Chapter 4

Importance of the Arterial Blood Supply to the Rabbit and Guinea Pig Spinal Cord in Experimental Ischemia

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Additional information is available at the end of the chapter

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Abstract

Spinal cord ischemia belongs to the one of the most frequently occurring results of spinal cord damage, with broad range of several symptoms and complications. The superficial position of fine arterial system of the spinal cord predicts the spinal cord ischemic injury. The laboratory animals, such as rabbits and guinea pigs, serve for the study of spinal cord ischemic injury. The aim of this work was to describe the arterial blood supply to the spinal cord in New Zealand White rabbits and English self guinea pigs, using the corrosion and dissecting technique. In both species, we found variations in arrangement and origin of segmental arteries of descending aorta, the basilar artery, the ventral spinal artery, the dorsal spinal arteries, the artery of Adamkiewicz, and the segmental dorsal and ventral branches arising from the arterial spinal branches. The presence of the artery of Adamkiewicz and nearly regular segmental blood supply to the spinal cord are responsible for the use of rabbit and guinea pig as a simple model of ischemic damage to the spinal cord. The understanding of the arterial arrangement to the spinal cord plays a very important role in avoiding the spinal cord ischemia or infarction during surgical interventions to the spine.

Keywords: artery, experiment, guinea pig, rabbit, spinal cord injury

1. Introduction

1.1. Spinal cord injury

Spinal cord injury represents a significant health problem associated with lifelong disability and a broad range of secondary complications. Although spinal cord trauma causes loss of neuronal cell bodies as well as myelinated axons, the dysfunction of the white matter tracts is

the factor that determines most of the clinical symptoms. In addition, the management of spinal cord injury patients is challenging. According to the damaged part of the spinal cord, the spinal cord injury can lead to the respiratory insufficiency due to the paralysis of breathing muscles, necessitating mechanical ventilators, phrenic nerve pacing, loss of sensory and motor functions, recurrent kidney stones, urinary tract infection, pressure sores, and cardiac and respiratory dysfunction. In the majority of cases, the spinal cord injury results into the spinal cord ischemia due to the superficial position of fine arterial system of the spinal cord.

1.2. Laboratory animals in experimental spinal cord injury

The advances made so far with the benefit of animal model have been primarily in understanding the cell biology of the injured nervous system. Rodent models are the common type of mammal employed in experimental spinal cord injury studies, and widespread research have been conducted using rats, mice, gerbils, guinea pigs, and hamsters [1]. Other animal experiments include cats, rabbits, and dogs [1, 2]. Of course, larger mammals such as nonhuman primate, goats, and pigs are also used but very rarely and are less experienced models based in spinal cord injury research, requiring expensive aftercare and housing as well as stringent ethical considerations [1–4].

1.2.1. Rabbit

Rabbits are commonly used in biomedical research. Currently, many strains of rabbit are available. Laboratory rabbits belong to the order Lagomorpha and are collectively referred to as lagomorphs [5]. The most popular strain for research purposes is a medium-sized (weighing between 3 and 5 kg), New Zealand White outbred rabbit. A number of advantages make rabbits a widely used animal in biomedical research. Their size, ease of handling, and relative ease of blood collection due to their large ear vessels make them suitable for many types of experiments [6]. The rabbits are suitable for long-term experiments, because most of them live for 5–8 years, but some individuals live to the age of 10 years or more.

1.2.2. Guinea pig

Guinea pigs occupy special place in research. This rodent species, with its unique physiology and anatomy, has come to symbolize all experimental subjects. The special place in research implies that guinea pigs are one of the most commonly used laboratory animals in research. They represent an appropriate animal for several types of experiments because of their small size, cleanliness, docileness, and relatively easy maintenance [7].

1.2.3. Rabbit and guinea pig in experimental spinal cord injury

The two before-mentioned species have been used as experimental models in the study of spinal cord ischemic injury, and the effect of various neuroprotective drugs on such way altered the nervous tissue [8–13]. The more detailed knowledge of anatomy of the spinal cord blood supply with focus on all possible variations can contribute to the protection of the spinal cord. The aim of our study was to describe the arterial blood supply to rabbit and guinea pig spinal

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cord using the corrosion and dissecting technique. We described some variations of the principal arteries and the segmental arteries contributing to the arterial blood supply in the corresponding region.

2. Materials and methods

2.1. Experimental animals

2.1.1. Rabbit

Adult New Zealand White rabbits in number of 20, at 140 days of age (weight range 2.5–3 kg) consisting of 10 males and 10 females, were used in this study. The work was performed in an accredited experimental laboratory at the University of Veterinary Medicine and Pharmacy in Košice, Slovak Republic. Standard conditions were ensured to all animals: approved cages, relative humidity (45%), temperature (15–20°C), light period (12 hours), feed (granular mixed, KLASIK, de Heus, Bučovice, Czech Republic), and drinking water (ad libitum). The corrosion technique was used for 10 rabbits, females (n = 5) and males (n = 5), and the dissecting technique for 10 rabbits, females (n = 5) and males (n = 5).

2.1.2. Guinea pig

Adult English self guinea pigs in number of twenty, at 220 days of age (weight range 0.8–1 kg) consisting of 10 males and 10 females, were used in this study. The work was performed in an accredited experimental laboratory at the University of Veterinary Medicine and Pharmacy in Košice, Slovak Republic. Standard conditions were ensured to all animals: approved cages, relative humidity (45%), temperature (15–20°C), light period (12 hours), feed (FANTASIA, Tatrapet, Liptovský Mikuláš, Slovak Republic), and drinking water (ad libitum). The corrosion technique was used for 10 guinea pigs, females (n = 5) and males (n = 5), and the dissecting technique for 10 guinea pigs, females (n = 5) and males (n = 5).

2.2. Casting media

2.2.1. Corrosion technique

In the corrosion technique, Spofacryl (SpofaDental, Czech Republic) was used as a casting medium. It consists of a powdered component (copolymer of methyl methacrylate, copolymer of methacrylate, sodium p-toluenesulfinate, pigments, and fluorescent agent), liquid component (methyl methacrylate, methacrylic acid, ethylene glycol dimethacrylate, stabilizers, and amine), and red pigment (1,2-benzenedicarboxylic acid, bis[2-ethylhexyl ester], epoxidized soybean oil, and 2-naphthalenecarboxylic acid).

