ABSTRACT

Key words: brown bear, morphology, anatomy, digestive system.

The PhD thesis entitled "**Normal morphology of the digestive system in brown bear (*Ursus arctos*)**" was elaborated within the Doctoral School of "*Ion Ionescu de la Brad*" *University of Agricultural Sciences and Veterinary Medicine* of Iaşi, during four years of studies, between October 1st, 2009, and October 1st, 2013, and it is structured according to the present legal provisions in two main parts: the first part, with the title "Literature review", contains 37 pages, and it represents 27%, and the second part, with the title "Personal research", is extended on 97 pages, and it represents 73%.

The first part, structured in two chapters, succinctly presents information from the literature referring to the subject of the PhD thesis, information that was subsequently used for the interpretation of the data obtained in the second part. This part is illustrated by 14 figures selected as suggestive for detailing the information synthesised.

The first chapter - **"Normal morphology of the digestive system at domestic carnivores"-** presents the normal morphology of the digestive system of the dog, because the bibliographical titles refer to the morphology of the digestive systems of the bears compared with that of the dogs. The main characteristics, both anatomical and histological of this species were presented, pointing the main morphological aspects.

Chapter II - " **Bibliographic data on normal morphology of the digestive system of the brown bear (*Ursus arctos*)**"- presents the data found in the literature, both from Romania and abroad. The data on the morphology of the digestive system of the brown bear are insufficient and rather succinct, being fragmentary and general, without suggestive pictures to exemplify the information provided.

The second part is structured in five chapters (chapters III-VII), as follows: chapter III – ***Aim and objectives***, chapter IV –***Material and methods***, chapter V –***Normal morphology of the prediaphragmatic digestive tract in brown bear* (*Ursus arctos*)**, chapter VI -***Normal morphology of the postdiaphragmatic digestive tract in brown bear* (*Ursus arctos*)**, chapter VII - ***Normal morphology of the glands associated with digestive tract in brown bear (Ursus arctos)***, ending this part with ***final conclusions***. All the images in the second part are original,

taken on the field or in the laboratories where the research took place, except those taken from two skulls of adult bears, one from *Natural History Museum* in Iaşi and one from *Faculty of Biology*, Iaşi.

In chapter III– ***Aim and objectives*** – we motivated the choice of the subject. The main purpose of the research is the description of the normal morphology of the digestive system of the brown bear (*Ursus arctos*), from anatomical and histological points of view, with the realisation of images to illustrate the main morphological aspects.

The study of the literature showed the existence of little preoccupation concerning the morphology of the digestive system of the brown bear, and also the morphology of this species in general. Most of the bibliographical titles studying the brown bear (*Ursus arctos*) contain information concerning the ethology and the conservation of the species.

The bibliographical titles analysing the normal morphologyof the digestive system of the brown bear, both from Romania and abroad, did not focus on the description of all the segments, presenting images and enumerating the main macroscopic and microscopic aspects; instead, they only made a brief description.

The literature studied contained the description of morfopathological and histopathological lesions in bears living in captivity, but without comparisons, and without any references to the normal morphology.

The research for the knowledge of the anatomy of cinegetic fauna on the territory of our country is incomplete, and the field of research belongs equally to biologists, veterinary doctors, and silviculture workers. This is the reason of the present PhD thesis.

In a continuous increase of the competences of knowledge in the veterinary medicine field, and with an extension of the syllabus within the Faculty of Veterinary Medicine of Iaşi, with subjects like "*Anatomy and physiology of animals for fur*" and "*Management of cinegetic production*", the present research support this disciplines, required in preparing for future veterinarians.

The knowledge of the morphology of the digestive system of the brown bear does not have only a biological importance; it also has a practical one. A quality of this knowledge may help to provide data that could be used in surgical interventions on different segments of the digestive system, and also on the abdominal cavity.

Consequently, the main objectives and the corresponding activitie of the PhD thesis were pre-established in agreement with the scientific coordinators of the doctoral thesis, and they are as follows:

- identification of hunting funds with populations of relatively big bears;

- participation in organised hunting;
- participation in the realisation of bear necropsy;
- realisation of morphological and macroscopic investigations in the necropsied individuals;
- collection of fragments of the segments of digestive tube and annex glands, with specific fixators;
- paraffin inclusion and sectioning of the paraffin blocks (5 µm thickness);
- staining the sections;
- interpretation under the microscope, taking pictures of the main histological aspects.

