

Morphofunctional changes assessment in newborn piglets in early postnatal ontogenesis complicated by isoimmunization symptoms

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Abstract. It is known that the pathological course of pregnancy is associated with a violation in the mutually dependent complex “mother-fetus” and leads to specific clinical consequences at the molecular-cellular, tissue, organ, organismic and population levels of the organization. Pathogenic action mechanism of the isoimmunization effect is very complex and still remains insufficiently deciphered. However, changes in the system of organs responsible for adaptation and survival play an important role in the pathogenesis of morphofunctional disorders associated with isoimmunization. Therefore, in farms where the fact of isoimmunization state is established, it is necessary to pay special attention to the creation of conditions that contribute to the normalization of the functional systems of adaptation of newborn animals. The material for research was the lungs, intestines, kidneys, and thymus. Only fresh material was examined. Samples up to 0.5 cm thick were taken for histological examination after forced slaughter. Fixation was performed with a neutral aqueous 10% formalin solution. Histological sections 4-6 microns thick were made from the obtained paraffin blocks, which were stained with hematoxylin and eosin for review purposes. When describing the morphofunctional state of organs, the structure, cellular composition, and condition of blood vessels were observed. Isoimmunization of sows during pregnancy is manifested by a characteristic clinical and morphological complex that differs from other infectious and non-infectious diseases. The most characteristic pathomorphological signs are: hypoplasia, delayed differentiation, dystrophic changes in the respiratory, digestive, urinary systems and organs of immunogenesis.

1 Introduction

Taking into account the high intensity of growth and development processes in newborns of multiple farm animals, as well as the fact that the fetus is fed through the placenta, intrauterine development and the fullness of the placental barrier are important [1-3].

The barrier function of the placenta is manifested only in physiological conditions. Under the influence of pathogenic factors, the barrier function of the placenta is disrupted,

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and it becomes permeable even to such substances that in normal physiological conditions pass through it in limited quantities [4-7].

The effect of previous isoimmunization may be associated with the formation of circulating antibodies [8, 9], which neutralize the antigen introduced during tolerance induction. As a result, a large dose of tolerogen or its repeated administration is required to obtain reactivity. This is confirmed by the results of a number of authors [10, 11], who showed that the introduction of specific antibodies early after the injection of tolerogen, when full tolerance has not yet occurred, prevented its development.

Considering the results of research conducted by various authors [12, 13], it can be concluded that preliminary isoimmunization (sensitization) in some cases creates certain difficulties in the induction of immunological reactivity. At the same time, a number of morphofunctional rearrangements of the homeostasis system occur in animals. In this connection, a comparative morphofunctional assessment of pathological changes in the organs of newborn offspring obtained from mothers exposed to isoimmunization during pregnancy becomes relevant in the theoretical and practical aspects of intensive animal husbandry.

The aim of research is to conduct a comparative morphofunctional assessment of pathological changes in the organs of newborn piglets obtained from sows exposed to isoimmunization during pregnancy.

2 Materials and methods

Experimental studies were conducted on sows of a large white breed on the farms in the Kurskoy district of the Stavropol territory. Histological studies were performed in the histological laboratory of the Scientific Diagnostic and Therapeutic Veterinary Center of the Stavropol State Agrarian University.

The test group included animals with detected signs of isoimmunization. The control group included animals that did not have an isoimmunization effect.

The morphofunctional state of the organs of newborn piglets obtained from sows exposed to isoimmunization during pregnancy was evaluated. The lungs, intestines, kidneys, and thymus served as materials for research. Исследовался только свежий материал. Samples up to 0.5 cm thick were taken for histological examination after forced slaughter. Fixation was performed with a neutral aqueous 10% formalin solution. From the obtained paraffin blocks, histological sections 4-6 microns thick were made, which, for the studying purpose, were stained with hematoxylin and eosin, according to the recommendations set out in the guide of V. V. Semchenko and co-authors in 2006 [14].

Section microscopy was performed using an Olympus BX45 light microscope with a built-in C 300 camera (Japan). Oculars $\times 10$ and lenses $\times 4$, $\times 10$, $\times 20$, $\times 40$, $\times 100$ were used for microscopy.

Morphometric studies were performed using the program "Videotestmaster Morphology 4.0" for Windows. The obtained digital data were analyzed using the statistical method of one-factor analysis of variance "Biostatistics 4.03" for Windows. Reliable differences were considered when $P \leq 0,05$.

3 Results

During isoimmunization, deep morphological changes develop in the organs and tissues of sows and piglets, indicating intoxication and circulatory disorders, violation of carbohydrate, fat, protein and other metabolism, which predispose to the development of pathological processes of a dystrophic and inflammatory nature in vital organs and are

accompanied by severe disorders of respiration, digestion, excretion, growth, development, etc.

In piglets subjected to isoimmunization, even in the absence of macroscopic deviations in the shape, color, and consistency of the lung, cystic-like extensions of the intra-lobular bronchi and emphysematous swellings of the entire lobule with ischemic vascular damage were histologically detected.

