Anatomical Studies on the Morphology and Topography of Cavity Lymphnodes in Pigs

ABSTRACT

The lymphatic system, supported by the lymphatic vessels, is a vascular network in higher vertebrates, having essential roles, such as regulating tissue pressure, monitoring the immune system and absorbing fats from the diet. The lymphatic vessels constitute an unidirectional system that transports fluids and proteins, taking them from the interstitial space and returning them to the bloodstream. **The objectives** pursued in this study are the following: to identify the topography of the main cavity lymph nodes and the morphological and topographic variants that may occur; to identify the main interspecific histological characteristics of vessels and lymph nodes in pigs. **Materials and methods.** The study was carried out on 20 animals weighing 25-30 kg, aged about 3.5 months, of both sexes. They came from private breeders. Ultrasonographic investigations were also performed on half of them. The macro- and microscopic anatomical studies as well as the ultrasonographic investigations were performed at the Faculty of Veterinary Medicine in Bucharest. For macroscopic and topographic analysis, lymph nodes in both the thoracic and abdominal cavities were examined "in situ". **Results.** Caudal mediastinal lymph nodes were always fewer in number (between one and three). They were associated with the esophagus, immediately behind the tracheobronchial and ventral lymph nodes of the aorta. The afferents originated from the pericardium, the caudal part of the mediastinum and the corresponding part of the esophagus. The efferents connected to the tracheobronchial lymph nodes or could also approach some thoraco-aortic lymph nodes. The lumbo-aortic lymph nodes were located on the lateral surfaces of the aorta, starting from the diaphragmatic orifice to the origin of the deep iliac circumflex artery. The iliac lymphocenters were represented by large lymph nodes that formed a bundle at the origin of the celiac artery. Colic lymph nodes were located along the path of the right colonic artery. Splenic lymph nodes were placed on the path of the homonymous vessels. Their topography was between the aorta and the hilum of the spleen. In this sector there could be 2-4 lymph nodes, but there were others (2-5) located along the upper quarter of the spleen. The jejunal lymph nodes formed a double chain in the middle of the jejuno-ileum meso, about 30 cm long. The caudal mesenteric lymph nodes were reduced, located dorsally by the descending colon on the path of the caudal mesenteric artery. **Conclusions.** In the case of mediastinal lymph nodes, the anterior ones are present, but their topography differs significantly from individual to individual. The most important groups of visceral lymph nodes are jejunal and colic. The most important parietal lymphocenter is the ileosacral one. In it, we were able to describe for the first time some individual variants, namely: lateral iliac lymph nodes dominant in volume compared to the medial ones, the absence in some cases (10%) of anorectal lymph nodes in some specimens, in which, however, a compensation was observed by the development of a chain of small units on the path of the median sacral artery.

Key words: lymphatic system, thoracic lymphocenter, mediastinal lymphocenter, swine.

1. INTRODUCTION

“The lymphatic system has been mentioned in the history of medicine for many centuries.

Hippocrates referred to "white blood in knots", and Aristotle pointed out the existence of fibers containing a colorless fluid between blood vessels and nerves. In 1627, Gasparo Aselli was the first to officially recognize the lymphatic system, which was constantly studied in the seventeenth and eighteenth centuries, a period in which medicine began to explore more deeply aspects related to lymph, lymphatic drainage and lymphatic anatomy. However, it is only in recent decades that we have witnessed an explosion of scientific research on this system, highlighting its involvement in numerous conditions, including cancer" (Padera et al. 2016).

"The lymphatic system, supported by the lymphatic vessels, is a vascular network in higher vertebrates, having essential roles, such as regulating tissue pressure, monitoring the immune system and absorbing fats from the diet. The lymphatic vessels constitute a unidirectional system that transports fluids and proteins, taking them from the interstitial space and returning them to the bloodstream. The lymphatic system has several important functions, the first forms a link from the interstitial space to the venous system. This connection helps maintain interstitial fluid volume" (Margaris and Black 2012) and allows for the transport of proteins, peptides, and macromolecules (Randolph et al. 2017). "The second well-known function of the lymphatic system is the transport of antigens and antigen-presenting cells to the lymph nodes, in order to mount targeted immune responses if needed" (Padera et al. 2016).

"The notion of "animal model" in medicine generally represents a non-human living animal in which a pathological process or lesion transmitted parentally, naturally acquired or induced will be investigated. The purpose of its use is to solve a research hypothesis of a similar condition in the target human species" (Crisóstomo, et al., 2016; Dove and Alworth, 2015).

"Over time, many species of animals have served as experimental models. It is known that rodents are species that have great advantages, this makes them some of the most favorable in biomedical research. Although cardiovascular biology has benefited from an immense amount of information, especially from the molecular and cellular segment of biology, the limitation of rodent use has been due to intrinsic differences between rodents and humans in terms of heart rate, adrenergic receptor ratio, oxygen consumption, reaction to loss, and other physiological processes such as the absence of the plateau phase and the expression of contractile protein critical for the coupling process excitation-contraction, as well as the spontaneous reversal of experimentally induced ventricular fibrillation in normal sinus rhythm, which make the use of rodents and the extrapolation of data derived from these models problematic" (Lelovas et al., 2014). "Over the past two decades, researchers have agreed that among the animal species that lend themselves to being used as experimental models, the closest in terms of anatomy and physiology of the cardiovascular system is the porcine model" (Lelovas et al., 2014; Ntonas et al., 2020, Nykonenko et al., 2017; Shah, 2022; Skjennald, 1982).

"Both structurally and functionally, the body of this animal has many similarities with the human one. In recent years, the species *Sus scrofa domesticus* has been intensively used as experimental material, especially in research carried out on various diseases of the circulatory system. In order to implement new surgical procedures and techniques, researchers first tried to know perfectly the morphological substrate on which, in the case of the above-mentioned field, vascular anatomy acts. Different procedures were used, such as injections of vessels with contrast agents, followed by dissections under a stereomicroscope or their follow-up by various paraclinical methods such as X-rays, computer tomography or MRI. Starting in 1930, when a study on the anatomy of the circulatory system of pigs appeared, in a classic reference work" (Sisson et al. 1975), and until now, when morphology works based on more modern investigation procedures appeared, the creation of a database necessary for experimental medicine and veterinary practice has been constantly pursued.

