## COMMENTS ON HISTOPATHOLOGICAL CHANGES IN RABBIT LIVER WITH EIMERIOSIS

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#### Abstract

The study was conducted on a total of 117 White New Zeeland breed rabbits, which were identified by feces examination with varying degrees of infestations with Eimeria sp., Eimeriosis hepatic lesions being identified in 20 of them.

Specific lesions were hepatic hypertrophy with presence of necrotic miliary nodular centres, vesicular looking angiocolitis, apostematous hepatitis and cirrhosis. Histopathological examination pursued in particular the consequences of sexual ongoing phase. Biliary ducts were dilated with hyperplastic epithelial reaction and the formation of papillary reactions with the presence of asexual and sexual stages of development.

Key words: angiocolitis, eimeriosis, hepatitis, rabbits.

## INTRODUCTION

Considered of lesser extent than intestinal Eimeriosis, hepatic Eimeriosis caused by *Eimeria stiedae* is common worldwide and affects domestic rabbits and wild ones equally and possibly other leporidae. It is a serious form of coccidiosis which particularly affects rabbits older than 2-3 weeks.

Rabbits are responsive only from the 16 th day of life and at the age of 6 months are totally unresponsive due to an immunity installed after permanent contact with oocysts.

Immunity is installed in 21 days after infection. Full immunity under natural conditions is set after 3 months (Chowdhury and Fraser, 2008).

Main pathological lesions are angiocolitis, diffuse cirrhosis, nodular hepatitis, compression atrophy of hepatocytes around the bile ducts affected (Militaru et all., 1997).

Hyperplasia of bile duct epithelium is accelerated along sexual phase and epithelial damage and desquamation will prevail during the invasion of merozoites and schizonts (Chowdhury and Fraser, 2008). In histopathological analysis, severe congestion and dilation of central veins were observed and the endothelial lining were ruptured (Naimi et all., 2012).

Severe hyperplasia of the lining epithelium of the portal areas were detected forming fingerlike projections in the lumen of the bile duct (Ebtesam and Mathal, 2008).

Sinusoids were also congested in this connection and dilated with haemorrhagic spots.

Multiple areas of coagulative necrosis of hepatic cells surrounded with inflammatory cells also happened. In addition, cellular infiltration of lymphocytes in the infected liver was noticed (Zerrin and Yesari, 2007).

# MATERIALS AND METHODS

The study was conducted on a total of 117 rabbits of New Zealand White breed (medium breeds) and half-breed from research stations, identified with parasitic infestations of *Eimeria* sp. in 97 (82.9%) of the total number of rabbits examined; the diagnosis was established by identifying oocysts of *Eimeria* sp. uninfested. Coproparasitological examination was performed through qualitative methods-Willis amended by Lungu and quantitative.

In heavy infestations or clinical, rabbits were slaughtered and morphopathological examination was performed upon the liver and biliar ducts, followed by histopathological examination after sampling the specific lesions, fixed in neutral formalin saline and further processed for inclusion in paraffin. Paraffin blocks were sectioned at 6  $\mu$ m, stained preparations were obtained by the trichromatic method of Mallory, examined and microphotographied.

## **RESULTS AND DISCUSSION**

In intestinal Eimeriosis, a distinction depending on the species couldn't be done, but *Eimeria stidae* species responsible for Eimeriosis liver could be differentiated due to the particularities of size (31-42 x 17-25  $\mu$ m), shape (ellipsoidal, with one pole elongated), colour (pink orange) and layout of sporont (fig.1). Specific lesions were observed in the liver parenchyma and bile ducts caused by *Eimeria stidae* in Eimeriosis liver disease. In the center of the necrotic focus one could observe the presence of "old oocysts" with altered the absence of sporont, changed shape and altered wall structure.

Histopathological lesions in the liver showed the following symptoms: severe congestion and dilation of central veins, rupturing of the endothelial lining, hyperplasia of the epithelial lining of portal areas with finger-like projections in lumen of the bile duct, congestion and dilation of sinusoids with haemorrhage areas (fig.2).

