small vessels. The concentration of plasma cells in different zones of the spleen indicates a certain increase in the organ reactivity, particularly evident in piglets of Groups III and IV (Fig. 4).

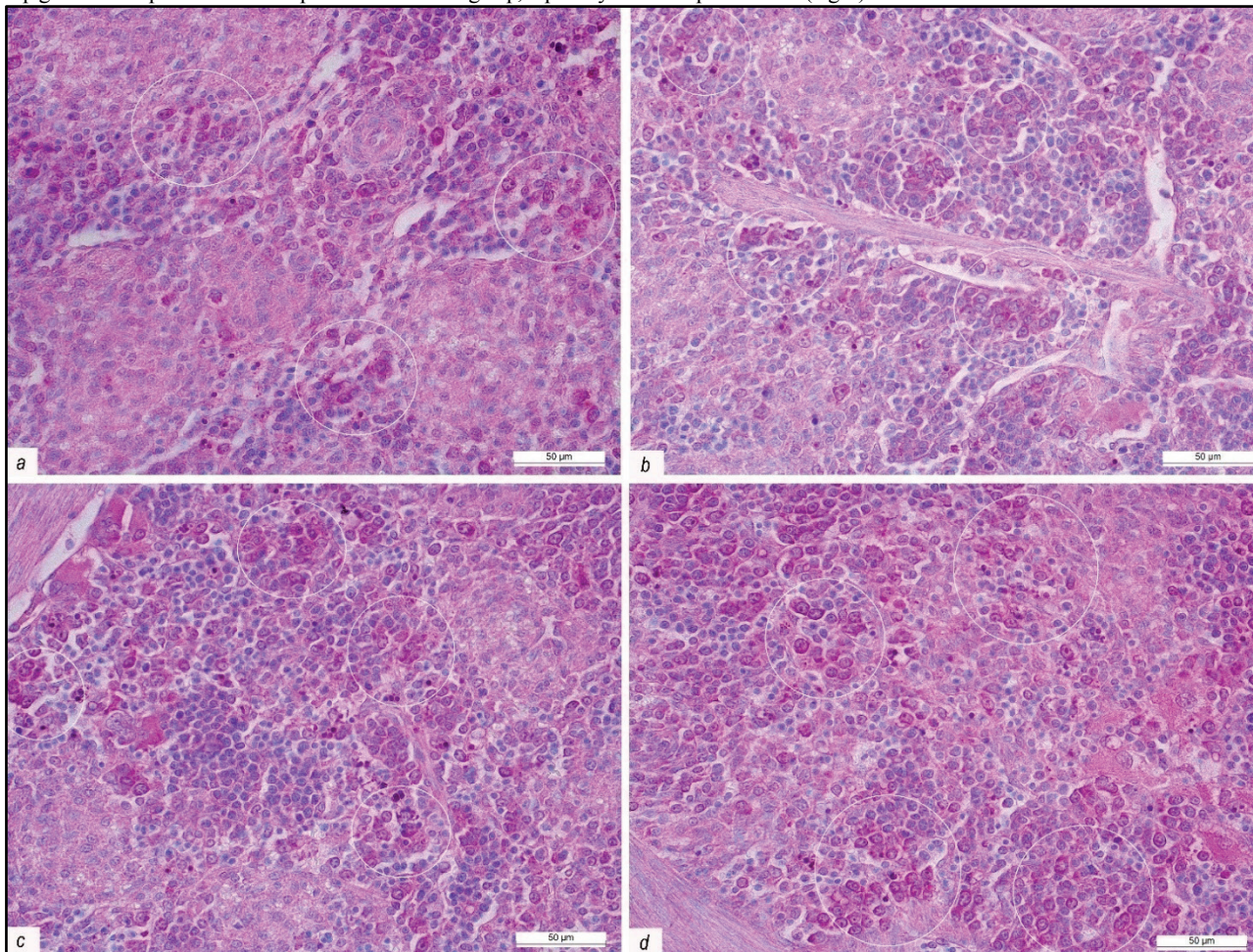


Fig. 4. Spleen: *a* – control group, *b* – Group II, *c* – Group III, *d* – Group IV; localization of eosinophilic plasma cells in the subcapsular zone near trabeculae and vessels; Brachet staining

Histological examination of the duodenum mucous membrane in piglets from all experimental groups revealed typical finger-like projections of the mucous membrane called villi, which tightly adhered to each other. A small number of goblet cells were observed between enterocytes. On day 28 of the research, a slight infiltration of lymphocytes into the lamina propria of the mucous membrane was detected in all piglets. The intestinal mucosa is known to be one of the largest immune organs in piglets, housing all types of immunocompetent cells. The intestinal immune system is divided into the inductive (Peyer's patches, bronchial lymph nodes) and effector (lamina propria and mucosal epithelium) parts. The mucosal epithelium plays a crucial immunological role in transporting immunoglobulin A. In all piglets from the experimental groups on day 28 of the study, we found a relatively high concentration of plasma cells, which were diffusely localized, mainly in the lymphoid nodules of the lamina propria and in the crypt areas of the duodenum (Fig. 5). Piglets from Groups III and IV showed an increase in the plasma cells number with pronounced eosinophilic cytoplasm and eccentrically placed nuclei.

