

Pathomorphological changes in the small intestine and liver of the European beaver (*Castor fiber* L. 1758): a case study

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Abstract: *Pathomorphological changes in the small intestine and liver of the European beaver (Castor fiber L. 1758): a case study.* The aim of the study is to examine and describe the morphology and patomorphology of small intestine and liver of European beaver (*Castor fiber*). In natural environment beavers are in constant contact with various both internal and external parasites. Histological analysis revealed the pathological changes within the digestive track and liver indicating long-term inflammation. It was hypothesized that the observed inflammation was caused by the parasites not related to beavers so far.

Key words: beaver, small intestine, liver, pathology, parasites

INTRODUCTION

European beaver is the largest rodent occurring in the area of Eurasia (Wilson 1971, Żurowski and Kasperczyk 1986, Halley et al. 2012). It is adapted to a semi aquatic lifestyle. This type of lifestyle is currently possible due to specific body adaptations that developed naturally in successive generations living in

such environment. In the course of evolution, interdigital webbing developed between the chest limb fingers while the animal tail was specifically widened. These adaptations allowed beaver good motility in water. Moreover, other organs are also well accommodated. Thanks to one of those adaptations beaver is able to dive in water for 15 min without a break. This allows beavers to avoid natural enemies as well as feed underwater (Czech 2010). Beaver is relatively unique animal that can adapt the environment to its particular needs. Such action requires specific type of logic and forecasting thinking and in the case of beaver it is possible due to relatively large brain. Rodents are characterized by the greatest ratio of brain to body mass. By cutting down the trees, which diameter can reach up to 1 m, beaver creates dams in the small rivers and streams (Hägglund and Sjöberg 1999). Backwaters that are formed this way become the territory where beavers create lodges, the areas

of breeding and reproduction (Żurowski 1992). They feed on almost all species of coastal and aquatic plants that grow in its space of living (Lahti and Helminen 1974, Vaughan et al. 2000, Czech 2010). Together with food, beavers absorb parasites, mainly flukes (Máca et al. 2015) and nematodes (Drózdź et al. 2000). Beaver intestines can colonize protozoa like *Cryptosporidium* and *Giardia*, which spread via contaminated aquatic environment (Paziewska et al. 2007). Based on the samples taken from three individuals, pathological lesions in the small intestine and liver typical for the European beaver was describe in the paper.

MATERIAL AND METHODS

Three beaver individuals were retrieved in the Population Reducing Program of the National Fishnig Farm in Kobryn, Belarus with the official permission of the Belarusian Minister of Environment. For histological examination, tissue was harvested from the middle of the small intestine and liver. Obtained tissues were fixed with 4% buffered formalin and then washed in water for 12 h. Afterwards the tissue was dehydrated in portions of ethanol with increasing concentrations of 70, 96 and 99.8%, respectively. After dehydration tissue fragments were triplicate rinsed in xylene and paraffin for 2 h. Prepared tissues were embedded in paraffin using Microm EC 350-1 and 350-2 EC cooling plate. Paraffin blocks were sliced using a microtome Microm HM 355S into the sections with the thickness of 3.5 μm , which were dried for 24 h at 37°C. Then the sections were deparaffinized in xylene and rehydrated

in ethanol in concentrations of 99.8, 96 and 70%. Then tissues were immersed in double-distilled water. The staining was performed with hematoxylin and eosin. Following staining, samples were again dehydrated with ethanol of increasing concentrations. Dehydrated and stained slides were treated with xylene and covered by coverslip with synthetic resin DPX. Slides were dried for 78 h at 37°C. Microscopic evaluation was conducted by light microscope Leica DM 750, equipped with a color camera Leica ICC 50 HD. Five separated areas from the intestine and liver samples taken from three animals were studied. Tissues examination were performed using Leica LAS EZ software.

RESULTS AND DISCUSSION

Intestinal villi and enterocytes were partially autolyzed post-mortally due to literature methodology (Madej et al. 2007). Leukocytes infiltrations, consisting of lymphocytes and eosinophils was observed within studied villi (Fig. 1).

The slides of the small intestine were of typical characteristic structure, however intestinal villi were partially distorted and significantly thickened. Paneth cells were clearly visible and normally located in the bottom of the mucous crypts. The cells contained the characteristic eosinophilic granules (Fig. 2). The lymph nodules were widespread in the intestinal villi in normal fashion. They represent large groups of lymphoblasts, clearly distinguishable in the wall of intestine. Significant number of macrophages capable of phagocytosis was noticed as well.