There was also a violation of the mucociliary function of the bronchial apparatus, expressed by a change in the ratio of mucoid and protein (serous) secretion, epithelium, increased serous secretion and weakening of mucosecretion. The number of goblet cells is reduced. There is edema of the alveolar tissue (Figure 2).

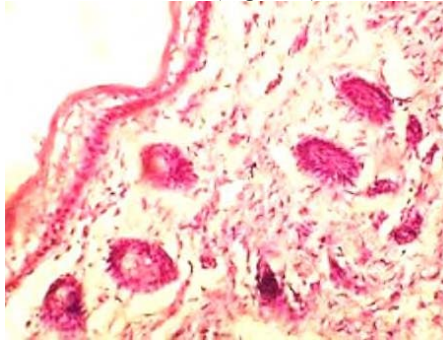


Fig. 1. Postpartum swelling of the connective tissue with ischemic damage to the blood vessels (staining with hematoxylin and eosin, $\times 400$).

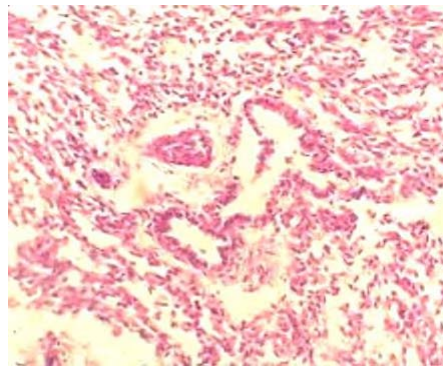


Fig. 2. Alveolar tissue edema (staining with hematoxylin and eosin, $\times 400$).

Changes in the alveolar parenchyma and mucociliary clearance are usually combined with inhibition of mitosis in the bronchiolar epithelium, weakening of phagocytic function, and underdevelopment of intramural and regional immunocompetent tissue.

There were differences in the morphofunctional structure of the piglets' kidneys during the newborn period born by sows, isoimmunized during gestation period. Histologically, certain changes in the glomeruli and tubules of the nephrons were detected in the cortical substance. The cortical substance is full-blooded, the vessels are filled, and in places there are hemorrhages in the glomerular lumen and interstitial tissue. There were underdeveloped, non-functioning nephrons, especially near the organ capsule. Smaller glomeruli are deformed, with shrunken cell elements. The tubules near these glomeruli have no lumen and are covered with juicy epithelium with granular cytoplasm (Figure 3).

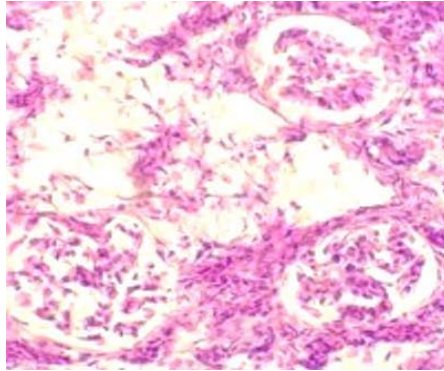


Fig. 3. Kidney tubules edema with necrotic lesion (staining with hematoxylin and eosin, $\times 400$).

In the endothelium of glomerular capillaries, pericytes, mesangial cells and epithelium lining the glomerulus capsule, fat, granular and carbohydrate dystrophy is noted. The epithelium of the proximal parts of the nephron in some areas is swollen, enlarged in volume, its cytoplasm is dim, almost completely covers the lumen of the tubule. The nuclei are large, light, with lumps of chromatin and a nucleolus. Some of the cells are located on the basement membrane, while others are rejected into the lumen of the tubule. The lumen of these tubules is filled with a fine-grained protein mass. The boundaries of the tubule cells are not defined, in some cells the cytoplasm is completely lysed, and only the nuclei and shells are visible, which seem to lie on the basement membrane.

Pathoanatomic and histomorphological changes in the gastrointestinal tract were most pronounced in the distal small intestine. In the stomach and large intestine, changes were not found in all piglets from the experimental group, and in the duodenum they were almost not manifested. In piglets subjected to isoimmunization, hyposecretion and dystrophic changes of prismatic cells by the type of coagulation necrosis, hypofunction of glandular cells were revealed. The greatest changes were observed in the small intestine, especially in the jejunum. They were characterized by dystrophic changes in the marginal epithelium of the apices and lower lateral surfaces of some villi, serous, and sometimes round-cell infiltration of their connective tissue stroma.

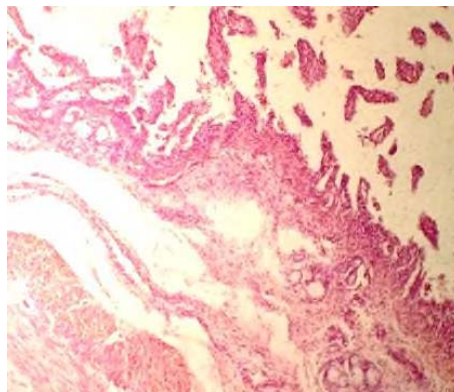


Fig. 4. Desquamation of the jejunal wall epithelium with signs of acute edema (staining with hematoxylin and eosin, $\times 400$).

