EVALUATION OF INTESTINAL BARRIER FUNCTION IN BROILER CHICKENS USING POLYMERASE CHAIN REACTION METHOD

Shatalov S.A.[1], Baselga R.[2], Benito A.[2], Arnal J. L.[2], Nedzvetsky V.S.[1]

1 - EXOPOL SL Polígono Rio, San Mateo de Gállego Zaragoza, España
2 - Dnipro State Agrarian and Economic University, Dnipro, Ukraine
shatalov.s.a@dsau.dp.ua

Relevance. In the current industrial conditions of broiler chicken farming, studying the intestinal barrier function is an extremely important task. Recently, the high intensification of poultry farming has caused several health issues in chicks. One critical aspect is the disruption of the intestinal barrier function, leading to reduced immunity and increased susceptibility to infectious diseases. Additionally, such disruptions negatively affect feed absorption efficiency, resulting in economic losses. Research in this area helps to evaluate the effectiveness of various drugs and bio-additives used to enhance the immunity and productivity of birds. Generally accepted indicators of intestinal health are morphological parameters and the species composition of the intestinal microbiota. However, considering recent scientific achievements, molecular markers of intercellular adhesion, cytokine production, and programmed cell death are increasingly used to assess intestinal barrier function. The use of molecular biological methods for evaluating the intestinal barrier function in broiler chickens is particularly relevant and appropriate. Determining the expression levels of tight junction proteins and inflammatory factors provides information about the health and functionality of the intestine.

The aim of this study was to develop criteria for evaluating the intestinal barrier function in broiler chickens by determining the gene expression of molecular markers using the RT-PCR method.

Materials and Methods. For PCR research, primers for the conserved regions of the molecular marker transcripts were selected using the GenBank genetic sequence database from NCBI (National Center for Biotechnology Information, USA) and the BLAST tool. Primers were chosen for occludin (reference sequence in NCBI NM_205128), E-cadherin (reference sequence in NCBI NM_001039258.3), and chicken β-actin, against which the expression intensity of the selected genes was compared.

Samples of organs (duodenum) were taken from broiler chickens of the ROSS 308 cross: on days 14, 20, 27, 34, and 43, five from each group. The control group of birds was raised according to the standard scheme adopted at the "Agro Oven" poultry farm. The experimental group of chicks additionally received a mixture of short-chain fatty acids with monoglycerides starting from the 14th day of life.

Nucleic acids were extracted from the studied samples using the automated PurePrep96 station with BioExtract® SuperBall® reagents manufactured by BioSellal, France. For reverse transcription PCR, One-step NZYSpeedy RT-qPCR Green kit reagents manufactured by Nzytech, Spain, were used. Amplification was performed on a Biorad CFX96 thermal cycler, which allows precise measurement of gene expression levels due to its high sensitivity and specificity.

The obtained data were analyzed and normalized using the 2^(-ΔΔCt) method with MS Excel. For further visualization of the results, the GraphPad Prism program was used to create illustrative graphs and perform statistical analysis.

Overall, the comprehensive approach to data collection, processing, and analysis, along with the use of modern molecular biology methods, ensured a high level of adequacy and reliability of the results obtained.

Results. The introduction of fatty acid preparations into the diet of broiler chickens significantly affects the expression of occludin and E-cadherin genes. In the experimental group, a stable increase in the levels of these proteins was observed, especially pronounced on days 20 and 34. The expression of occludin significantly increased in the experimental group throughout the

experiment, with the highest values on day 34. This indicates an improvement in the intestinal barrier function, which can reduce the risk of pathogen penetration and improve the overall health of the chicks. E-cadherin expression also noticeably increased in the experimental group, particularly on day 20. The increased levels of this protein indicate improved cellular adhesion and epithelial layer integrity, which helps protect against infections. The beneficial impact of fatty acids confirms their potential benefit for enhancing bird immunity and productivity.

Conclusion. The obtained results demonstrated the importance of using molecular biological methods, particularly RT-PCR, for assessing the intestinal barrier function in broiler chickens. The study results indicate a significant impact of fatty acid preparations on the expression of markers such as occludin and E-cadherin. Considering that intercellular adhesion proteins play a key role in maintaining the integrity and functionality of the intestinal barrier, the obtained data suggest the prospect of using the developed strategy for assessing intestinal health and, accordingly, productive animals. Thus, the results confirm the potential benefits of including fatty acids in the diet of broiler chickens to enhance their immunity and productivity. This comprehensive approach to evaluating intestinal barrier function and using modern molecular biology methods allows obtaining data on chick health and feeding correction.

INDICATORS OF PROTEIN METABOLISM OF BLOOD AND MODULATION OF THE BARRIER FUNCTION OF THE PIGLET'S INTESTINE BY THE ACTION OF AN ISOTONIC PROTEIN MIXTURE

Sheptukha O. [1]*, PhD student,*
Stefan G. Buzoianu [2]*, PhD, R&D Project Manager,*
Masiuk D. [1]*, D.Vet.Sc., Head of the Department of Animal Physiology, Biochemistry, and Laboratory Diagnostics,*

[1]*Dniprovskiy State Agrarian and Economic University, Dnipro, Ukraine*
[2]*Tonisity International, Dublin, Ireland*
shepto@icloud.com

Relevance. The main task in modern pig breeding is to obtain the maximum number of piglets per sow per year. On average, 2.2-2.3 farrowings per year can be obtained from a sow, but the number of piglets born to a sow varies from farm to farm. The number of piglets born per sow in Ukraine on average ranges from 13.5 to 17.5 heads, based on 12-16 working nipples per sow. Improving the viability of newborn piglets in the early postnatal period of ontogeny is a priority for any pork producer. The safety of piglets in the lactation period in Ukraine is on average 85-90% of the piglets born. The main losses of piglets occur in the first week of life associated with a lack of colostrum, milk, impaired intestinal barrier function and the manifestation of diarrhea due to a complex of pathogens of viral-bacterial etiology (Rota A, C, E. Coli, Cl. perfringens A, C). In the first week after birth, piglets face the most severe stress associated with zooveterinary treatments (removal of canines, docking of tails, tagging, castration, injections of iron-containing drugs, antibiotics, coccidiostats)

Therefore, new strategies to prevent the development of infections by initiating the protective mechanisms of innate immunity and intestinal barrier function are very relevant. The use of additives that can increase the resistance of the intestines to external damage is an important strategy in modern animal husbandry. The barrier function of the intestine is provided by proteins involved in the intercellular adhesion of the epithelial layers of the intestine. Weakening of the