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CLINICAL AND PARACLINICAL ASPECTS IN HAEMORRHAGICAL ENTERITIS OF YOUNG BIRDS

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ABSTRACT

PhD thesis entitled "CLINICAL AND PARACLINICAL ASPECTS IN HEMORRHAGIC ENTERITIS OF YOUNG BIRDS" had as motivation the narrow area of research on this disease with major impact on poultry reared in intensive and extensive system, reducing weight gain, carcass depreciation and last but not least the mortality. Therefore early diagnosis and appropriate action is a necessity aiming to reduce losses. The thesis has a deeply original character from the fact that until now, in Vrancea county, no study on comparative epidemiological between the intensive and extensive farming systems, allowing to shape an idea on differential etiology of this syndrome - hemorrhagic enteritis - was conducted before.

The thesis has 266 pages and it is structured in two parts.

The bibliography is composed of 54 pages, being systematized into five chapters.

presenting selected data from bibliographic sources in the literature on the etiology, pathogenesis, clinical manifestations and diagnosis of pathological changes in hemorrhagic enteritis in young birds.

Part Two, the *Personal contributions* is presented in a number of 169 pages and contains 9 chapters, the research and objectives on this study, organizational and institutional framework in which the research took place, materials and methods, results, related discussions and general conclusions that have emerged from research.

The data presented are supported by 2 tables and 19 figures inserted in the *Bibliographic study*, 54 tables and 107 figures inserted in the *Personal contributions* part. The bibliographic list sums 237 titles in domestic and international literature and internet files containing dedicated subjects.

In the thesis there can be found personal data published on scientific papers that were presented at the annual Symposium organized by Veterinary Medicine in Iasi city.

The importance of the theme comes from the fact that hemorrhagic enteritis has a negative impact on weight gain and health in young birds.

Starting from these considerations, our research aims to identify and study the main causes involved in the etiology of hemorrhagic enteritis to poultry. In order to achieve this goal there were pursued the following objectives:

- ✓ study on intensive and extensive pathogens involved in producing hemorrhagic enteritis of infectious (bacterial, viral) and parasitic nature to poultry in Vrancea county both in intensive systems and into the household environment;

- ✓ determination of the incidence in hemorrhagic enteritis to poultry according to their etiology;
- ✓ tracking clinical and paraclinical aspects in hemorrhagic enteritis of infectious (bacterial, viral) and parasitic nature to poultry;
- ✓ aspects of differential diagnosis in hemorrhagic enteritis to poultry.

To achieve the proposed study, the researches have been conducted in the county of Vrancea. Research on the county were made in three broiler farms with intensive system from unit A and in 12 localities (Răcoasa, Suraia, Milcov, Vulturu, Vânători, Mircești, Cotești, Dumbrăveni, Garoafa, Năruja, Golești, Vidra), where the poultry are reared in the traditional system. Laboratory investigations were conducted in the LSVSA from the Sanitary Veterinary and Food Safety Department of Vrancea county.

In chapter VIII it is presented a study on the prevalence, geographic distribution and clinical aspects of infection / infestation by protozoa, nematodes and cestodes but also bacterial and / or viral infections producing hemorrhagic enteritis to poultry reared in intensive and extensive systems. From the conducted research, resulted that the prevalence of parasitic diseases in poultry has been reduced in the intensive system, but in the extensive one, the species of parasites (protozoa and helminth) are widespread.

The only parasites involved in the production of hemorrhagic enteritis in poultry in the unit A, took for study, was eimeriosis, in proportion of 54.54%, compared with the extensive growth system, traditional where eimeriosis to chickens was found in a proportion of 42.65 % being accompanied by other parasites too, namely: Ascaridia galli infestation rate of 24.85%, with a rate of 9.03% cestodosis (Raillietina Hymenolepis spp and pens) and other parasites (Histomonas, Heterakis), a rate of 7.62% poliparasitism.

All cases of eimeriosis were diagnosed in the intensive system during winter season, between November to February contrary to extensive farming systems where the eimeriosis cases were found throughout the year, in contradiction to the specifications found in the literature.

In the intensive system bacterial infections showed a higher incidence compared to the extensively system. This is due to stress caused by congestion in the halls (approx. 15 000 chicken / hall) and permanent litter growth system that favors engraftment and conditional pathogenic germs. Thus the prevalence of intestinal bacterial infections in chickens, farmed extensively, in areas under study was conducted, showed a rate of 21,31% (15,22% colibacillosis, 4,06% clostridiosis and 2,03% salmonellosis) while for the intensive system was 51.5% (38% colibacillosis, 10% clostridiosis and 3.5% salmonellosis), resulting that most common intestinal bacteriosis encountered were colibacilosis, in both farming systems.

The incidence of viral infection with Newcastle disease virus in extensive farming systems during the study period was 3,04%, while for the intensive system were not found clinical cases of intestinal viral infections due to the bio-security and preventive immunizations conditions applied.