"Currently, in order to understand the topography and distribution of the main cavity lymph nodes in pigs, those interested generally turn to the descriptions in anatomy treatises by established authors such as Barone, König, Sisson or Dyce" (Sisson et al. 1975; König & Bragulla, 2007; Barone, 2012; König, 2014; Dyce et al., 2018;). However, they do not always provide the necessary details and we have found, as well as other authors (Yanina et al. 2024 Hsu and Itkin, 2016), that in the recent literature there are not enough morphometric studies of the lymph nodes of the thoracic and abdominal cavities. "For this reason, problems arise especially in experimental surgery, in the case of using devices (probes, implants) incorrectly calibrated to the dimensions of the animal used. In order to determine morphometric details, the authors used both paraclinical methods, such as high-frequency ultrasonography (UHFUS)" (Damian et al., 2015, Storkholm et al., 1997) or radiological examinations with contrast agents, but also classic dissection methods with the use of contrast agents injected into the arterial bed (Chirilean et al., 2010; Predoi et al., 2002).

"Since the middle of the last century, anatomists who have been studying the lymphatic system in mammals have found an interesting thing, namely that in pigs they are structurally different from those in other domestic animals. On the one hand, it has been observed that in the central part of the lymph node the cortical is arranged" (Trautmann and Fiebiger 1952, Spalding and Heath 1986, Anderson 2019) and on the other hand functionally, the passage of lymphocytes from the lymph to the blood takes place inside the lymph node (Drexhage et al. 1973, Bennell and Husband 1981). "It has also been observed that the hilum, present in other species, is not well defined on the surface of the lymph node in pigs" (Semeraro and DAVIES 1986, Drexhage et al. 1973).

"With the advent of new imaging techniques for investigation, it was possible to describe in detail not only the lymph nodes but also the lymphatic vessels in this species. Moreover, by using microinjection techniques, it was possible to define the lymphatic drainage territories by drawing up the so-called lymphosomes" (Suami and Scaglioni 2018, Suami et al. 2017). The authors concluded that this species could be used as a large animal model for research of the lymphatic system due to the anatomical structure of lymphosomes and the considerable caliber of lymphatic vessels.

Since we found on the one hand that in the literature there are some small inconsistencies related to the topography of the abdominal lymph nodes in this species and on the other hand, that we did not find any source in which these structures are examined by the ultrasound method in this species, we tried to approach in this study a detailed description of the morphology and topography of these lymph nodes as well as the possibility of identifying the clusters by the ultrasound method.

The objectives pursued in this study are the following: to identify the topography of the main cavity lymph nodes and the morphological and topographic variants that may occur; to identify the main interspecific histological characteristics of vessels and lymph nodes in pigs; to identify the best incidences of ultasonographic examination of lymph nodes in pigs.

2. MATERIALS AND METHODS

The study was carried out on 20 animals weighing 25-30 kg, aged about 3.5 months, of both sexes, 13 males and 7 females. They came from private breeders. Ultrasonographic investigations were also performed on half of them. The macro- and microscopic anatomical studies as well as the ultrasonographic investigations were performed at the Faculty of Veterinary Medicine in Bucharest.

For macroscopic and topographic analysis, lymph nodes in both the thoracic and abdominal cavities were examined "in situ". They were identified, from a terminological point of view, measured with the stool and photographed.

For the histological examinations after the euthanasia stage, tissue fragments were taken (thoracic aortic segment, pulmonary trunk, caudal vena cava, mesenteric vessels at the level of the jejunal insertion, mesenteric lymph node, tracheobronchial lymph node, superficial inguinal lymph node, thymus, artery and caudal mesenteric vein), segments with a longitudinal axis length of about 1.0 cm that were later immersed to fix, in a buffered solution of formaldehyde, with a concentration of 10%, for 24 hours.

After partial fixation, the tissue samples obtained reached the modeling stage, each piece being sectioned into several fragments with a thickness of about 2-3 mm, which were placed in cassettes with large or small meshes and put in another 10% formaldehyde solution, and then remained to be fixed for another 12 hours.

The next step was the introduction of samples into the histoprocessor for dehydration and pre-inclusion in paraffin. This technique consisted of carrying out standard steps: fixation of the samples in 10% formaldehyde buffered solution for 24 hours, followed by washing, dehydration in successive baths of 70%, 96% and absolute alcohol, followed by inclusion in paraffin. Processing and paraffin inclusion were done automatically in the Thermo Scientific STP 120-3 tissue processor, and paraffin inclusion was done using the Thermo Scientific Microm EC 350-1 station. The formation of the paraffin blocks was carried out by using the "Tissue-Tek Tec Sakura" embedding station, and the sectioning of the parts to the size of 4-5 µm was done with the "Thermo Scientific HM 340E" microtome. The sections thus obtained were spread on histological slides, stained and then fixed with Canada balm. For the histopathological study, the preparations were usually stained with Hematoxylin-Eosin (HE).

***Table 1*.**
Hematoxylin-Eosin (HE) staining stages

Stage	***Reagents***	***Duration***
Color	Hematoxylin	8 min
Washing	Water	10 min
Color	Eosin	1 min
Washing	Water	-
Dehydration	4 successive baths in alcohol of different concentrations.	-

After inclusion, the blocks were sectioned to 4 µm using the Leica RM 125RTS microtome and spread on histological slides. The slides were colored using the Thermo Scientific Microm HMS 70 automatic slide coloring machine. Hematoxylin-eosin (HE) staining was used, which allows a good differentiation of cell types (Table 1).

Analysis of microscopic images - The microscopic examination of histopathological preparations was performed using the Olympus CX23 microscope, equipped with a digital camera for capturing images, model Olympus SC50. The complete and detailed examination of the preparations was carried out by successively using the x4, x10, x20 and x40 lenses.

Our experiment was carried out in the anatomy laboratory of the Faculty of Veterinary Medicine in Bucharest, benefiting from the favorable approval of the Ethics Commission of this institution and met the conditions required by EU Directive 63/2010

3. RESULTS AND DISCUSSIONS

3.1. Morphology and topography of lymph nodes from thoracic and abdominal

3.1.1. Lymph nodes

In pigs, lymph nodes had an apparently different organization from that found in other species. The numerous nodules that made up their composition had a deep location and the medullary was slightly distinct (Fig. 1, Fig. 2). For this reason, their structure seemed inverted, the cortex being deeply organized, surrounded by the medullary, which had become a peripheral part. This particular type of lymph node was considered an inverted lymph node (*Nodus lymphaticus inversus*).

The–grouping of nodules on the inside and medullary sinuses pushes pushed towards the periphery were-observed.