Discussion

At birth, the immune system of piglets is immature and relies on passive maternal protection until weaning. The immune system of the mucous membrane in piglets develops during the first few weeks but does not reach maturity at the time of weaning. During weaning, piglets are exposed to a wide range of microbial agents through feed, frequently leading to

various illnesses and sometimes resulting in piglet mortality. It is during these critical periods that antibiotics have been widely used. However, numerous studies on piglets and poultry have shown the effectiveness of using probiotic feed additives and feeds containing essential oils. The additives have a positive impact on digestion stimulation, leading to better nutrient absorption and increased average daily weight gains (Franz et al., 2010; Windisch et al., 2010). The authors noted improvements in growth rates, feed consumption, and feed conversion ratios. The varied results reported by other researchers in their studies may be attributed to differences in digestive physiology, the origin of the feed, the quantity of the added mixture, and environmental conditions during the trials.

It is often claimed that essential oils enhance the taste of feed, thereby stimulating its consumption. This ultimately leads to a better feed absorption and increased piglet weight. However, in an experiment using a pre-mix with fennel and caraway oil as flavor additives or appetite stimulants, it was found that pigs took several days to adapt to the specific feed taste, and there was no subsequent improvement in consumption (Schöne et al., 2006). To date, experimental research is being conducted in this direction to confirm the assumption that herbs, spices, and their extracts improve feed consumption by pigs. The addition of essential oils to the diet of sows, especially during the lactation period, is attracting growing interest. Miller et al. (2009) reported that adding 2 g per kilogram of an essential oils mixture for 10 days before the expected farrowing date improved feed consumption by sows, reduced weight loss in sows during the first week of lactation, and increased piglet body mass at weaning.