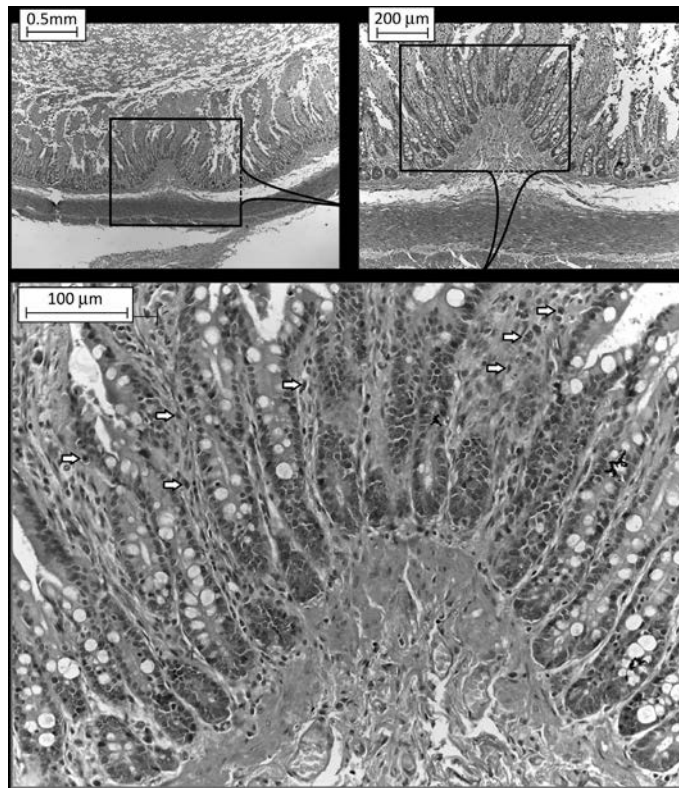


FIGURE 1. Intestine villi with eosinophils indicated by arrows

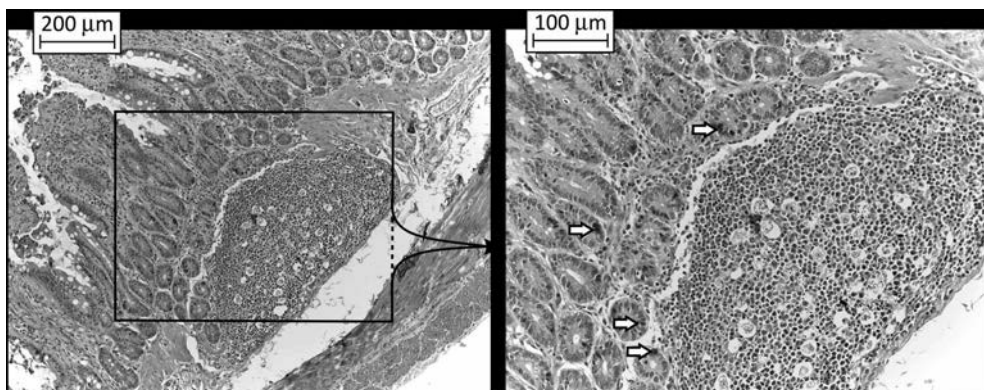


FIGURE 2. Intestine crypts with visible Paneth cells (arrows), lymph nodules of the small intestine

Slides of the liver tissue, allow for an accurate observations of its structure. Beaver liver has a macroscopic structure characteristic rodents livers. Liver lobes are not clearly separated due to small amount of the connective tissue between

them therefore borders are not clearly visible (Kuryszko and Zarzycki 2000) – Figure 3. Olive color tissues occurred between the hepatocytes. It represented the bile, which was stagnated in the liver bile tubules and ducts. It may be referred to the necrosis of the hepatocytes, noticed in the analyzed slides. The necrosis was presented by the shrinking and destructive changes within cell nuclei. Some of cell nuclei undergo contraction that results in cell destruction and death.

connective tissue, resulted from chronic inflammation (Madej and Rotkiewicz 2007, 2011). It is also confirmed by the presence of the inflammatory infiltrates of the lymphocytes and eosinophils. The presence of the eosinophilic suggests the presence of the parasites in the analyzed organism. It should be noted that Maca et al. (2015) found the flukes of the *Stichorchis subtriquetrus* in the gastrointestinal tract of the beavers body examined in their study. It was hypothesize that