As with other domestic mammals, the capsule is was approached by a series of related lymphatic vessels. They would open into very narrow capsular sinuses under which the lymph nodes are were located. Much more numerous were the afferents that directly penetrated the trabeculae, then followed a rectilinear path to the depth (Yanina et al., 2024). They would open into irregular and relatively wide trabecular sinuses. In the immediate vicinity of these, most of the nodules developed. Their crown was very thick at the opposite pole of the sinus, where it formed a kind of cap (Fig. 2). Grouped around the nodules and in a middle area, there are were clusters of cells that clearly equated to a paracortex. The assembly was integrated into a lax, unorganized connective tissue, where macrophages and plasma cells mixed with lymphocytes (Hsu and Itkin, 2016). In general, there was no distinct hilum, but only an area of emergence of multiple efferent lymphatic vessels. At the level of this area, the peripheral tissue was organized into several medullary cords in the middle of a discrete network of medullary sinuses (Fig. 1).

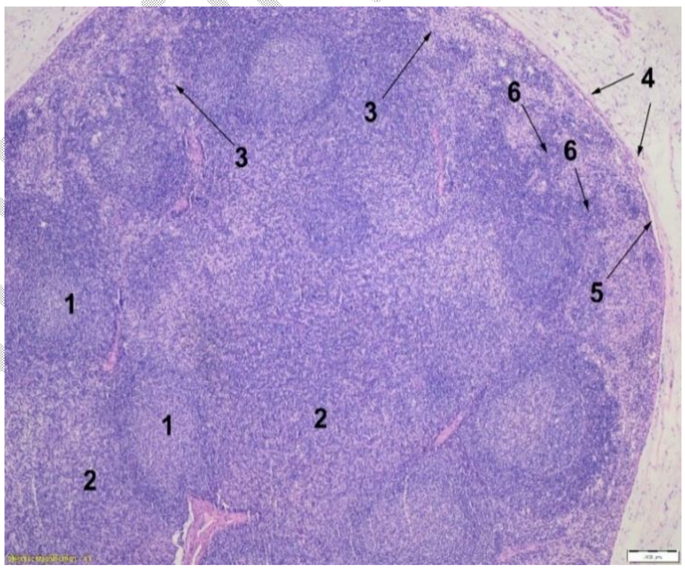

Fig. 1 Section through mesenteric lymph node, ob x4, col. HE (original)
1-lymphoid nodules with evident germinal centers; 2-diffuse lymphoid tissue; 3-medullary sinuses; 4-capsule; 5-subcapsular sinus; 6-medullary cords.

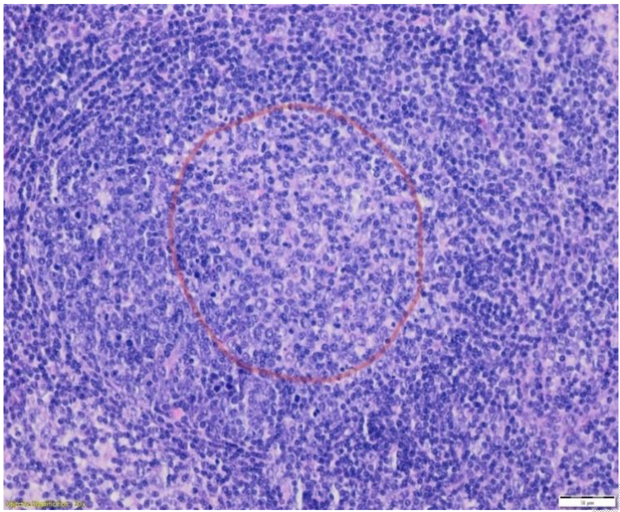

Fig. 2 Section through mesenteric lymph node, ob x20, col. HE (original).
Surrounded by red color is a lymphoid node (white pulp), the cortical area can be seen around it.

We could appreciate that the direction of lymph circulation did not differ in the case of these lymph nodes compared to those of other species. The lymph initially reached the trabecular and subcapsular sinuses, coming into contact with the nodules before diffusing into the parenchyma, medullary sinuses and efferent vessels (Padera et al., 2016).

The blood vessels approached the periphery of the lymph node and accompanied the trabeculae before feeding the capillary network. A few veins left the lymph node along with the efferent lymphatic vessels.

3.1.2. Lymph nodes of the thoracic cavity

In the thoracic cavity there were four lymphocenters:

1. Dorsal thoracic lymphocenter. “The thoraco-aortic lymph nodes were the only representatives of this lymphocenter. From the level of the sixth thoracic vertebra, it occupied the space between the aorta and the spine, up to the level of the last vertebra of this region. Slightly lateral to the aorta, they were somewhat irregularly arranged, both on the left and on the right. Their number was variable, observing 2-10 lymph nodes. Lymph was collected from the dorsal half of the chest wall, basically from all its layers, but also from the mediastinal septum and diaphragm. It also received some efferents of the caudal mediastinal lymph nodes. The efferent vessels, through successive junctions, reached the thoracic canal and some the cranial mediastinal lymph nodes” (Randolph et al., 2017).

2. Ventral thoracic lymphocenter. Only the group of cranial sternal lymph nodes were present, relatively well represented. Their topography was dorsal to the first sternum, between it and the internal thoracic vessels (Fig. 3). “It collects collected lymph from the ventral part of the chest wall, from the pectoral muscles but also from the most anterior part of the ventral abdominal muscles, including the regional breasts. Apart from these structures, lymph was also taken from the mediastinum, diaphragm, thymus, trachea and the thoracic portion of the esophagus. The destination of the efferent vessels was different: the thoracic duct, the right lymphatic duct, the external jugular vein and the brachiocephalic vein. A few efferents were tributary to the caudal deep cervical lymph nodes" (Randolph et al., 2017).