2.2.2. Dissecting technique

Batson's No. 17 Plastic Replica and Corrosion Kit (Polysciences Europe GmbH, Germany) was used as a casting medium in the dissecting technique. It consists of base solution A (polymethyl methacrylate; methyl methacrylate; dibutyl phthalate; 2-methyl-, 1,2-ethanediyl ester; and 2-propenoic acid), catalyst (dibutyl phthalate, benzoyl peroxide, and acetone), promoter C (N,N-dimethyl-4-toluidine and dibutyl phthalate), and red pigment (2-naphthalenecarboxylic acid, 1,2-benzenedicarboxylic acid, epoxidized soybean oil, and bis[2-ethylhexyl ester]).

2.3. Methods

2.3.1. Surgical preparation of animals

First step in surgical preparation of animals was the intravenous application of heparin (50,000 UI/kg) 30 minutes before the animals were euthanized using the embutramide (T-61, 0.3 mL/kg) also intravenously. For better manipulation during the dissection and prevention from the hair sticking to the corrosive casts during maceration process was the skin subsequently as far as possibly removed. The entrance into the thoracic cavity was performed from the left side by removing of the parts of the ribs. Before the introduction of a ligature to the initial part of ascending aorta, the pericardial cavity was opened. Plastic cannula was inserted into the ascending aorta through the opened left ventricle. After the cannula was fixed in the ascending aorta, the perfusion started. The decrease of pressure in arteries and veins and performing of good injection were accomplished by opening of the right auricle of the heart. The manual perfusion of the arterial and venous system using the cannula by means of 2.5–3 l of warm (37°C), pH 7.3, and 0.9% NaOH in 0.01 M phosphate took 15–20 min [14].

2.3.2. Casting medium preparation

2.3.2.1. Corrosion technique

Powdered component with a weight of 20 g was added to the red pigment. To this mixture, liquid component in amount of 10 mL was added, and both components were mixed together.

2.3.2.2. Dissecting technique

The red pigment was added to the base solution A prior to mixing the catalyst and promoter C. The pigment was added in the amount of 5% of the base solution A. After mixing of the base solution A and the pigment together, the mixture was divided into two similar parts. Each of them has a volume of 10 mL. The first of these parts was mixed with the catalyst in volume of 6 mL. The second part was mixed with six drops of promoter C. After the initial mixing, these two parts were fused and mixed together.

2.3.3. Casting medium application

The same cannula fixed to the ascending aorta serves for manual filling of the arterial system with the casting medium. The red casting medium in the superficial body vessels determined

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the adequate filling of arteries and an even distribution. After the completion of arterial casting, for at least 30 minutes, the bodies must not be manipulated. After this time period, the bodies were submersed in water (40–60°C; 24 hours) to ensure adequate polymerization of the applied casting medium in the arterial system.

2.3.4. Corrosion technique

The variable soft tissues, which surrounded the polymerized casting medium, were dissolved by the potassium hydroxide (KOH) in concentration of 2–4% for 2 days. For the faster corrosion, a constant temperature (40°C) of the used solution must be achieved [15]. Every 12 hours, the corrosion solution was changed. After the dissolution, the rest of the surrounding soft tissues were removed from the corrosion casts in running water. Then, the corrosion casts were dried at the room temperature.

2.3.5. Dissecting technique

By the dissecting technique, 10% formaldehyde was injected into the vertebral canal between the last lumbar vertebra and sacrum, between the last cervical and first thoracic vertebra, and between the occipital bone and the first cervical vertebra to fix the spinal cord. After 1-week fixation, the vertebral canal was opened by removing vertebral arches in sacral, lumbar, thoracic, and cervical regions. Also, the occipital bone was partly removed. The prepared spinal cords were fixed in 10% formaldehyde (Section 1).

3. Results

3.1. Rabbit

3.1.1. Cervical spinal cord

In the cervical spinal cord, we found more complex arterial blood supply in comparison with the rest of the spinal cord. The most cranial section of cervical spinal cord was supplied with blood by means of small branches arising from the posterior inferior cerebellar artery. This artery originated from the vertebral artery bilaterally. The bilateral vertebral arteries entered into the vertebral canal through the lateral vertebral opening of the atlas. The fusion of the bilateral vertebral arteries was located on the caudal border of the dorsal surface of the basilar part of the occipital bone. From this fusion, the basilar artery which participated on the formation of arterial cerebral ring continued cranially. The fusion of vertebral arteries was present in 50% of cases without gap (**Figure 1**) and in 30% of cases with one longitudinal gap (**Figure 2**). In 20% of cases, we found two gaps. At the level of fusion of bilateral vertebral arteries originated the ventral spinal artery. Its origin was from the right-sided vertebral artery, from the left-sided vertebral artery (**Figure 2**), and from the anastomosis of two branches, each coming from the medial surface of the corresponding vertebral artery (**Figure 1**). The frequency of origins of ventral spinal artery is shown in **Table 1**.

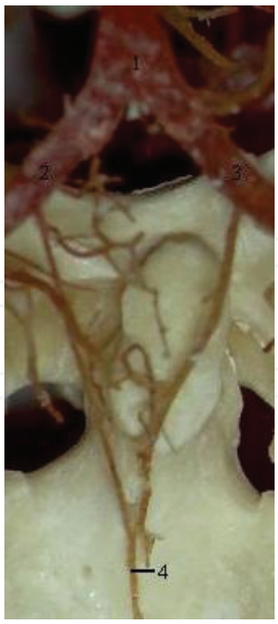

Figure 1. The origin of the ventral spinal artery from the anastomosis of two branches, each coming from the medial surface of the corresponding vertebral artery. (1) Basilar artery, (2) left vertebral artery, (3) right vertebral artery, and (4) ventral spinal artery. Macerated specimen, dorsal view, magnification 8x.

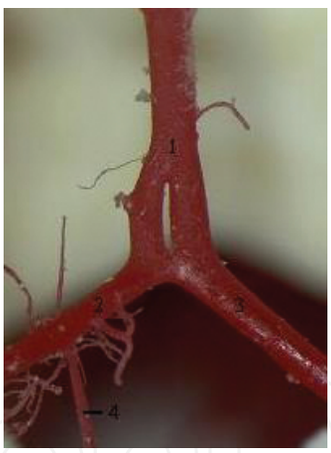

Figure 2. The origin of the ventral spinal artery from the left-sided vertebral artery. The fusion of bilateral vertebral arteries is visible on one longitudinal gap. (1) Basilar artery, (2) left vertebral artery, (3) right vertebral artery, and (4) ventral spinal artery. Macerated specimen, dorsal view, magnification 12.5x.