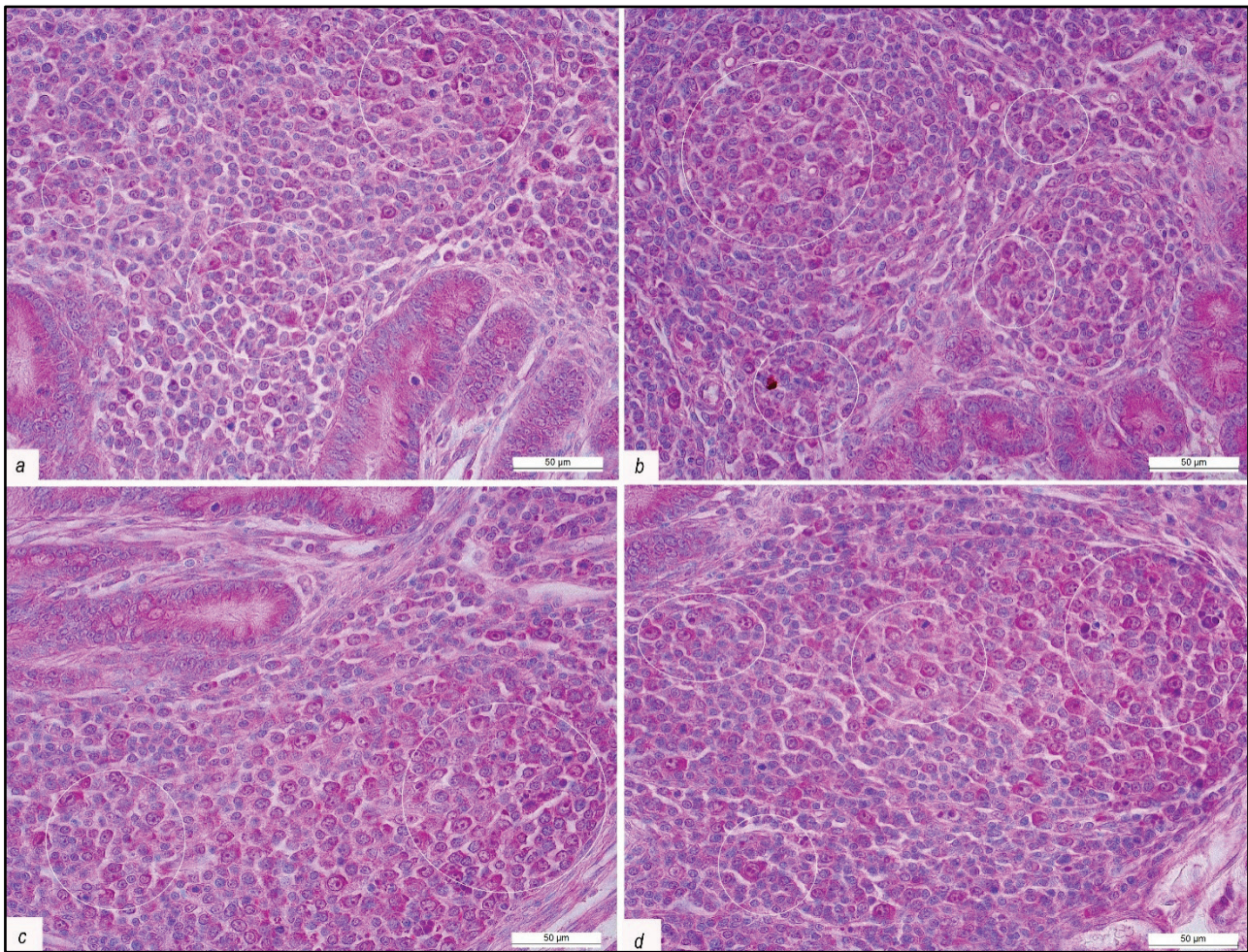


Fig. 5. Duodenum of piglets: *a* – control group, *b* – II group, *c* – III group, *d* – IV – group; localization of pyroninophilic plasma cells in lymphoid nodules of the submucosal layer; Brachet staining

Allan & Bilkei (2005) reported that sows fed a diet containing 1 g per kilogram of oregano, consumed feed more efficiently and had a lower annual piglet mortality rate. Similar results were obtained by other authors who added supplements with essential oils to the basic feed. It is well known that essential oils and aromatic plants exhibit antibacterial, antifungal, and antiviral effects in *in vitro* experiments (Windisch et al., 2008). It is also widely recognized that essential oils are more active against gram-positive bacteria than gram-negative ones (Burt, 2004; Muhl & Liebert, 2007; Tiihonen et al., 2010; Baker et al., 2012). It has been documented that essential oils improve nutrient digestibility in pigs (Janz et al., 2007; Li et al., 2012; Ahmed et al., 2013). This is partly explained by increased saliva secretion, bile production, and enhanced enzyme activity (Lambert et al., 2001; Rodriguez-Garcia et al., 2016).

However, Muhl & Liebert (2007) did not observe an improvement in nutrient digestibility and an enhancement of pancreatic and duodenal trypsin and amylase activity in weaned piglets fed diets containing carvacrol, thymol, and tannins as key components of the feed additive. Ambiguous results in nutrient absorption may be attributed to endogenous loss due to stimulated mucin secretion induced by plant extracts. Reduction in the quantity of pathogenic bacteria in the intestine enhances the regenerative capacity of epithelial cells in the villi, thus increasing the absorptive capability of the intestine. However, scientific literature lacks consensus regarding the effectiveness of using essential oils as feed additives that positively impact intestinal morphology. Reports show varying effects on villi length and crypt depth in the small and large intestine of broiler chickens and piglets fed supplements enriched with essential oils, including instances of increase, no change, or decrease in these parameters (Manzanilla et al., 2004; Walter & Bilkei, 2004; Zeng et al., 2015). Considering the diverse reactions in intestinal morphology, Windisch et al. (2009) hypothesized that one aspect of the phytotherapeutic action of essential oils is the irritation of intestinal tissues leading to a reduction in the intestinal

surface. Conversely, a favorable impact on intestinal health (i.e., reduction in pathogen pressure) may contribute to an increase in villi length and intestinal surface. Therefore, the overall influence of essential oils on intestinal morphology likely depends on the balance between tissue irritation and the favorable impact on intestinal hygiene. However, these mechanisms still need to be studied in detail.

Conclusion

The morphological characteristics of the immune system organs of pigs fed with various feed additives indicated their active morphofunctional state. It was observed that the partial structure of the thymus was well expressed in pigs from all experimental groups, maintaining the division into cortical and medullary zones. The thymocyte population density was high. However, in pigs treated with feed additives "Activo" and "Globigen Jump Start" throughout the research, the organ showed a more pronounced functional activity on both the 14th and 28th day compared to other groups. This was evidenced by the increase in the relative area of the thymic lobules' cortical substance and the increase in the number of plasma cells.

Well-formed lymph nodes, mainly of small and medium sizes, were observed in lymph nodes and spleen. Some lymph nodes had reactive centers. The concentration of plasma cells in different areas of the spleen and lymph nodes indicated a certain increase in the organ's reactivity, especially notable in pigs from groups III and IV.

The effectiveness and expediency of using feed additives were confirmed in all experimental groups of pigs. However, the most significant differences in indicators were observed in pigs fed with "Activo" supplement containing a mixture of essential oils and plant extracts, and the feed additive "Globigen Jump Start", which includes dry yeast and egg powder enriched with immunoglobulins during the weaning period.

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