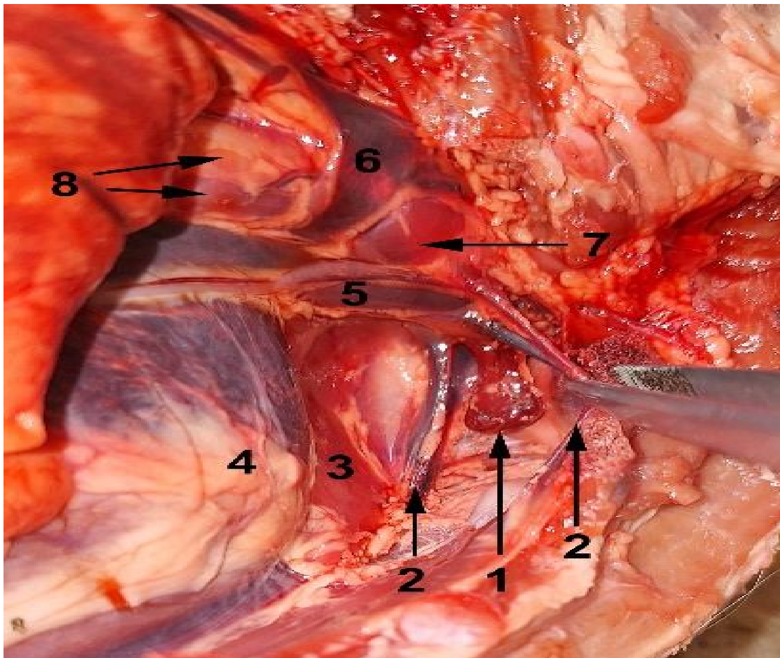

Fig. 3 Topography of lymph nodes in the cranial mediastinum (original)
1 - group of cranial sternal lymph nodes; **2** - internal thoracic arteries and veins; **3** - thymus; **4** - cord covered by pericardium; **5** - cranial vena cava; **6** - costocervical venous trunk; **7** - cranial mediastinal lymph nodes; **8** - cranial tracheobronchial lymph nodes (eparterial).

3. Mediastinal lymphocenter. It included cranial and caudal mediastinal lymph nodes.

Cranial mediastinal lymph nodes had a lower topographic constancy. Even their number was variable (1-10). They accompanied the trachea, the esophagus but also the large vessels especially on the right side. The ones in the anterior part bordered the cranial sternal ones and the most caudal ones were close to the tracheobronchial ones. The areas they served were: the structures in the caudal areas of the neck, those in the cranial parts of the walls of the thoracic cavity, the scapular regions, the cranial mediastinum, the trachea, the esophagus, the thymus and the pericardium. They also received the efferents of the thoraco-aortic and tracheo-bronchial lymph nodes. The efferent vessels had different destinations, those on the right side reached the right lymphatic duct and those on the left side reached the thoracic duct. Some reached the cranial sternal lymph nodes.

Caudal mediastinal lymph nodes were always fewer in number (between one and three). They were associated with the esophagus, immediately behind the tracheobronchial and ventral lymph nodes of the aorta. The afferents originated from the pericardium, the caudal part of the mediastinum and the corresponding part of the esophagus (Ntonas et al.,2020). The efferents connected to the tracheobronchial lymph nodes or could also approach some thoraco-aortic lymph nodes.

4. Bronchial lymphocenter

The right tracheobronchial lymph nodes, up to three in number, were positioned in contact with the trachea, before the origin of the right main bronchus (Fig. 4). They drained the right lung and the terminal portion of the trachea. The efferents reached the left tracheo-bronchial lymph nodes and cranial or cranial mediastinal lymph nodes.

The left tracheobronchial lymph nodes (2-7) were found in contact with the trachea, cranially by the left main bronchus, dorsally masked by the left azygose vein. The afferent vessels came from the left lung but also from the heart, trachea and esophagus and for some to the caudal mediastinal and right tracheobronchial lymph nodes. The efferent vessels reached the thoracic duct or the caudal mediastinal lymph nodes.

The middle tracheobronchial lymph nodes, 2-5 in number, were placed dorsally by the angle of separation of the main bronchi (Fig. 5). The afferent vessels came from the lungs, pericardium, trachea, esophagus and mediastinum (Crisóstomo et al., 2016). The efferent vessels reached the left tracheobronchial and cranial lymphodes as well as the cranial mediastinal lymph nodes.

The cranial tracheobronchial lymph nodes (1-5) were placed ventrocranially from the origin of the tracheal bronchus or slightly cranially from it, at the ventral part of the trachea (Fig. 4, 5 and 6). In specimens in which there was only one lymph node, it was bulky. The afferent vessels came from the lungs, heart and pericardium, but also from other tracheobronchial lymphodes. The efferent vessels were directed to the cranial mediastinal lymph nodes.

3.1.3. Lymph nodes of the abdominal cavity

In the abdominal and pelvic cavities there were five lymphocenters:

1. Lumbar lymphocenter. It was represented by five groups of lymph nodes: lumbo-aortic, renal, fernic-abdominal and testicular.

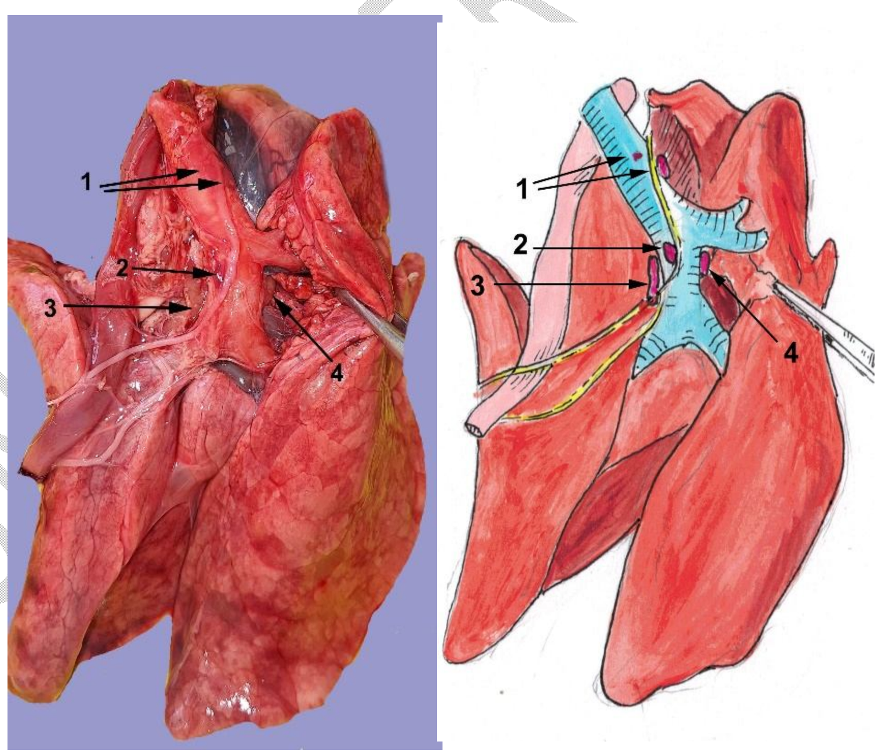

Fig. 4 Topography of lymph nodes belonging to the tracheobronchial lymph center, photograph (A) and schematic representation (B) (original)

1 - cranial tracheobronchial lymph nodes; **2** - intermediate tracheobronchial lymph nodes; **3** - left tracheobronchial lymph nodes; **4** - right tracheobronchial lymph nodes.