Relatively low mortality (3.12% -13%), from the intensive system compared with the extensive growth system (8% -92%), but also with that specified in the literature (10% -100%) is due to early diagnosis and rapid establishment of therapeutic medication.

In chapter IX there were conducted laboratory researches for paraclinical aspects on hemorrhagic enteritis on young poultry with infectious and parasitic etiology. The diversity of diseases that cause hemorrhagic enteritis in chickens requires a detailed study of their etiology. This can be achieved only by clinical test, indication of their etiology requiring laboratory studies, too.

Laboratory studies on broiler chickens, aged 1-6 weeks and intensively farmed, as well as on chickens of different ages reared in traditional household system, were conducted in the diagnostic profiles from the Vrancea LSVSA as follows: species identification of the parasites involved in the etiology of hemorrhagic enteritis in the parasitology profile, biochemical and hematological examination of blood in the hematological profile, histopathological examination in the morphopatological profile, bacterioscopic examination, bacteriological examination in the bacteriological in profile and virological examination (viral biology and serology) in the profile of virology / molecular biology.

Following the study, we have noted that the average values of biochemical indices: total protein, fat, cholesterol, alkaline phosphatase and hematological (leukocytes, erythrocytes, hemoglobin, hematocrytes, MCHC) were below the values cited in the literature, for clinical evolution of eimeriosis and in physiological limits for the development of cestodosis and ascaridiosis. In the intensive system, VEM recorded values over the physiological limits cited in the literature in year 2009 and 2010; in the extensively growth system have been reported values of VEM's beyond physiological limits; medium urea limits, registered during clinical episodes of eimeriosis, were well above those cited in the literature in both systems.

In the intensive system, the average of oocysts Eimeria spp copro-elimination was variable from one farm to another and from one year to another, and the maximum value of OPG's always coincided with clinical manifestations of the disease while in extensively system, the maximum value of oocysts Eimeria spp copro-elimination didn't always coincided with the onset of clinical disease. We mention the cases from 2007 in Suraia commune and from 2009 in Năruja. Also there was discovered that there is no fixed number of oocysts in which the disease appears and

no direct correlation between the number of oocysts counted and the disease evolution in any of the systems.

It was observed that age of onset of the disease varies from one system to another, in the intensive system the disease occurring at age 19 to 28 days with maximum frequency 20 to 23 days, unlike the extensive system where the disease occurred in specimens aged 4-12 weeks, with maximum frequency at 7 weeks. This is due to the fact that broilers reared in intensive system (unit A) are slaughtered at the age of 42-45 days and have a continuous chemopreventive, unlike chickens raised in the traditional system with a much larger economic life.

In the intensive system, we encounter a big frequency of E. tenella species, with a percentage of 72.61, followed by E. necatrix with a rate of 60.1, E. acervulina 44,62, E. Brunetti 30.9 and maximum 15.45; in extensive farming systems E. tenella is on the first place with a frequency of 77.5%, E. acervulina 42.5% and E. necatrix with 41.25%.

During investigations it was found the simultaneous evolution of eimeriosis with bacteriosis in the intensive and traditional system as follows: *Eimeria-Salmonella* - (0,25%), *Eimeria-Escherichia* 1,76% (intensive system - 1,26% and extensive system-0,5%), *Eimeria-Escherichia-Clostridium*-1,26% (intensive system - 0,76% and extensive system - 0,5%), *Eimeria-Salmonella-Clostridium* 0,25% şi *Eimeria-Salmonella-Escherichia* 0,25%.

Salmonella spp strains isolated for 5 years, belonged to serogroups DO, BO, CO, most frequently encountered species being Salmonella enteritidis.

The virus strain, isolated in the pseudo-pest hole was classified in the mesogenes category by IPIC's value (IPIC – 1,31) causing illness even among immunized birds on the holding.

Chapter X presents a detailed study of common and specific pathological changes found in diseases with parasitic and infectious etiology that cause hemorrhagic enteritis in chickens.

Common lesions seen both in parasitic and infection diseases, ranged from the duodenum, the jejunum and duodenum catarrhal typhlitis, jejunum and congestive catarrhal typhlitis and congestive-hemorrhagic depending of the form of the disease, the identified species but also on the origin and strain existing at the effective time. Fibrino-hemorrhagic lesions of typhlitis were found in a biger percentage (3-25%), especially in herds where it developed among eimeriosis / colibacillosis (Vulturu and Dumbrăveni) or eimeriosis / histomoniasis (in Dumbrăveni and Coteşti commune).

Specific lesions found in some infectious diseases with bacterial etiology and / or viral, diagnosed, such as: congestion and hemorrhages on serous and internal organs, especially in the liver and spleen in salmonellosis, hepatic congestion in clostridiosis, serofibrinous inflammation of serous and organs in colibacillosis, hemorrhagic necrotic proventriculitis and the presence of pseudo pesto buttons on the intestine in Newcastle disease, helping to establish the diagnosis.