	Corrosion technique (%)	Dissecting technique (%)	Average (%)
Bilateral origin	20	30	25
Right-sided origin	40	40	40
Left-sided origin	40	30	35

Table 1. Origin of ventral spinal artery using the corrosion technique (10 rabbits) and dissecting technique (10 rabbits).

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	Occurrence of spinal branches (%)	
	Right	**Left**
C 1	0	0
C 2	70	50
C 3	50	30
C 4	50	50
C 5	30	50
C 6	20	70
C 7	30	50
C 8	50	50

C, cervical segment of the spinal cord.

Table 2. Frequency of occurrence of spinal branches in the cervical spinal cord using the dissecting technique (10 rabbits).

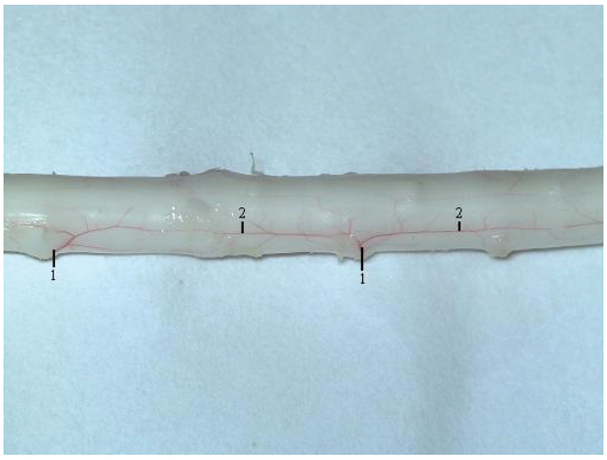

Figure 3. Dorsal branches of the spinal branches joining the irregular dorsal spinal arteries. (1) Dorsal branch of spinal branch and (2) irregular dorsal spinal artery. Dissected specimen, dorsolateral view, magnification 5x.

The ventral spinal artery was located along the ventral median fissure on the ventral surface of the cervical spinal cord. Bilateral vertebral arteries gave off spinal branches which entered the vertebral canal through the intervertebral openings. Inside the vertebral canal, the spinal branches divided into the dorsal and ventral branch with direction to the spinal cord. The ventral branches joined the ventral spinal artery. The frequency of occurrence of individual ventral branches joining the ventral spinal artery is shown in **Table 2**. In the cervical spinal cord, the ventral branches joining the ventral spinal artery were present as right-sided in 46.2% and as left-sided in 53.8% of cases.

We found two irregular longitudinal dorsal spinal arteries receiving dorsal branches of spinal arteries (**Figure 3**) or the absence of the dorsal spinal arteries (**Figure 4**) on the dorsal surface of the spinal cord. In the case of the presence of two longitudinal dorsal spinal arteries, their

arrangement was very variable. These two longitudinal dorsal spinal arteries were formed only by fusion of the small cranially and caudally directed branches originating from the dorsal branches. In the case of the absence of dorsal spinal arteries, the dorsal surface of the spinal cord was supplied by means of dorsal branches forming irregular loops between each other on the same and on the opposite side. There was no origin of the dorsal spinal arteries present in the area of fusion of bilateral vertebral arteries. The frequency of occurrence of dorsal branches was the same as of the ventral branches.

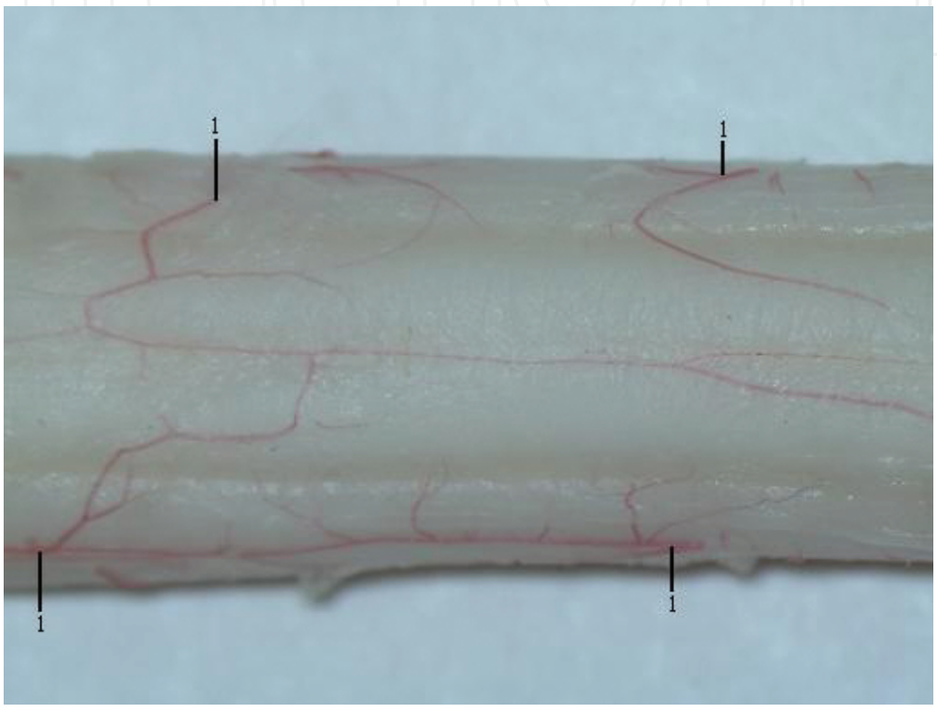

Figure 4. Dorsal branches of the spinal branches in the form of irregular loops. (1) Dorsal branch of spinal branch. Dissected specimen, dorsal view, magnification 12.5x.