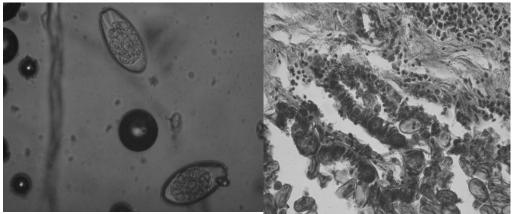


Fig. 1- Eimeria stiedae - unsporulated oocyst (20x)

Fig. 2 – Hiperplastic bile ducts; Col. Mallory trichromic; Ob. 40x

Multiple areas of coagulative necrosis of hepatic cells surrounded with inflammatory cells were found.

On the surface of the parenchyma one could observe white-yellowish areas with irregular shape with sizes ranging from 0.3 cm to 1.5 to 2 cm (fig.3).



Fig. 3 – Nodular necrotic hepatitis

These are centres of necrosis bounded by a congestive-haemorrhagic edge produced by trophozoites and merozoites during asexual multiplication in the destruction of hepatocytes and sinusoidal capillaries rupture.

The bile ducts could be observed with necrotic lesions of diffuse or nodular type and colonies of *Eimeria*.

In the bile ducts located within bile-port space, hyperplastic reactions both intraepithelial and in the lumen occurred with presence of coccidia (fig.4).

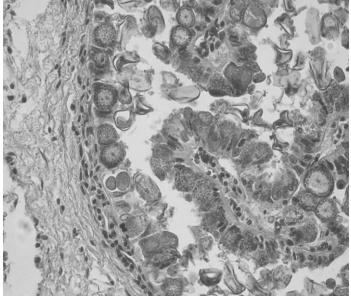


Fig. 4 - Angiocolitis; Col. Mallory trichromic; Ob. 20x

Hyperplastic bile ducts were surrounded by a conjunctival reaction type, accompanied by an inflammatory reaction of histiocytic lymph type.

The bile ducts were clearly dilated with hyperplastic reaction of simple prismatic epithelium, with formation of papillary reactions and the presence of asexual and sexual stages of development (micro- and macrogametocytes and oocysts in training).

In the lumen of the bile ducts there are numerous oocysts with thin shell of around 50  $\mu$ m with a specific structure of *Eimeria stiedae* (fig.5).

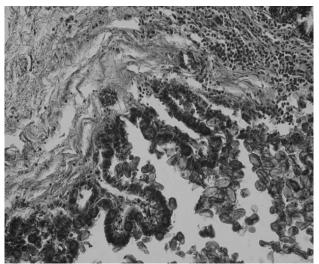


Fig. 5 – Angiocolitis with hyperplastic reaction of simple prismatic epithelium, with formation of papillary reactions; Col. Mallory trichromic; Ob. 20x

The liver lobules outlined necrotic lesions due to traumatic mechanical action of asexual forms (schizont and merozoite) and the destruction of liver parenchyma (Remack cords and sinusoidal capillaries). In the center of the necrotic area one can observe the presence of 'old' oocysts, with altered structure, characterized by the absence of sporont, changed shape and altered wall structure (fig.6).

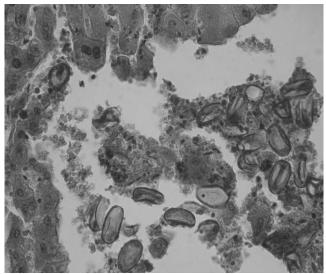


Fig. 6 - Necrotic hepatitis; Col. Mallory trichromic; Ob. 40x

#### CONCLUSIONS

The intensive growing conditions require the emergence and evolution of parasitary diseases where contributory factors (humidity 43-52%, temperature 17-20°C), feeding and manure disposal systems affect the development of infested forms (oocystes, eggs) in hepatic - Eimeriosis, intestinal Eimeriosis and some nematodosis – *Passalurus* sp.

Infestation with *Eimeria* sp. was identified in 97 (82.9%) of the total number of rabbits examined, 150, the highest incidence being in rabbits of 3-6 weeks which were identified in 82% of specimens examined.

Clinical expressions and lesions were evident in the liver Eimeriosis caused by Eimeria stiedae, characterized by necrotic hepatitis and angiocolitis in diffuse outbreaks.

Histological lesions are evident in hepatic Eimeriosis, in which bile duct hyperplasia and the occurrence of papillary reactions at the epithelial level with the presence of asexual stages of development could be noticed.