Chapter XI presents an experimental eimeriosis episode on 3 groups of broilers aged 15 days. Lots consisting of 15 chickens, each received the same maintenance conditions but feeding conditions were different, as follows:

- *Group I* was fed with feed without eimeriostatic and subject to experimental infection by administering suspension of evolved Eimeria spp oocysts for 2 consecutive days. They received a treatment with curative Coccistop tablets, taken once the bloody diarrhea appeared;

- *Group II* was fed with feed with eimeriostatic (Kokcisan - 0.5 g / kg) and subjected to experimental infection by administering suspension of evolved Eimeria spp oocysts for 2 consecutive days. They did not benefit from curative treatment;

- *Group III* was fed with feed with eimeriostatic (Kokcisan - 0.5 g / kg) and subjected to experimental infection by administering suspension of evolved Eimeria spp oocysts for 2 consecutive days and they received curative treatment with Coccistop tablets, taken once the clinical signs appeared.

The evolved oocysts suspension that was used for the infection was composed by the most commonly found species of eimeria in unit A as follows: Eimeria tenella (68%) followed by Eimeria acervulina (24.5%) and Eimeria Brunetti (7.5%). Postinfectious, they were identified the same species, but the report was different: Eimeria tenella (77.6%) followed by Eimeria acervulina (21.5%) and Eimeria Brunetti (0.9%).

In terms of morphometric, the oocysts used to produce experimental infection were within the limits cited in the literature.

In group I, which has not received an eimeriostatic contribution, there were identified both forms of asexual propagation (schizonts with merozoites and free merozoites) as well as sexual (oocysts of E. tenella in caecums and E. acervulina in the duodenum) and dimensions of E. tenella schizonts like or slightly higher than those cited in the literature.

Clinical manifestations and dynamics of mortality differed in the three groups according to the time of usage and applying preventive and curative therapy. In group I which did not received any feed with eimeriostatic, the symptoms appeared after 6 days of infection, and in group II and III that received feed with eimeriostatic the symptoms appeared later, after 10 days and 9 days postinfection, which demonstrates the need for an eimeriostatic in the food, especially in intensive farming. If the group I, the curative treatment applied after the first cases of death, led to its decline (66.6%), in group III, there were not reported cases of mortality due to application of curative treatment at the same time the clinical signs appeared. Lot II, that has not benefited from the contribution of a curative treatment, had a mortality range that has been constant throughout the observation period (86.6%).

From the analysis of the identified constant values after the infection, we can see a decrease in total number of protein (hypoproteinemia), lipids (hypolipidemia), alkaline phosphatase, cholesterol and glucose, of the haematological constants (WBC, RBC, hemoglobin, hematocrits, MCV, HEM, CHEM) and an increase level of urea (uremia).

The oocysts of Eimeria spp copro-elimination showed higher values in group II, of 45. 780 OPG, with a maximum of 108.400 OPG age 30 days, while on group I the value was 24. 899 OPG, with a maximum of 148.350 OPG at the age of 22 days and in group III of 9.900 OPG with a maximum of 58. 000 OPG at the age of 24 days. We note that although the average of copro-elimination was higher on group II, fed with feed with eimeriostatic and did not received curative therapy, the copro-elimination peak was recorded on group I, fed with feed without eimeriostatic and were the curative therapy was applied 8 days after the infection.

The lesions observed at the necropsy examination were duodenitis and congestive and hemorrhagic catarrhal typhlitis (in the outbreak) for group I, fed with feed without eimeriostatic but benefited from the contribution of curative therapy, with a lower lesion score of 6, 62 (average 3.31) compared with group II, fed with feed with eimeriostatic, but without the benefit of curative treatment, at which lesions were duodenitis and catarrhal typhlitis, congestive-catarrhal and hemorrhagic typhlitis, pericarditis, fibrinous perihepatitis and bleedings (as consequence of conditioning pathogenic flora graft), the lesion score being higher by 6.8 (average 3.4). In group III, the lesions were catarrhal type, and the value of lesion score was low 0.12 (average 0.6).

In chapter XII is presented the differential diagnosis of hemorrhagic enteritis diagnosed both in intensive and extensive farming systems to establish the parasitic etiology (eimeriosis, cestodosis, ascaridiosis), viral (Newcastle disease, avian flu) or bacterial (pasteurellosis, salmonellosis, clostridiosis, colibacillosis), diseases and injuries that have some common clinical manifestations. In this respect, there were taken samples from the bodies and faeces, for diagnosis of Newcastle disease by inoculating on conventional embryonated eggs, for avian flu by RT-PCR test, and there were performed inseminations from long bone and organs on common environments and selective one for the diagnosis of bacterial diseases and to diagnose the parasitosis there were used direct methods from curette of the intestinal mucosa and qualitative flotation methods to study the morphology of the present parasitic elements.

Therefore we conclude that the changes found in diseases, manifested by digestive disorders and enteritis under various forms, on young poultry, have negative repercussions by reducing weight gain, carcass depreciation and not least by death. It is therefore appropriate an early etiologic diagnosis and appropriate measures in order to reduce the losses.