Chapter IV- ***Material and methods*** – presents the research material and the methods used. The research was done on cases obtained from organised hunting in Braşov and Bacău Counties, and also from the Zoo in Focşani, from the "*Rehabilitation Centre of Orphan Bears*" in Bălan, Harghita County, and from two skulls of adult bears, one from *Natural History Museum* in Iaşi and one from *Faculty of Biology*, Iaşi.

Also, two adult bears hunted in Braşov area were examined briefly in field conditions. Only the gastrointestinal mass was examined at the bear from Bacău County, within the Faculty of Veterinary Medicine of Iaşi, in the laboratory of anatomy.

In Focşani we made the laboratory necropsy of a brown bear female from the Zoo.

We also examined two brown bear cubs, aged approximately 3 and 4 months, one on the field, the other within the Faculty of Veterinary Medicine of Iaşi, in the laboratory of anatomy.

The investigations on the dentition of the brown bear were realised also on two skulls ofadult bear, in order to take pictures and to analyse the main aspects.

In what concerns the work methods, we made the macroscopic and microscopic examination.

The macroscopic examination was realised briefly on the field, and in the laboratory, at the same time with the necropsy examination realised according to *Kitt's* holoptic method, which means opening the great serous cavities, a previous *in situ* inspection of organic systems and apparatuses, their evisceration in block, and their examination according to the common methodology.

A first stage consisted in skinning the corpses which were positioned in dorsal decubitus, and the second stage consisted in opening and examining the abdominal cavity and the peritoneal cavity.

The actual examination of the digestive system was performed by inspection, analysing the aspect and the dimension of the organs, the topographic reports and the fixation and

connection means intermediated by the serous ligaments, in the case of those placed intracavitary.

The microscopic examination consisted in the collection, fixation, paraffin inclusion, microtome sectioning, staining, mounting and actual microscopic examination -operating steps of the technique of obtaining the permanent histological preparation using the method of paraffin inclusion.

The histological preparations were examined with *Motic B1-211A* optical microscope, with *Moticam 1000* video camera attached, and they were histologically interpreted.

The staining methods used werethe following: HEA, Masson, PAS, Van Gieson and Gömori, for general tissue aspects.

Chapter V - ***Normal morphology of the prediaphragmatic digestive tract in brown bear (Ursus arctos)*** - was carried out to systematise the main characteristics of this segment belonging to the digestive tube. We realised the examination of the oral cavity, of the pharynx, and of the esophagus.

In the oral cavity we intended to see the carnivorous character regarding the dentition, and to establish if the brown bear is an authentic carnivore. The characteristics of the dentition at the age of 3 months and at the adult age were described on the skulls. The floor, the roof, the side walls, and the tongue were also described in the oral cavity. This chapter also contains the description of the pharynx and of the esophagus, analysing the main histological characteristics ofthe latter.

The roof of the oral cavity is approximately lyre-shaped, having a thick palate mucosa, which structures 8-10 palatine crests arranged under the shape of symmetrical bows, along the entire length of the hard palate, joining in the sagittal plane, without being separated by the palatine raphe, which is barely shaped.

The palatine velum extends up to the basis of the epiglottis. It is oriented almost horizontally, so as its aboral surface appears as an extension of the nasal roof.

The tongue is flattened dorso-ventrally, without lingual protuberance, with a thin tip, slightly flattened, with sharp free edges. The ventral surface of the tongue presents the *lyssa*, underlying the mucosa, in the median plane, between the genioglossus muscles. *Lyssa* is a fusiform fibrous cord reaching the tip of the tongue. The twenty caliciform papillae, with circular aspect, are arranged on two divergentV-shaped rows, with oral opening. The foliate papillae are small, without forming a gustative organ.

The brown bear has a heterodont, diphyodont, brachyodont, anisognathous dentition. The temporary dentition has 32 teeth with isognathous arrangement: I 3/3, C 1/1, M 4/4. The

permanent dentition has 42 teeth, with anisognathous arrangement: I 3/3, C 1/1, P 4/4, M 2/3. The brown bear is far from having the dentition of an authentic carnivore, because the molars do not have sharp crests, prerogative of canines and felines. All the molars experienced a morphological modification, as a result of the influence of the omnivorous nutrition, as a necessity to adapt to a more active mastication. The cuspid teeth are small, rounded, and flattenedin the mediolateral position.