3.1.2. Thoracolumbar spinal cord

The thoracic spinal cord received the arterial blood by means of spinal branches originating from the dorsal intercostal arteries which were present in 13 pairs. Dorsal intercostal arteries as paired branches arising from the dorsal surface of the thoracic aorta were present in nine pairs in 70% of cases, in eight pairs in 20% of cases, and in 10 pairs in 10% of cases. The remaining three to five pairs originated from the supreme intercostal artery. The more cranial origin of the left-sided dorsal intercostal arteries than the right-sided was present in 60% of cases (**Figure 5**). The origin of right- and left-sided dorsal intercostal arteries at the same level was present in 20% of cases (**Figure 6**). The origin of first nine pairs at the same level and the more cranial origin of the left-sided arteries than the right-sided by the remaining pairs were present in 10% of cases. The more cranially located origin of the right-sided arteries than the left-sided arteries by the first eight pairs and the origin of the remaining pairs at the same level were found in 10% of cases.

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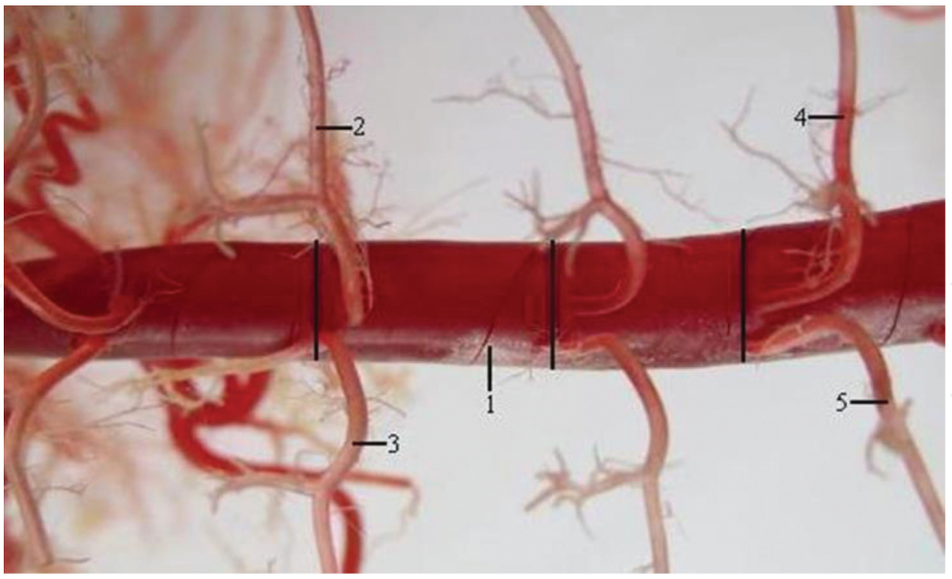

Figure 5. More cranially located origin of the left-sided dorsal intercostal arteries than the right-sided. (1) Thoracic aorta, (2) 10th left dorsal intercostal artery, (3) 10th right dorsal intercostal artery, (4) eighth left dorsal intercostal artery, and (5) eighth right dorsal intercostal artery. The black line indicates the shift of origin of dorsal intercostal arteries. Macerated specimen, dorsal view, macroscopic image.

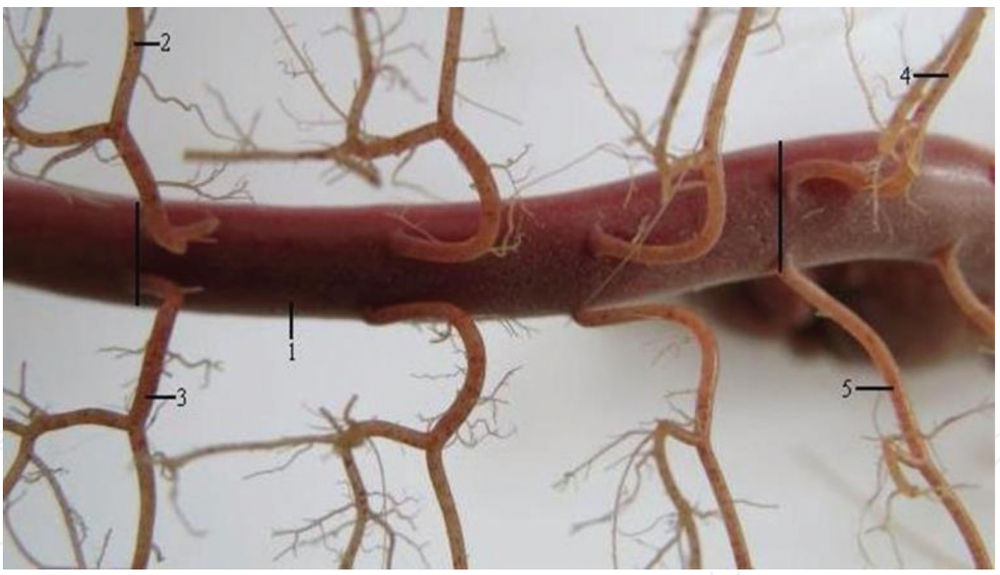

Figure 6. Origin of dorsal intercostal arteries at the same level. (1) Thoracic aorta, (2) 10th left dorsal intercostal artery, (3) 10th right dorsal intercostal artery, (4) seventh left dorsal intercostal artery, and (5) seventh right dorsal intercostal artery. The black line indicates the place of origin of dorsal intercostal arteries. Macerated specimen, dorsal view, macroscopic image.

The paired lumbar arteries originated from the dorsal surface of the abdominal aorta. Their spinal branches represent the arterial blood supply to the lumbar spinal cord. Lumbar arteries in number of six pairs were present in 90% of cases and in five pairs in 10% of cases. The remaining last pair was originating from the median sacral artery. In 60% of cases, the lumbar

arteries at the same level originated by means of a common trunk (**Figure 7**). The independent origin of first two pairs and the origin of the remaining pairs by means of a common trunk were present in 30% of cases (**Figure 8**). The more cranial origin of the left-sided lumbar arteries than the right-sided lumbar arteries by the first two pairs and the origin of lumbar arteries by the remaining pairs by means of a common trunk from the dorsal surface of the abdominal aorta were present in 10% of cases.