The lumbo-aortic lymph nodes formed an elongated group of about 10-15 relatively small formations, placed between the origins of the renal arteries and the terminal portion of the aortic artery, the latter closed to the group of medial iliac lymph nodes. They had relations with the lateral but especially dorsal parts of the aortic artery and vena cava. The afferent vessels drained the dorsal and lateral walls of the abdomen, the peritoneum, the kidneys and adrenal glands, the gonads and the tubular part of the genital tract. These lymph nodes were reached by part of the efferents of the medial and lateral iliac lymph nodes, caudal mesenteric lymphodes and other lymph nodes belonging to the lumbar lymphocenter. The efferent vessels associate associated with each other and finally approached the lumbar trunks or the cistern of the chyle (Yanina et al., 2024).

Renal lymph nodes were variable in number (1-4), placed on the path of the homonymous vessels, between the aorta or caudal vena cava and the kidney on that side (Fig. 7). It performed lymphatic drainage of the kidneys and adrenal glands, but also of the peritoneum and regional muscles. They received the efferents of the phrenic-abdominal lymph node and some lumbo-aortic lymph nodes in the vicinity. The efferent vessels reached the lumbar trunks or the chyle cistern.

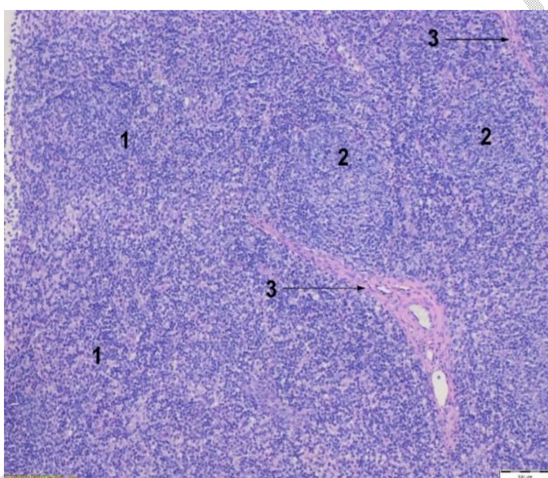

Fig. 5 Section through cranial tracheobronchial lymph node, ob x10, col. HE (original)
1-diffuse lymphoid tissue; 2-lymphoid nodules; 3-conjunctive-muscular trabecula.

The phrenic-abdominal lymph node was reduced, and would be present only unilaterally or even absent. When it existed, it was placed on the path of the caudal branch of the phrenic-abdominal vessels, near the lateral edge of the major psoas muscle. It drained the peritoneum and zonal muscles and could receive some efferents of the lateral iliac lymph nodes. The efferent vessels reached the lumbo-aortic or renal lymph nodes, even directly into the lumbar trunks.

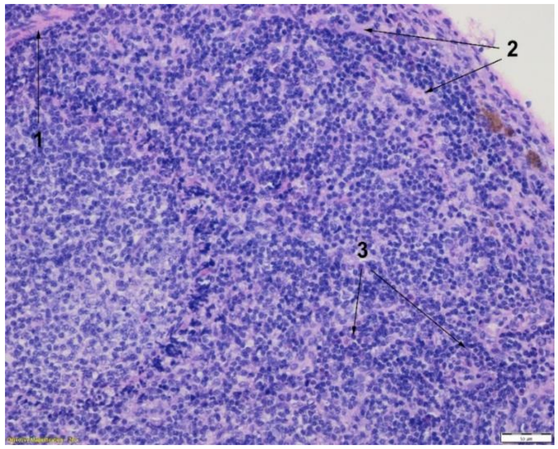

Fig. 6 Section through cranial tracheobronchial lymph node, ob x20, col. HE (original)
1-trabeculae with smooth muscle tissue; 2-medullary sinuses; 3-diffuse lymphoid tissue.

"The testicular lymph node was located subperitoneally, on the path of the testicular vessels. Sometimes it occurred near the origin of these vessels, but most commonly in the vicinity of the inguinal region. There were cases in which it appeared in duplicate but also cases in which it could be missing. It drained the testicle and epididymis and sent the efferents to the lumbo-aortic lymph nodes" (König, 2014).

Uterine lymph nodes, classified by some authors as belonging to the iloosacral lymphocenter, by their topography and the relationships they have with the lumbo-aortic lymph nodes, could rather be considered as belonging to the group in which the latter fall. There were one or two lymph nodes, which could be bilaterally or unilaterally absent.

It drained the ovary, fallopian tubes and the corresponding horn of the uterus. The efferent vessels were destined for the lumbo-aortic or medial iliac lymph nodes.

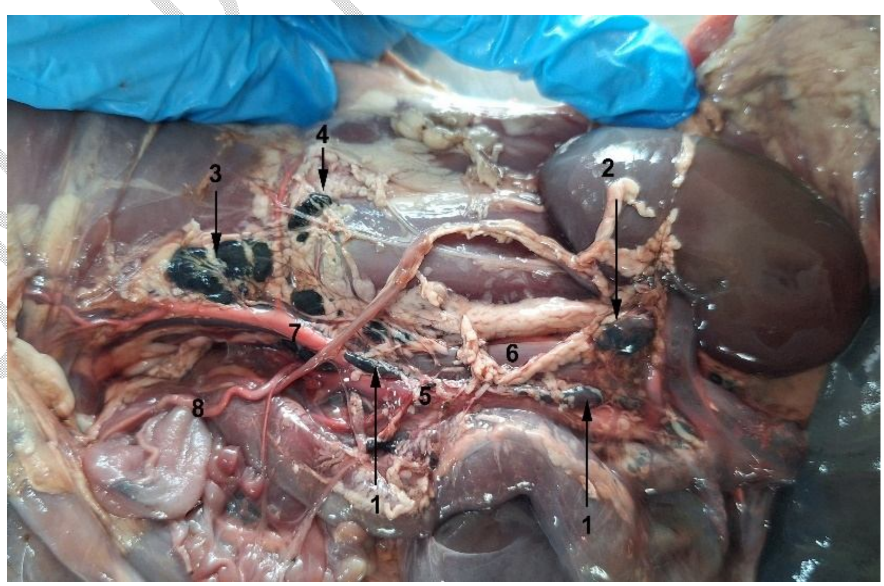

Fig. 7 Topography of lymph nodes on the roof of the abdominal cavity in pig (original)
1-lumbo-aortic lymph nodes; 2-renal lymph nodes; 3- medial iliac lymph nodes; 4-lateral iliac lymph nodes, 5-abdominal aorta; 6-caudal cava vein; 7-external iliac artery; 8-umbilical artery.