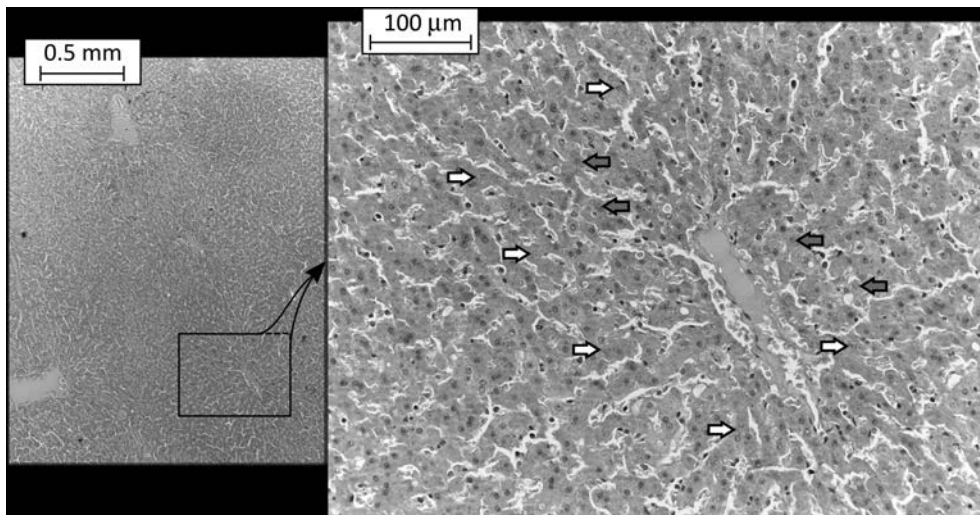


FIGURE 3. Liver with visible bile droplets (light arrows) and dark cell nuclei (dark arrows)

The degradation of intestinal tissue was most probably due to sampling long time after animal death. The tissues were improperly preserved and the enzymes partially digested the structures of tissues. Microscopic analysis of interstitial tissue indicated widespread and long-term inflammation. The presence of the chronic inflammation is also confirmed by the thickened villi. The increased thickness is associated with extensive growth of

mentioned parasite is the main factor of the inflammatory changes. Demiaszkiewicz et al. (2014) showed that *S. subtriquetrus* is a common parasite in beaver and the flukes usually locate in the further part of the digestive tract, inhabiting mainly the cecum that way. The small intestine is commonly inhabited by juvenile forms of the parasite. The inflammatory changes caused by the action of *S. subtriquetrus* were also observed in the

cecum and therefore the changes were potentially caused by the presence of juvenile forms of the flukes (Demiaszkiewicz et al. 2014). The study of Demiaszkiewicz et al. (2014) shows that the parasites such a *Psiloterma castoris* or *Trichostrongylus capricola* may also appear within beaver small intestine.

Slack of bile found in the liver can be associated with the blockage of bile outflow caused by parasites or gallstones (Madej and Rotkiewicz 2007). Cholestasis observed was most probably related to the hepatocytes necrosis, however, unequivocal identification of its robust reason was difficult. The liver changes occurred most likely during lifespan but after the animal death. It was hypothesized that the liver changes occurred due to the action of toxic products of parasites metabolism. The infection of the rodents may also be caused by the *Babesia* parasites. Demiaszkiewicz et al. (2014) did not report presence of Piroplasmid or any other parasites in the beaver blood samples. Kadulski (1998) reported the presence of ticks: *Ixodes apronophorus* and *Ixodes hexagonus*, in the beaver skin. *Ixodes hexagonus* is especially interesting as it may transfer other parasites that cause babesiosis, like *Babesia microti* (Bristol University, tick ID). Babesiosis is considered to be the type parasitosis which does not occur among the beavers.

Beaver, which is the subject of many studies, remains interesting research object as its physiological and pathological nature is unknown. Its biology certainly still hides many secrets and unraveling them could be a new discovery, also in the field of parasitology.

CONCLUSIONS

Beavers as the wild animals remain in the constant contact with different endo and ectoparasites. Microscopic observations of the small intestine revealed several pathological changes such an extensive connective tissue growth and the presence of lymphocytes and eosinophils, suggesting the state of long-term inflammation. It was assumed that the observed inflammation was caused by *Stichorchis subtriquetrus*, found in the gastrointestinal tract. Moreover cholestasis was observed in the liver samples. The genus *Ixodes* ticks found on the beaver skin, suggested the infection of parasites of the genus *Babesia*. Further studies did not find the parasites in the examined beaver's blood. Currently study results although of basic scientific knowledge can be soon applied for diagnostic of living animals health.

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Streszczenie: *Zmiany patomorfologiczne w jelicie cienkim i wątrobie bobra europejskiego (Castor fiber L.1758): studium przypadku.* Celem pracy jest ocena patomorfologiczna jelita cienkiego i wątroby bobra europejskiego (*Castor fiber*). W naturalnym środowisku bobry mają stały kontakt z pasożytami wewnętrznymi i zewnętrznymi. Ocena preparatów histologicznych wykazała zmiany patologiczne w obrębie układu pokarmowego i wątroby, wskazując na przewlekły stan zapalny. Założono hipotezę, że przyczyną obserwowanych patologicznych zmian w tkankach jest zapalenie wywołane pasożytami, jakich wcześniej nie stwierdzano u bobrów.

Słowa kluczowe: bóbr, jelito cienkie, wątroba, patomorfologia, pasożyty

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