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# CLINICAL, RADIOLOGICAL AND MORPHOLOGICAL ASPECTS IN CHYLOTHORAX IN CATS – 5 CASES REPORT

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#### Abstract

Chylothorax represents the acumulation of lymph (chyle) within the pleural space. In the radiology service of FMV Iasi, during the last year (2012), 13 cases of pleural effusion were radiologically diagnosed.

Thoracocenthesis was performed on the cats and macroscopical, cytological and microbiological exams followed.

Five cases were diagnosed with chylothorax. The etiological diagnostic was established: one case was trauma determined, two of them were caused by mediastinal lymphoma, one was congenital recurrent chylothorax and one was caused by cardiac insufficiency.

Key words: chylothorax, lymphosarcome, pleural effusion, thoracocenthesis.

#### INTRODUCTION

Chylothorax represents the acumulation of lymph (chyle) within the pleural space. It results from the leakage of lymph from disrupted, obstructed or abnormal thoracic lymph channels (Fossum, 1993).

Common causes are: thoracic duct rupture (traumatic), lymphangiectasia of the anterior portion of the thoracic duct due to compresion from anterior mediastinal masses on anterior vena cava, cardiac insufficiency (dirofilariasis, cardiomyopathy), hyperthyroidism and anterior vena cava thrombosis (Fossum, 1993; Leib, Monroe, 1997). Other causes are mediastinal lymphome, torsion of a lung lobe, trauma, idiopathic or congenital

Clinical symptoms are: respiratory distress, cough, fainted heart sounds, fatigue and polydipsia. When the chylothorax is old, weight loss and anorexia are present (Leib, Monroe, 1997).

#### MATERIAL AND METHOD

In the radiology service of FMV Iasi, during the last year (2012), 13 cats were radiologically diagnosed with pleural effusion. The pacient's age varied from 6 months to 17 years. The symptoms were those of respiratory distress (tachipnea,

abdominal and open mouth breathing), weight loss, fatigue, none of them presented polidipsia.

Clinical examination revealed the loss of the thoracic elasticity, increased matitee on thoracic percussion, decreased lung sounds ventrally and increased bronchoalveolar sounds dorsally and bilaterally muffled heart sounds.

The thoracic x-ray examination was performed in left and right lateral incidence and dorso-ventral incidence, trying to keep the pacient as confortable as possible, knowing that respiratory distress may increase when the animal is stressed, especially in lateral recumbency.

The presence of various quantities of liquid was diagnosed by the large area of radioopacity that covered the lower part of the thorax in lateral incidences, covering the heart silhouette and different parts of the lung lobes, displacing the trachea dorsally and pressing the remaining normal lung lobes, modifying their possition and making them look like leaves, with round edges. In dorso-ventral position, the radioopacity – presence of liquid – surrounded the lungs, displacing them from the thoracic wall.

Thoracocentesis was performed on the cats, uni- or bilaterally, and macroscopic and cytological exams followed. Chylothorax was diagnosed in 5 cats.

## RESULTS

In all the 5 cases of chylothorax, the radiographs were made before and after the fluid drainage, as seen in Fig.1 and 2. bellow. This helps evaluating the pulmonary tissue, respiratory capacity heart shape. The Fig. 3 presents the same case, after complete drainage and specific treatment.

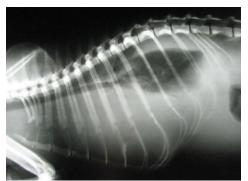


Fig. 1.—Rx lat, cat, plural and peritoneal effusion (before drainage)

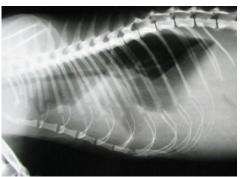


Fig. 2.—Rx lat, cat, plural and peritoneal effusion (after partial drainage)

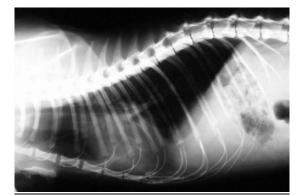


Fig. 3.—Rx lat, cat, plural and peritoneal effusion (after complete drainage and treatment)

In the first image the small respiratory space is evident, the collapsed lungs, the elevated trachea, the radioopaque liquid that covers everything, even the shape of the heart. In the second image, the lungs are seen better, closer to the right position, the heart appears enlarged, still masked by the liquid and the trachea displaced dorsally.