3.1.4. Celiac lymphocenter

Celiac lymph nodes (3-4) were placed around the celiac artery, often difficult to isolate from splenic or gastric lymph nodes. They received afferent vessels from the lung, mediastinum, diaphragm, liver, spleen, adrenal glands, adjacent muscles, as well as the efferents of all the other lymph nodes belonging to this lymphocenter. The efferent vessels came together to form the celiac lymphatic trunk (Suami și Scaglioni 2017).

The hepatic (portal) lymph nodes (2-7) were located along the portal vein and in the area of the hepatic hilum (Fig. 9). The afferent vessels came from the liver, gallbladder and pancreas. The efferents of the pancreaticoduodenal lymph nodes were also collected. The efferent vessels reached the celiac lymph nodes or the celiac trunk.

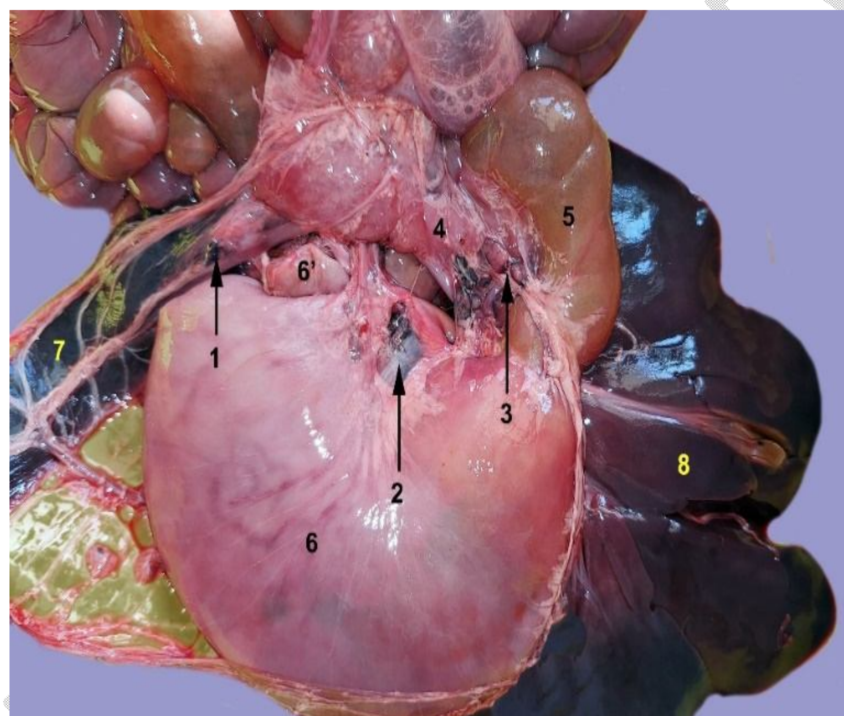

Fig. 8 Topography of the lymph nodes belonging to the celiac lymph center (original)

1- splenic lymph nodes; **2** - gastric lymph nodes; **3** - pancreaticoduodenal lymph nodes; **4** - pancreas; **5** - duodenum; **6** - stomach; **6'** - ventricular diverticulum; **7** - spleen; **8** - liver

"Splenic lymph nodes were placed on the path of the homonymous vessels. Their topography was between the aorta and the hilum of the spleen (Fig. 8). In this sector there could be 2-4 lymph nodes, but there are were others (2-5) located along the upper quarter of the spleen. They received the efferent vessels from the spleen, from the great epiplon, from the left portions of the stomach and pancreas. The efferent vessels are were directed to the celiac lymph nodes or the celiac trunk" (Ntonas et al.,2020).

Gastric lymph nodes, which vary in number (1-5), were placed especially in the first part of the left gastric artery to near the cardia. It received the efferent vessels of the mediastinum, the caudal portion of the esophagus, the stomach and pancreas. The efferent vessels were directed to the celiac lymph nodes or the celiac trunk.

The more numerous pancreatico-duodenal lymph nodes (5-10) were located along the cranial pancreatico-duodenal artery, between the descending portion of the duodenum and the right lobe of the pancreas. They received related vessels from these organs, from the right side of the stomach and the great epiplon (Dove and Alworth, 2015). The efferent vessels approached the celiac lymph nodes and some could reach the colic lymph nodes.

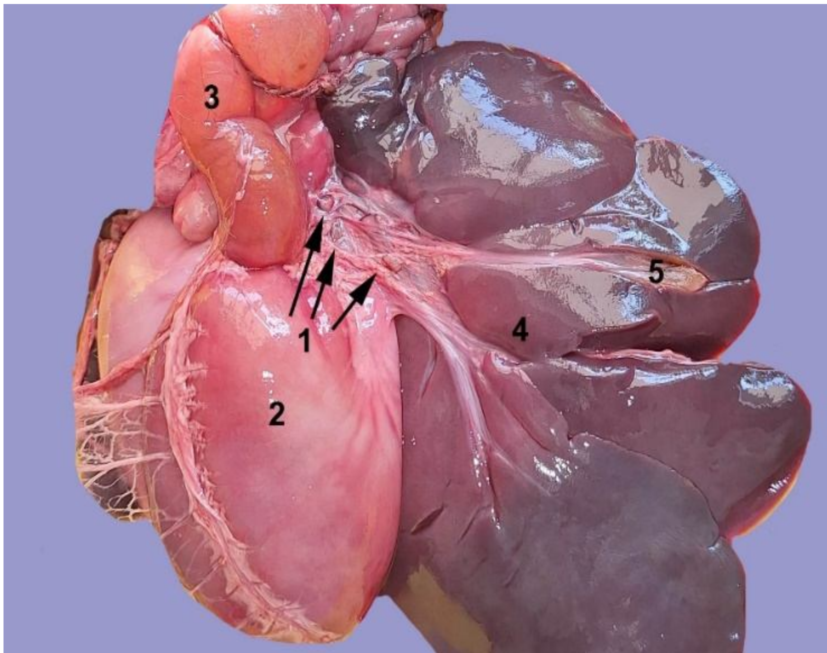

Fig. 9 Portal lymph node topography (original)
1 - portal lymph nodes; **2** - stomach; **3** - duodenum; **4** - liver; **5** - gallbladder

3.1.5. Cranial mesenteric lymphocenter

Cranial mesenteric lymph nodes were located around the origin of the cranial mesenteric artery. They could be confused with some of the pancreatico-duodenal and celiac lymph nodes. It received the afferent vessels from the colon and some efferent vessels from the jejunal and colic lymph nodes. The efferent vessels reached the visceral trunk and intestinal trunk. From the capillaries, the interstitial fluid is transported to the lymphatic precollectors that have a diameter of 70-150 µm (Suami și Scaglioni 2018).