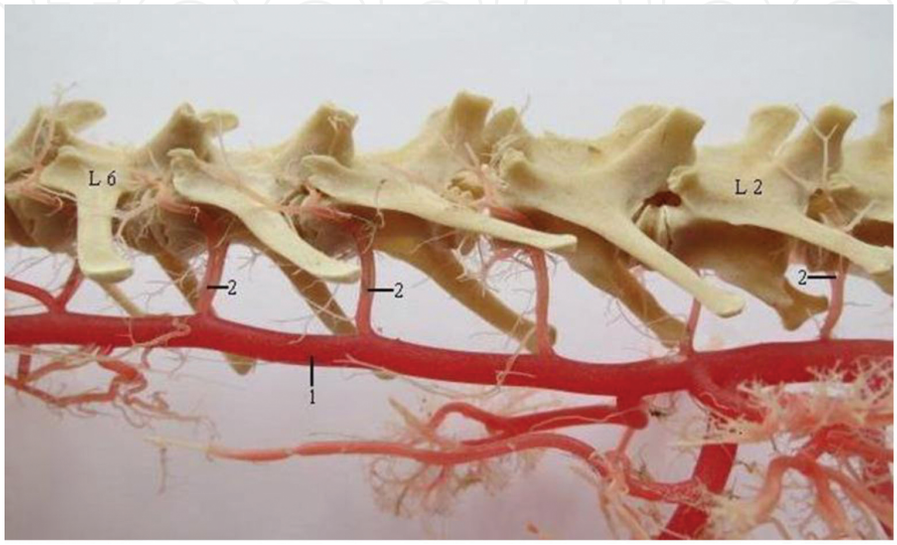

Figure 7. Origin of lumbar arteries by means of a common trunk. (1) Abdominal aorta and (2) the point of division of common trunk for bilateral lumbar arteries. L 2—second lumbar vertebra and L 6—sixth lumbar vertebra. Macerated specimen, lateral view, macroscopic image.

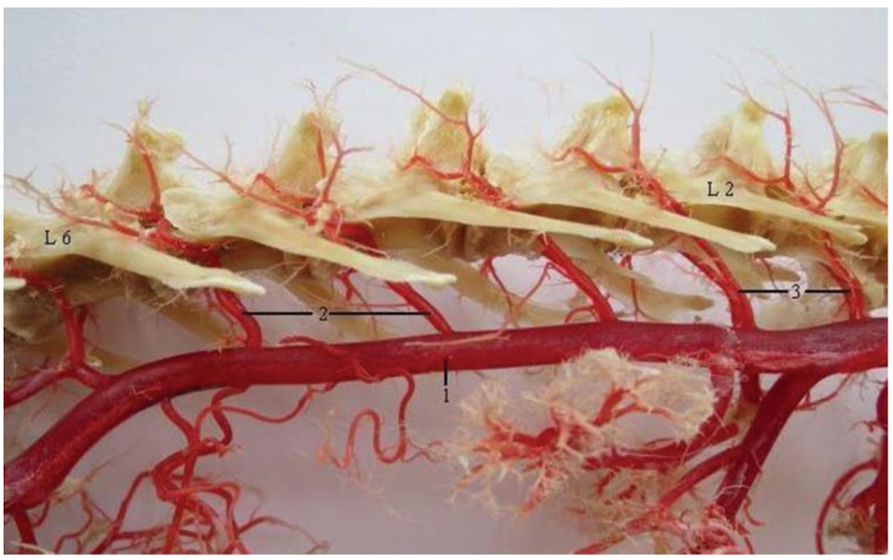

Figure 8. Origin of lumbar arteries by means of a common trunk. The first two pairs originated as independent branches. (1) Abdominal aorta, (2) common trunk for bilateral lumbar arteries, and (3) the separate origin of bilateral lumbar arteries. Macerated specimen, lateral view, macroscopic image.

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The spinal branches with origin from the dorsal intercostal and lumbar arteries entered the vertebral canal through the intervertebral openings in association with the respective spinal nerve roots. Inside the vertebral canal, each spinal branch was divided into the dorsal and ventral branches. The ventral branches joined the ventral spinal artery.

Level	Occurrence of spinal branches (%)			
	Ventral		Dorsal	
	Right	Left	Right	Left
Th 1	20	50	40	50
Th 2	60	70	70	40
Th 3	0	30	50	70
Th 4	30	60	70	90
Th 5	0	90	40	100
Th 6	20	100	30	60
Th 7	70	70	100	60
Th 8	20	100	60	100
Th 9	10	30	0	70
Th 10	20	80	60	20
Th 11	30	30	50	80
Th 12	10	40	0	80
Th 13	40	60	30	100
L 1	50	50	50	40
L 2	30	20	40	80
L 3	50	60	60	60
L 4	70	80	30	40
L 5	50	70	60	50
L 6	50	50	50	50

L, lumbar segment of the spinal cord and Th, thoracic segment of the spinal cord.

Table 3. Occurrence of ventral and dorsal branches of arterial spinal branches in the thoracolumbar spinal cord (dissecting technique, 10 rabbits).

The frequency of occurrence of individual ventral branches is shown in **Table 3**. The left-sided ventral branches entering the ventral spinal artery in thoracic spinal cord were present in 71.1% of cases and the right-sided ventral branches in 28.9% of cases. The left-sided ventral branches entering the ventral spinal artery in lumbar spinal cord were present in 52.4% of cases and right-sided in 47.6% of cases. Along the entire thoracolumbar spinal cord, the left-sided ventral

branches were present in 64.4% of cases and the right-sided in 35.6% of cases, which is most likely related to left-sided localization of the descending aorta.

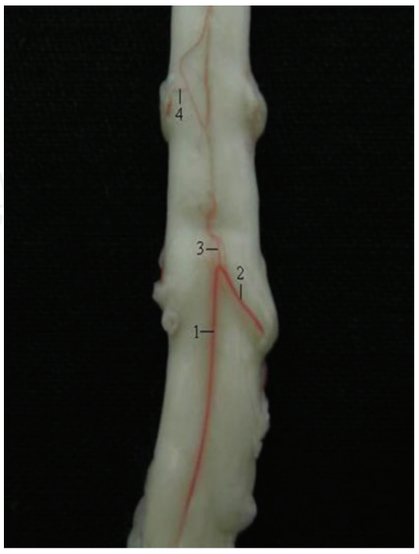

Figure 9. Left-sided localization of the artery of Adamkiewicz. (1) Ventral spinal artery, (2) the artery of Adamkiewicz, (3) branch of the artery of Adamkiewicz running cranially, and (4) ventral branch of spinal branch of the fifth right lumbar artery. Dissected specimen, ventral view, macroscopic image.