The pharynx has narrow and elongated *choanae*, and the palatopharyngeal arches are small, and they do not join at the level of the esophagus vestibule.

The esophagus mucosahas a stratified keratinised pavimentous epithelium and a very vascularised *lamina propria.* The latter is separated by a circular mucous fold (*Lymen pharyngoesophageum*) from the pharyngeal mucous.

Chapter VI - ***Normal morphology of the postdiaphragmatic digestive tract in brown bear* (*Ursus arctos*)** - was realised in order to describe macroscopically every segment belonging to the postdiaphragmatic digestive tube, topographic position, relationships with the neighbouring organs, collection of fragments belonging to it, with specific fixators, paraffin inclusion and sectioning of the paraffin blocks (of 5µm thickness), staining of the sections, interpretation under the microscope, taking pictures of the main histological aspects.

The research activities from this chapter are fundamental research, because the integral anatomical and histological study of the post-diaphragmatic digestive tube of the brown bear (*Ursus arctos*) is described for the first time in Romania. Acquiring this knowledge provides data that can be used in surgical interventions on different segments of the digestive system, and on the abdominal cavity.

The stomach of the brown bear has a single cavity, and it is relatively small as compared to the size of the animal. It is shaped like a recurved sack, slightly flattened antero-posteriorly, with piriform aspect, highlighted by the position of the cardia and the deep ness of the angular incisure. The dimensions for the greater and lesser curvatures are 30 and 60 cm, respectively, in full state. The position of the stomach in the abdominal cavity is transversal, from left to right, in contact with the ventral abdominal wall, immediately caudal by the hypochondral arch and the xiphoid appendix. The cardiac orifice is structured by a little developed, relatively wide, with permissive sphincter. The pyloric orifice circumscribed by a strong sphincter is provided with a pyloric torus. From a macroscopic point of view, we can distinguish two areas of the gastric mucosa. The main area, of grey colour, which is occupied by the actual gastric glands, covers approximately two thirds of the internal surface. The other portion, occupying a third, of a lighter colour, contains pyloric glands. The proventricular and cardiac mucosaeare little extended, being

placed around the cardia. The cardial glands are simple, made of mucous cells, and rare parietal cells. The fundic glands are structured from mucous cells, main cells, parietal cells, and argentaffin cells. The pyloric glands are shorter, but more ramified, formed mostly of mucous cells, and very rare parietal cells.

Greater omentum is formed of two laminae, one cranial, and the other caudal. The cranial lamina of the greater omentum covers the lateral left lobe and the medial left lobe of the liver. The caudal lamina of the greater omentum partially covers the intestinal mass, therefore the jejun loops are in direct contact with the walls of the flanks, and with the ventral wall of the caudal half of the abdominal cavity.

The duodenum with the descending part, with the caudal flexure, and with the ascending part form the U-shaped duodenal loop, with the concavity oriented cranially. The major duodenal papilla, and the minor duodenal papilla are in the lumen, in the proximal segment belonging to the descending portion of the duodenum. Both are located on the lesser curvature of the duodenum, at 2 cm distance one from the other. In what concerns their dimensions, the major duodenal papilla has a smaller diameter than the minor duodenal papilla, 0.7 cm as compared to 1 cm, which means that the minor duodenal papilla corresponding to the opening of the accessory pancreatic duct, has a more important role.

The jejunal loops are big, and they fill most of the abdominal cavity, reaching cranially the stomach and the liver, caudally the entrance to the pelvic cavity, and ventrally leaning on the floor of the abdominal cavity, from which they are separated only in the cranial part by greater omentum .

The ileum could not be identified macroscopically, due to the absence of the caecum, and of the ileocecal fold, whose length is delimiting the ileum, at the others domestic animals.

The epithelium of the small intestine mucosais prismatic simple, with striated border, structured by intestinal villi with finger-likeaspect.

The large intestine is short, uniformly calibrated, with no haustra present. The caecum could not be identified in any of the individuals examined macroscopically. The small intestine continues at the exterior with only a visible thickness of the intestinal wall.

The colon has a length of approximately 1.2-1.5 m, and it is U-shaped with its three parts, with caudal opening.

The anal cutaneous zone has bilateral openings of the para-anal glands, also called para-anal sinuses (*Sinus paranales*).