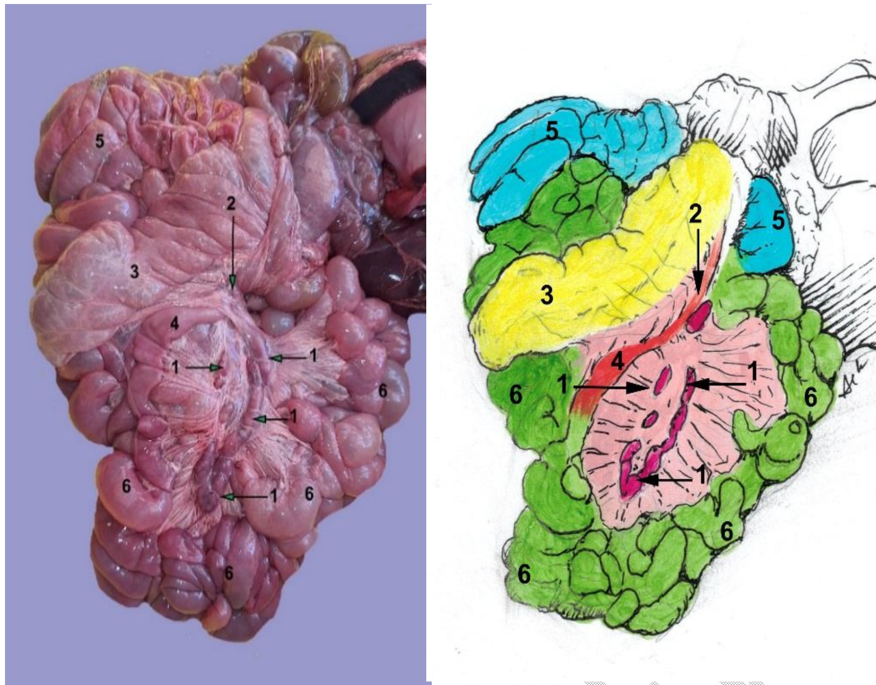

Fig. 10 Topography of lymph nodes belonging to the cranial mesenteric lymph center, photograph (A) and schematic representation (B) (original)

1 - jejunal lymph nodes; **2** - ileocecal lymph nodes; **3** - cecum; **4** - ileum; **5** - ascending colon; **6** - jejunal loops

The jejunal lymph nodes formed a double chain in the middle of the jejuno-ileum meso, about 30 cm long (Fig. 10, Fig. 11). The two portions of this chain, each in contact with the peritoneum of one side of the meso, were separated by a layer of adipose tissue, crossed by the jejunal vessels (Ntonas et al.,2020). In the area corresponding to the ileum, the jejunal lymph nodes were more dispersed.

"The afferent vessels drained the ascending portion of the duodenum, the jejunum and the ileum. The efferent vessels converged in the mesum to form the roots of the intestinal triuncle. The vessels that came from the lymph nodes close to the duodenum reached this trunk directly. On the other hand, those close to the ileum addressed the ileocolic lymph nodes" (Yanina et al., 2024).

The ileocolic lymph nodes (5-10) formed a group less well defined by the jejunal and colic lymph nodes in the vicinity. They were arranged on either side of the terminal portion of the ileum, both in the ileonic mesoum and in the ileocecal fold. The afferent vessels came from the cecum, the ileum and the terminal portion of the jejunum (Suami și Scaglioni 2018). Some came from the last jejunal lymph nodes. The efferent vessels reached the intestinal trunk.

Fig. 11 The mesenteric ganglion group - (B mode - longitudinal section) - It is identified as ovoid structures (arrows), with reduced echogenicity (inferior to intestinal loops and similar to regional vascular structures), the contour is regular, the echostructure is homogeneous

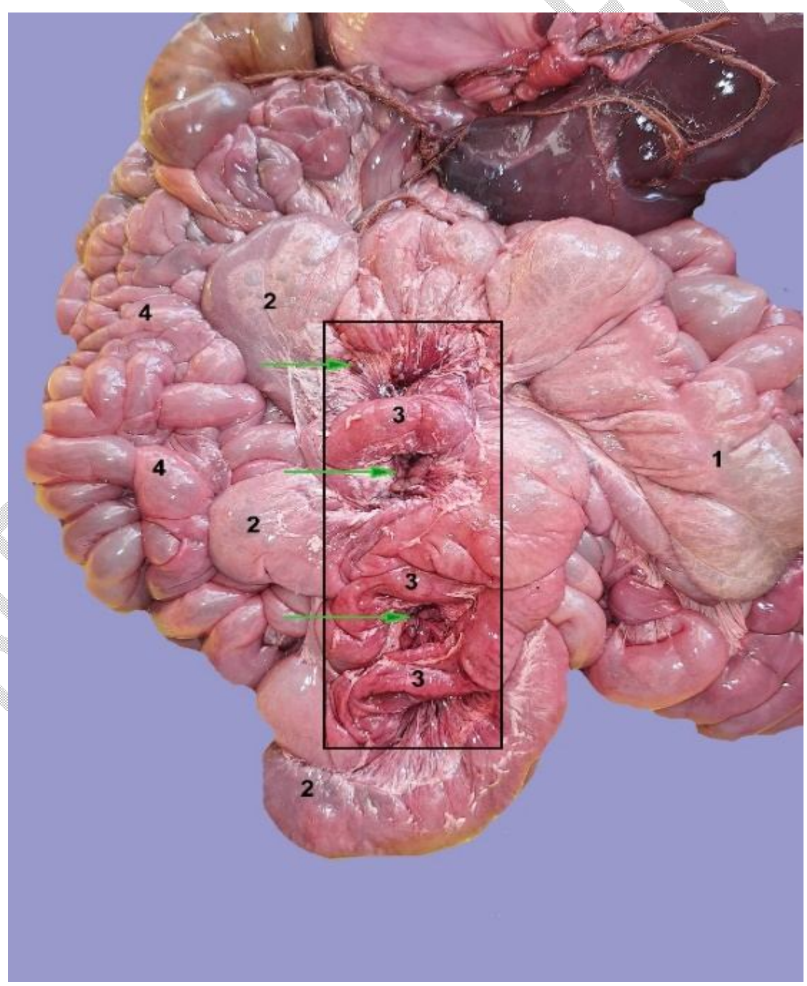

Fig. 12 Colic lymph nodes arranged in the connective axis of the helical colon indicated by green arrows (original)

1- caecum; **2 -** centripetal turns of the helical colon; **3 -** centrifugal turns of the helical colon; **4 -** jejunal convolutions.