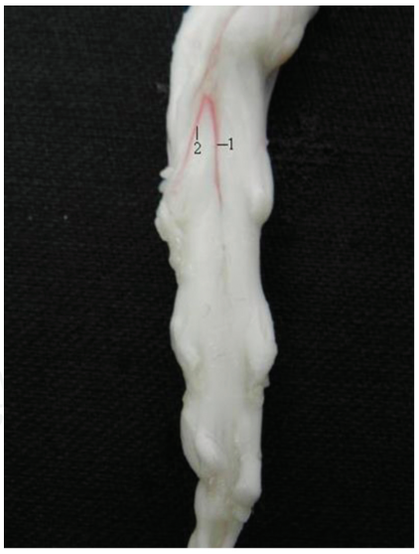

Figure 10. Right-sided localization of the artery of Adamkiewicz. (1) Ventral spinal artery and (2) the artery of Adamkiewicz. Dissected specimen, ventral view, macroscopic image.

A feeding artery with larger diameter entered the ventral spinal artery together with numerous weak spinal branches with smaller diameter. This bigger artery originated from the spinal branch which arose from the sixth lumbar artery. Thereafter, it arose and it ran through the intervertebral foramen to enter the vertebral canal. In all the studied specimens, we found this

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artery, which is known as the artery of Adamkiewicz or the arteria radicularis magna. It was present as left-sided artery in 50% of cases (**Figure 9**) and as right-sided artery also in 50% of cases (**Figure 10**). The artery of Adamkiewicz represented the arterial blood supply of the lumbar spinal cord caudally from the point of narrowing of the ventral spinal artery. After reaching median ventral fissure, it ran caudally replacing the ventral spinal artery and sent an important thin branch cranially to the thinning ventral spinal artery (**Figure 9**).

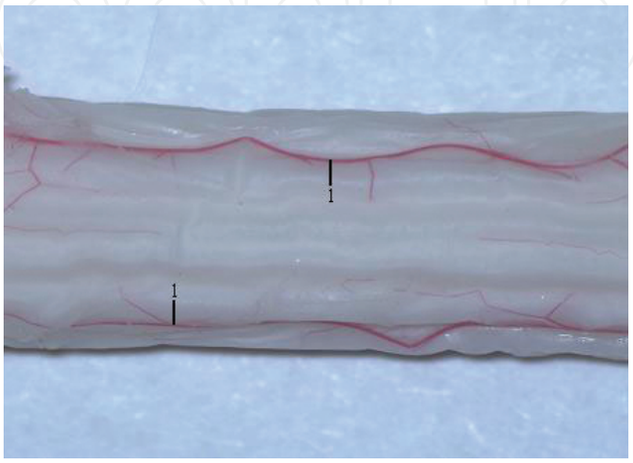

Figure 11. The presence of two irregular longitudinal dorsal spinal arteries. (1) Dorsal spinal artery. Dissected specimen, dorsal view, magnification 12.5x.

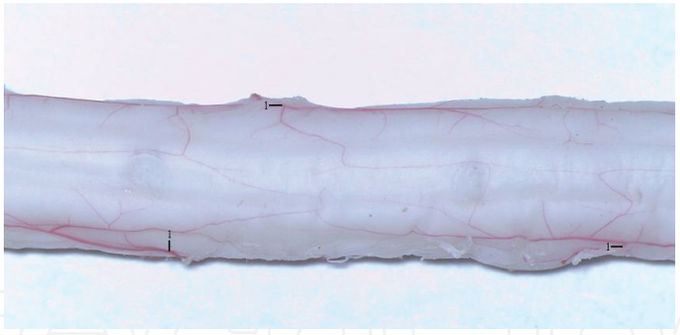

Figure 12. The absence of longitudinal dorsal spinal arteries. (1) Dorsal branch of spinal branch. Dissected specimen, dorsal view, magnification 12.5x.

On the dorsal surface of the thoracolumbar spinal cord, two irregular longitudinal dorsal spinal arteries were present in 70% of cases (**Figure 11**). They were located bilaterally in the lateral dorsal groove. We found the absence of longitudinal dorsal spinal arteries on the dorsal surface of thoracolumbar spinal cord in 20% of cases (**Figure 12**). Three irregular longitudinal dorsal spinal arteries receiving the dorsal branches were present in 10% of cases. The third artery was lying in the median dorsal groove (**Figure 13**). In the cases of the presence of two irregular longitudinal dorsal spinal arteries, these were formed only by the fusion of small cranially and caudally directed branches arising from the dorsal branches of the spinal branches. They

formed irregular loops between each other on the same and on the opposite side. The frequency of occurrence of individual dorsal branches is shown in **Table 3**. The dorsal branches in the thoracic spinal cord were present in 60.5% of cases as left-sided and in 39.5% of cases as right-sided. The dorsal branches in the lumbar spinal cord were present in 52.5% of cases as left-sided and in 47.5% of cases as right-sided. Along the entire thoracolumbar spinal cord, the left-sided dorsal branches were present in 58.2% of cases and the right-sided in 41.8% of cases; this is most likely related to the left-sided localization of the descending aorta.

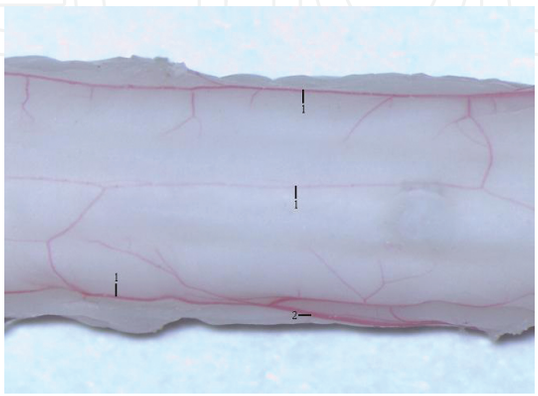

Figure 13. Dorsal branches of spinal branches forming three irregular longitudinal dorsal spinal arteries. (1) Dorsal spinal artery and (2) dorsal branch of spinal artery. Dissected specimen, dorsal view, magnification 12.5x.