The last image shows an almost normal appearence of the thorax, the trachea was less elevated, the heart was enlarged and the fissure lines between the lung lobes were present. At the moment of the first radiography, the nature of the collection wasn't suspected.

Pleural effusion was drain in every case, as much as possible. For this action, in the usual election place  $(7^{th} \text{ or } 8^{th} \text{ intercostal space, on right, at the costo-condral junction, and the 6^{th} or 7^{th} intercostal space on left) the hair was shaved and the asepsie was made with alcohol or betadine.$ 



Fig. 4.—Drainige of pleural fluid on the left side

The position of the pacients was on ventral recumbency, as confortable as possible, as seen on Fig. 4. At need, a light sedation was made.

The toracocentesis was performed using 20G or 22G intravenous catethers, depending on the density of the fluid. After the catether is placed, a sterile 2 ml seringe is drain out, then a 10 or 20 ml seringe can be used to drain the rest of the liquid.

Macroscopical examination of the fluid and refractometry vas performed.

The macroscopic appearance of the effusion and the quantity demonstrate a variety of colors (Meyer, Franks, 1987), as seen in Fig. 5-8. Chylothorax varied from milk white to pale rose and almost red. That depended on the etiology and the time of the accumulation. In traumas cases followed by the rupture of the thoracic channel, other vassels breake too. So blood was present in the effusion too. In lymphoma, cardiac insufficiency and in congenital chylothorax, the macroscopic aspect was dense white, pale white or pale rose.



Fig. 5.— Important quantity (300 ml) of milky white liquid—chylothorax.



Fig.7.— 200 ml of rose, dense fluid chylothorax with small quantity of blood cells.



Fig. 6.— Large quantity (250 ml) of red, dense liquid—chylothorax with large quantity of blood cells.



Fig. 8.— Important quantity (250 ml) of dense, almost red liquid—chylothorax and large quantity of old bood cells .

<u>Cytological examination</u> - during the punction of the pleural space, a sterile liquid sample is being prelevated on an EDTA tube (Fig. 9). The samples are cetrifugated, smeares are made and colored MGG and they are microsopically examined (Fig. 10 - 12)



Fig. 9.—Chylous effusion, lactescent aspect

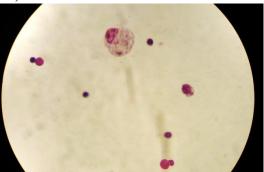


Fig. 10.— Cat. Chylous pleural effusion. Small lymfocites , G.N Macrophages. Col, MGG x 1000.

The EDTA samples are being sent to the cytology laboratory, were there are analysed. The samples were centrifuged, smeares were made from the sediment and coloured MGG, then examined under microscope, first using small lens (x10, x40) and later on immersion lens (x100).

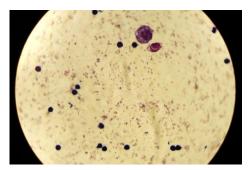


Fig. 11. - Cat. Chylous pleural effusion Small lymfocites, G.N.Macrophages, rare eritrocytes. Col, MGG x 1000.

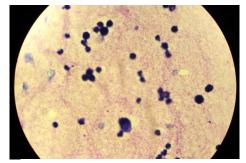


Fig. 12. - Cat. Chylous pleural effusion.Isolated and agglomerated lymfocites,G.N.Macrophages. Possible lymphoma. Col,MGG x 1000

<u>Ultrasound</u> examination of the mediastinum in search of mediastinal masses and the cardiac examination were performed. In one case a left heart dilatation was found.

Using all these methods, an ethiological diagnostic was established in each case. This is very important for the therapeuthic protocole that is to be followed.

# CONCLUSIONS

Five cats were diagnosed with chylothorax, by the use of radiography, pleural punction, macroscopical evaluation and cytological examination.

The ethiology of these cases was established: one traumatic old chylothorax, two mediastinal lymphomas, one left heart cardiomiopathy and one congenital, recurrent chylothorax, remittent under treatment.

The drainage of the pleural effusion is very important for the good quality radiography, the fluid analysis and for the releave of the pacient.

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