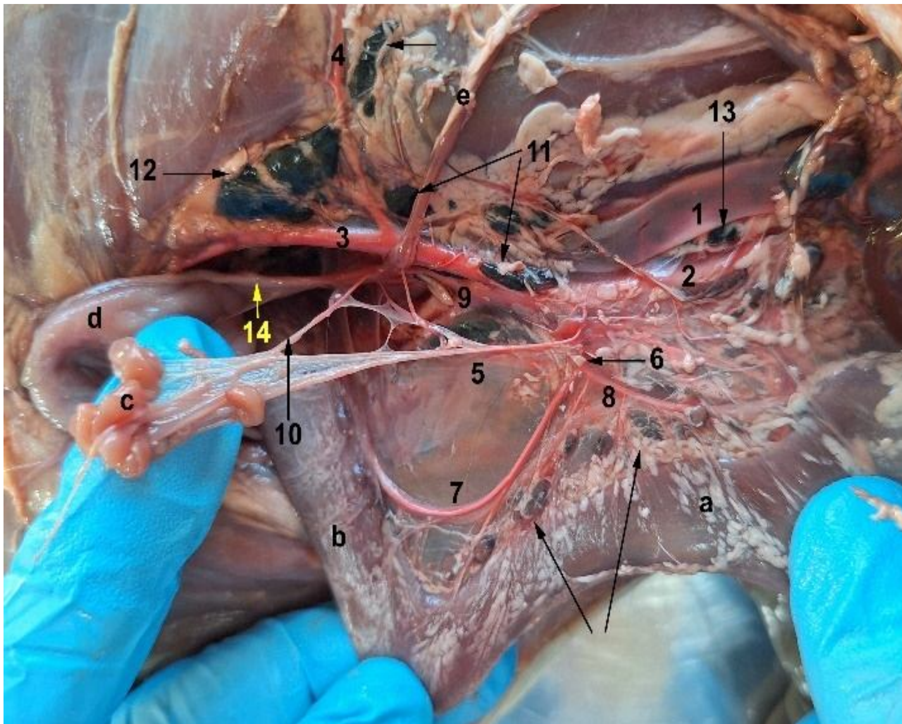

Fig. 13 Caudal mesenteric artery and lymph nodes in the pelvic cavity in pigs (original)
1-caudal vena cava; 2-abdominal aorta; 3-external iliac artery; 4-deep iliac circumflex artery; 5-descending mesocolon (small mesentery); 6-caudal mesenteric artery; 7-cranial rectal artery; 8-left colic artery; 9-the origin of the internal iliac artery; 10-uterine artery; 11-medial lymph nodes; 12-ileofemoral lymph nodes; 13-lumbo-aortic lymph nodes; 14-umbilical artery; a descending colon; b-rect; c-uterine horn; d-bladder, e-ureter.

Colic lymph nodes were very numerous, and could even reach 50. It formed a chain associated with the right colic artery in the thickness of the connective axis that supported the coils of the ascending colon (Fig. 12). "The afferent vessels drained the adjacent portion of the cecum, the ascending colon and the transverse colon. Some efferents of the pancreaticoduodenal lymph nodes could also reach the colic ones" (Done et al., 2006). The efferent vessels formed the roots of the colic trunk; some reached directly into the intestinal trunk.

Caudal mesenteric lymphocenter. The caudal mesenteric lymph nodes form formed a not very compact chain of about ten nodular structures, which accompanied the descending colon.

Ileosacral lymphocenter. This lymphocenter was represented by the medial iliac, lateral iliac, sacral and anorectal lymph nodes (Fig. 7, Fig. 13).

The **medial iliac lymph nodes** were a group of lymph nodes located at the origin of the deep iliac circumflex artery. At this level, three or four lymph nodes have been observed in 80% of cases, the most voluminous not exceeding 1.2 mm. The more rostral ones were not well delimited by the lumbo-aortic lymphodes (Lelovas et al., 2014).

The **lateral iliac lymph nodes**, generally two in number, were found at the cranial edge of the terminal bifurcation of the deep iliac circumflex artery, in the superitoneal adipose tissue organized on the lateral side of the psoas major muscle. In 10% of cases, they could be absent unilaterally or bilaterally.

The **sacral lymph nodes** were located at the origin of the median sacral artery, between the initial portions of the two internal iliac arteries. At this level, 2-3 lymph nodes have been observed, but on the path of the first half of the median sacral artery, accessory lymph nodular clusters were frequently found (75% of cases), (Lelovas et al., 2014).

The **anorectal lymph nodes**, numerically variable (2-10 units), were located on the dorsal side of the retroperitoneal portion of the rectum. In about 10% of cases they ~~are~~ were not present.

Ileofemoral lymphocenter. In pigs, this lymphocenter was represented only by **ileofemoral** lymph nodes. They were positioned along the external iliac vessels, from the origin of the deep iliac circumflex artery to the vicinity of the deep inguinal ring. In general, there were 3-4 lymph nodes, but less often (about 10-15% of cases, only one large lymph node appeared).

5. CONCLUSIONS

Although variable in number, thoraco-aortic lymph nodes were constant, but intercostal lymph nodes have been identified in any case. Similarly, the cranial sternal lymphocenter was represented is represented by constant cranial lymph nodes and the caudal ones are absent.

In the case of mediastinal lymph nodes, the anterior ones were present, but their topography differed significantly from individual to individual. Caudal mediastinal lymph nodes were reduced and we considered their value in the anatomo-pathological examination to be insignificant.

The most important groups of visceral lymph nodes were jejunal and colic. It can be mentioned that the jejunal ones could be examined relatively easily by the ultrasound method. On the other hand, the colic ones were masked in the thickness of the mesocolon root and surrounded by the coils of the ascending colon, which is why they were difficult to spot by ultrasound.

The most important parietal lymphocenter was the iliosacral one; in it, we were able to describe for the first time some individual variants, namely: lateral iliac lymph nodes dominant in volume compared to the medial ones, the absence in some cases (10%) of anorectal lymph nodes in some specimens, in which, however, a compensation was observed by the development of a chain of small units on the path of the median sacral artery.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

ETHICAL APPROVAL

Our experiment was carried out in the anatomy laboratory of the Faculty of Veterinary Medicine in Bucharest, benefiting from the favorable approval of the Ethics Commission of this institution and met the conditions required by EU Directive 63/2010. As per international standards or university standards written ethical approval has been collected and preserved by the author(s).

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