3.2. Guinea pig

3.2.1. Cervical spinal cord

The arterial blood supply to the cervical spinal cord was more complex in comparison with the rest of the spinal cord. Numerous small branches arising from the posterior inferior cerebellar artery supplied the most cranial section of the first segment of the cervical spinal cord. Bilateral vertebral arteries entered the vertebral canal through the lateral vertebral opening of the atlas. These two arteries fused together on the caudal margin of the basilar part of the occipital bone. From the fusion originated the cranially directed basilar artery which participated on the formation of the cerebral arterial circle. The fusion of bilateral vertebral arteries has no triangular gap in 60% of cases (**Figure 14**) and one longitudinal gap in 30% of cases. A communicating branch between bilateral vertebral arteries was present in 10% of cases (**Figure 15**). The ventral spinal artery originated at the place of fusion of bilateral vertebral arteries. This origin was from the right-sided vertebral artery (**Figure 15**), from the left-sided vertebral artery, and from the anastomosis of two branches originating from the medial surface of the corresponding vertebral artery (**Figure 14**). The frequency of rostral origins of the ventral spinal artery is shown in **Table 4**.

The ventral spinal artery runs along the ventral median fissure of the cervical spinal cord. Spinal branches originating from the bilateral vertebral arteries entered the vertebral canal through the intervertebral openings. Inside the vertebral canal, they were divided into the

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dorsal and ventral branches with direction to the spinal cord. Some of the ventral branches joined the ventral spinal artery. The frequency of occurrence of individual ventral branches joining the ventral spinal artery is shown in **Table 5**. The left-sided ventral branches joining the ventral spinal artery were present in 58.2% of cases and the right-sided in 41.8% of cases.

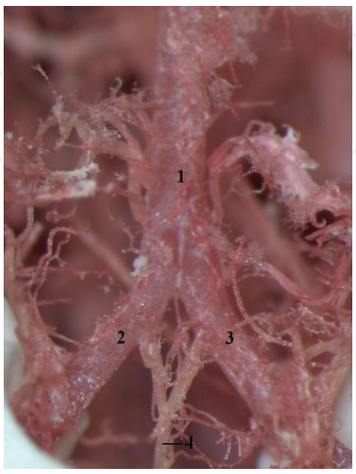

Figure 14. The anastomosis of two branches with origin on the medial surface of bilateral vertebral arteries forming the ventral spinal artery. (1) Basilar artery, (2) left vertebral artery, (3) right vertebral artery, and (4) ventral spinal artery. Macerated specimen, dorsal view, magnification 12.5x.

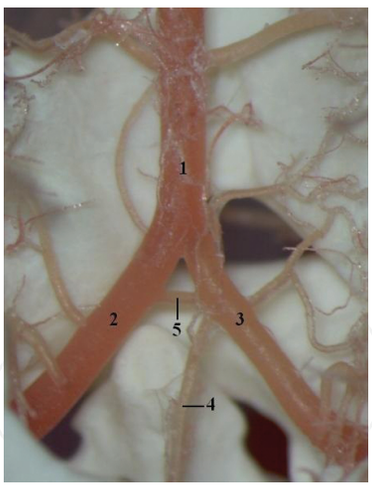

Figure 15. Ventral spinal artery originating from the right vertebral artery. Connection of both vertebral arteries by means of communicating branch. (1) Basilar artery, (2) left vertebral artery, (3) right vertebral artery, (4) ventral spinal artery, and (5) communicating branch. Macerated specimen, dorsal view, magnification 12.5x.

On the dorsal surface of the cervical spinal cord, we found two longitudinal dorsal spinal arteries in 60% of cases (**Figure 16**), three longitudinal dorsal spinal arteries in 30% of cases (**Figure 17**), or they were absent in 10% of cases. The fusion of the small cranially and caudally directed branches originating from the dorsal branches of spinal arteries represents the form of two longitudinal dorsal spinal arteries. In the cases of the absence of the dorsal spinal

arteries, the dorsal surface of the cervical spinal cord receives the arterial blood by means of dorsal branches of spinal arteries with very irregular arrangement (**Figure 18**). We found no rostral origins of dorsal spinal arteries in the place of fusion of bilateral vertebral arteries. The frequency of occurrence of individual dorsal branches reaching the cervical spinal cord is shown in **Table 5**. The left-sided dorsal branches were present in 63.3% of cases, and the right-sided dorsal branches were present in 36.7% of cases.

	Corrosion technique (%)	Dissecting technique (%)	Average (%)
Bilateral origin	20	30	25
Right-sided origin	40	40	40
Left-sided origin	40	30	35

Table 4. Rostral origin of ventral spinal artery using the corrosion technique (10 guinea pigs) and dissecting technique (10 guinea pigs).

Level	Occurrence of spinal branches (%)			
	Ventral		Dorsal	
	Right	Left	Right	Left
C 1	0	0	50	50
C 2	30	30	0	50
C 3	0	0	50	60
C 4	30	60	30	90
C 5	50	50	50	50
C 6	90	30	60	100
C 7	0	100	0	50
C 8	30	50	50	50

C, cervical segment of the spinal cord.

Table 5. Frequency of occurrence of ventral branches of spinal branches of the cervical spinal cord using the dissecting technique (10 guinea pigs).

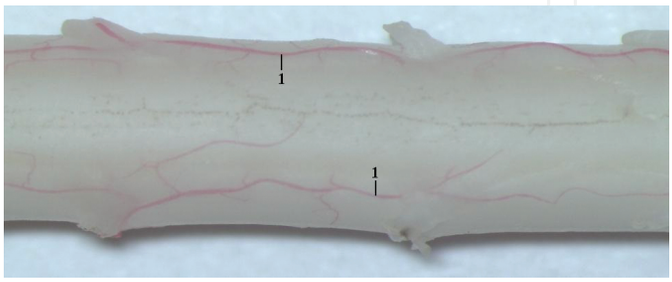

Figure 16. The presence of two dorsal spinal arteries. (1) Dorsal spinal artery. Dissected specimen, dorsal view, magnification 12.5x.