The large intestine mucosa has no intestinal villi, and it is thicker than the one of the small intestine, with a simple prismatic epithelium, and with predominance of caliciform cells.

Chapter VII – ***Normal morphology of the glands associated with digestive tract in brown bear (Ursus arctos)***– was realised to study the degree of development of the annex glands belonging to the digestive tract, and also to describe anatomically the main particularities, the topographic position, then collection of fragments belonging to them, then the stages of obtaining the permanent histological preparations using the method of paraffin inclusion, and interpretation under the microscope and taking pictures of the main histological aspects.

The images and the interpretation concerning the histological aspects of the annex glands of the digestive system of the brown bear (*Ursus arctos*) are realised for the first time, and they are presented in this thesis .

The major salivary glands, liver and pancreas were described anatomically and histologically.

The parotid salivary gland is small, triangular, with the dorsal extremity incised in two parts, with a biramous aspect, and the parotid duct has a lateral trajectory, over the masseteric region, and it opens in the superior lateral oral vestibule, in the plane of the third premolar, at the level of the salivary papilla, which is not very visible in bears. Structurally, the parotid salivary gland is mixt, and it has both serous and mucous structure, with the clear delimitation and differentiation of the two types of acini.

The mandibular salivary gland is globulous, smaller than the parotid, and with little lobulation, having a histological structure which confirms the fact that it is a mixt gland.

The monostomatic sublingual salivary gland is developed, made of a compact and elongated mass, extending from the median third of the tongue to the mandibular salivary gland, being also placed posteriorly to the polystomatic sublingual salivary gland. *Bartholin* duct has adorsal trajectory, parallel to *Wharton* duct, opening in the apical sublingual space, shared with the previous one, at the level of an orifice protected by a very small sublingual caruncle with triunghiular shape. The histological structure of the monostomatic sublingual salivary gland is predominantly mucous.

The polystomatic sublingual salivary gland is made of small glandular lobules, and it is placed rostrally to the monostomatic sublingual salivary gland, histologically being a mixt gland, with mucous predominance.

The liveris placed before the second to the last rib in the left side, and at the level of the last rib in the right side. Its colour is brownish-red, and it does not have a lobular aspect, due to the lack of abundance of subcapsular connective tissue.

The lobation of the liver is given by the two main incisures and by the two deep accessory ones, dividing the liver in six well shaped lobes. The interlobar incisures are deep, but

they do not reach the hepatic hilum. The caudate lobe is small, divided in two pedicled processes: an actual caudate process, oriented to the right, and a papillary process oriented to the left. The quadrate lobeis well developed and individualised, with dimensions comparable with medial right and left lobes.

The serous fixation means of the liver are as follows: the coronary ligament, the falciform ligament, the right triangular ligament, the left triangular ligament, and lesser omentum.

The bile duct, resulted from the joining of left and right hepatic ducts with the cystic duct, opens at the same place as the main pancreatic duct in the anterior segment belonging to the descending portion of the duodenum, at the major duodenal papilla.

The gall-bladder in adults is developed, with a capacity of approximately 80-100 ml, being visible on both visceral and diaphragmatic sides, with a bottom going beyond the ventral limit of the liver.

The histological structure of the liver shows that there is a low quantity of interlobular connective tissue structuring the septa, therefore the lobules are not well delimited, and *Glisson* capsule is thin.

The pancreas is very developed, elongated and recurved, U-shaped, with caudal opening, consisting of the following: dorso-ventrally flattened body, located caudally to the pyloric region of the stomach, and into the cranial flexure of the duodenum; the left lobe, which is shorter, but wider, is located between the sheets of the greater omentum which accompanies the greater curvature of the stomach, insinuating among the stomach, the left kidney, and the base of the spleen; the right lobe is longer than the left lobe, in close contact with the descending portion of the duodenum.

The main pancreatic duct opens at the same place as the bile duct, at the major papilla, in the proximal segment belonging to the descending portion of the duodenum, on its lesser curvature, and the accessory pancreatic duct opens at the minor papilla.

The pancreas is organised according to the model of the parenchymatous organs, structured from serous acini, having at the exterior a cover and support tissue made of capsule, connective-vascular septa, and reticular stroma. One-three *Langerhans* islands are located among the serou sacini of the lobules, with endocrine cells polarised at the sinusoidal capillaries.