In the jejunum (figure 4) of most piglets, increased mucosal folding was noted as a result of edema of the submucosal layer and spasmodic contractions of the intestinal muscles.

Violation of the histostructure of the villi in piglets with the effect of isoimmunization was characterized by the fact that they greatly shortened and expanded. The cylindrical epithelium is transformed into a cubic and then to a flat. In some piglets epithelium separated from the villi as a result of subepithelial edema. There were foci of necrosis and desquamation of the ciliated epithelium on the entire surface of the villi, sometimes necrosis of the apical part of the villi was noted.

In the large intestine, histomorphological changes were not detected in all piglets with the effect of isoimmunization, they had some differences both in the nature of the development of the pathological process and in the depth of the mucosal lesion.

Test group of piglets were characterized by similar morphological and functional changes in the thymus in the form of reducing its weight to 5 g and more, falling and destruction of the cortical substance of the collapse of thymocytes, erase layer structure (figure 5) and causing the medulla contains thymocytes as much or more than in the cortex (layers inversion). The thymus lobes are sharply enlarged in volume with an increase in the cortical substance (figure 6).

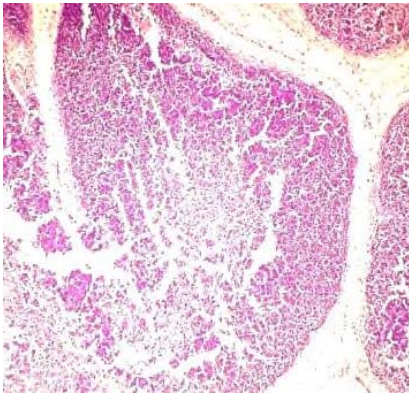


Fig. 5. Increase in the cortical substance of the thymus lobules (staining with hematoxylin and eosin, $\times 400$).

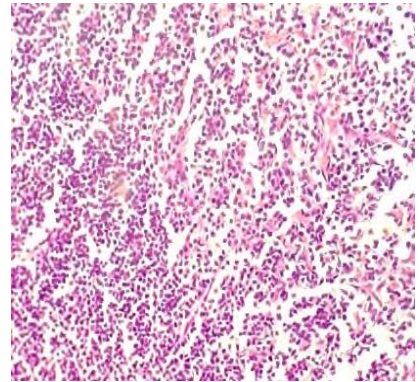


Fig. 6. Proliferative changes of lymphoid cells with vague boundaries of the cortical substance (staining with hematoxylin and eosin, $\times 400$).

The changes we found in the thymus of piglets mainly correspond to morphological reactions of functional (accidental) transformation and are a morphofunctional expression of overstrain, exhaustion, probably not compatible with life processes.

4 Conclusions

Our research has shown that in the test group of newborn piglets with isoimmunization effect, there are disorders of microcirculation, dystrophic and inflammatory processes in the kidneys. Histological studies indicate a high degree of damage to the atmosphere and membrane structures of the nephron. It is characterized by a decrease in the content and change in morphology in the protoplasm of the endothelium and tubular epithelium.

Analysis of the results of histomorphological studies of the gastrointestinal tract showed that the appearance of diarrhea is associated with dystrophic changes in the epithelial (suction) layer of the villi. The main changes were characterized by desquamation of the epithelium, atrophy and destruction of the villi of the mucous membrane of the small intestine. All this significantly reduces the suction surface and aggravates dystrophic changes in the epithelial layer of the intestinal mucosa.

At the time of birth, piglets' lungs are structurally fully formed. However, the elements responsible for the organ's resistance to adverse effects – the mucociliary system, immunocompetent elements, supramembrane and membrane structures-do not reach full differentiation, this occurs only by age of 3-4 months. The speed and degree of development and differentiation of the tissue elements of piglets' lungs largely depend on the usefulness of the immunobiological relationship in the functional system “mother-placenta-offspring”. From sows that have undergone isoimmunization during pregnancy, piglets are born with signs of pneumopathy and a predisposition to the disease of bronchitis and bronchopneumonia.

The changes we found in the thymus of piglets mainly correspond to morphological reactions of functional (accidental) transformation and are a morphofunctional expression of overstrain and exhaustion.

The mechanism of pathogenic action of the isoimmunization effect is very complex and still remains insufficiently deciphered. However, changes in the system of organs responsible for adaptation and survival play an important role in the pathogenesis of morphofunctional disorders associated with isoimmunization. Therefore, in farms where the fact of isoimmunization state is established, it is necessary to pay special attention to the creation of conditions that contribute to the normalization of the functional systems of adaptation of newborn animals.

Isoimmunization of sows during pregnancy is manifested by a characteristic clinical and morphological complex that differs from other infectious and non-infectious diseases. The most characteristic pathomorphological signs are: hypoplasia, delayed differentiation, dystrophic changes in the respiratory, digestive, urinary systems and organs of immunogenesis.

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