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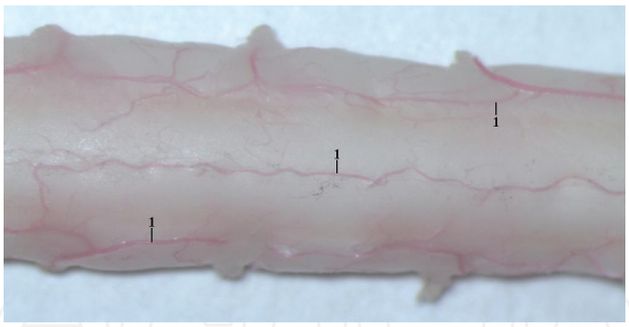

Figure 17. The presence of three dorsal spinal arteries. (1) Dorsal spinal artery. Dissected specimen, dorsal view, magnification 12.5x.

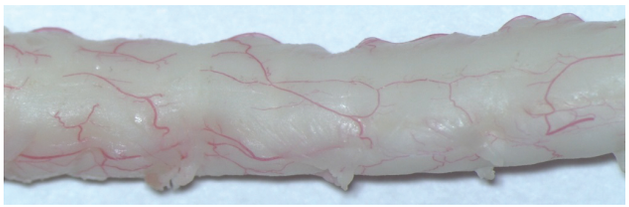

Figure 18. Dorsal branches of spinal branches with irregular arrangement. Dissected specimen, dorsolateral view, magnification 12.5x.

Figure 19. Dorsal intercostal arteries. (1) Thoracic aorta, (2) dorsal intercostal arteries with independent origin, (3) craniocaudal division of a common trunk of dorsal intercostal arteries, and (4) right-left division of common trunk of dorsal intercostal arteries. Macerated specimen, dorsal view, magnification 5x.

3.2.2. Thoracolumbar spinal cord

In the thoracic spinal cord, the arterial blood supply is performed by means of spinal branches arising from the dorsal intercostal arteries (**Figure 19**) which were present in number of 12

pairs. Dorsal intercostal arteries originated from the dorsal surface of the thoracic aorta in number of eight pairs in 70% of cases, in number of seven pairs in 20% of cases, and in number of nine pairs in 10% of cases. The remaining three to five pairs arose from the supreme intercostal artery. The origin of dorsal intercostal arteries by means of common trunk was present in 70% of cases. We found the division in right-left direction of common trunk in 60% of cases and in craniocaudal direction in 40% of cases. There was a high degree of variability present in the formation of common trunk. It was formed by two dorsal intercostal arteries in four cases, by three arteries in one case, by four arteries in one case, and by five arteries also in one case. The right- and left-sided arteries at the same level originated independently in 30% of cases.

The lumbar spinal cord received the arterial blood supply by means of spinal branches originating from the paired lumbar arteries. In all the cases, we found seven pairs of lumbar arteries. The first six pairs originated from the dorsal surface of the abdominal aorta, and the last one pair was a branch from the median sacral artery in 80% of cases. The origin of two last pairs from the median sacral artery was present in 10% of cases. Also in 10% of cases, we found the origin of all seven pairs from the abdominal aorta. The origin of lumbar arteries at the same level by means of a common trunk with the division in the right-left direction was present in 60% of cases. The independent origin of the right- and left-sided arteries at the same level was present in 40% of cases (**Figure 20**).

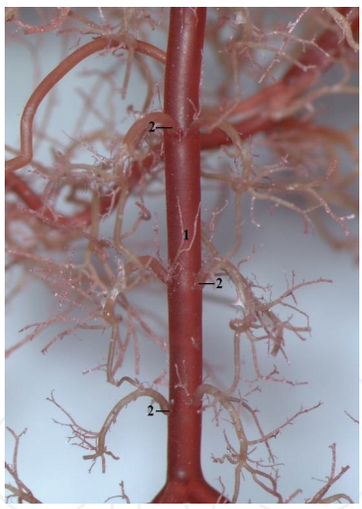

Figure 20. Origin of lumbar arteries. (1) Abdominal aorta and (2) independent origin of lumbar arteries. Macerated specimen, dorsal view, magnification 5x.

Dorsal intercostal arteries and lumbar arteries gave off spinal branches which entered the vertebral canal through the intervertebral openings. The entering was associated with the respective spinal nerve roots. After their entrance into the vertebral canal, the spinal branches divided into the dorsal and ventral branch. The ventral branches joined the ventral spinal artery. The occurrence of individual ventral branches joining the ventral spinal artery is shown in **Table 6**. The ventral spinal artery was located subdurally along the ventral median fissure of the thoracolumbar spinal cord. We found the left-sided ventral branches joining the

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ventral spinal artery in the thoracic spinal cord in 69.5% of cases and the right-sided in 30.5% of cases. We found the left-sided ventral branches joining the ventral spinal artery in the lumbar spinal cord in 54.2% of cases and right-sided in 45.8% of cases. Along the entire thoracolumbar spinal cord, the left-sided branches joining the ventral spinal artery were present in 63.8% of cases and right-sided in 36.2% of cases, which is most likely related to the left-sided localization of the descending aorta.

Level	Occurrence of spinal branches (%)			
	Ventral		Dorsal	
	Right	Left	Right	Left
Th 1	30	60	50	50
Th 2	30	30	50	60
Th 3	30	100	30	30
Th 4	0	30	0	30
Th 5	0	50	30	50
Th 6	50	50	30	30
Th 7	0	30	30	50
Th 8	30	60	30	0
Th 9	0	50	10	30
Th 10	50	50	30	50
Th 11	30	30	30	90
Th 12	0	30	60	30
L 1	0	0	50	30
L 2	0	90	30	30
L 3	30	0	0	30
L 4	60	30	0	30
L 5	50	50	0	60
L 6	50	0	90	100
L 7	30	90	30	100

L, lumbar segment of the spinal cord and Th, thoracic segment of the spinal cord.

Table 6. Occurrence of ventral and dorsal branches of arterial spinal branches in the thoracolumbar spinal cord (dissecting technique, 10 guinea pigs).

In addition to relatively small spinal branches, a bigger feeding artery with origin from the spinal branch of the fifth left lumbar artery in 60% of cases was present (**Figure 21**). The doubled artery of Adamkiewicz with two different levels of origin was present in 30% of cases. The left-sided artery originated from the spinal branch of the fourth lumbar artery and the right-sided

from the spinal branch of the fifth lumbar artery (**Figure 22**). The artery of Adamkiewicz with the origin from the spinal branch of the fifth right- and left-sided lumbar artery was present in 10% of cases. These two separated arteries were continuing caudally on the ventral surface of the lumbar spinal cord. These two arteries fused together at the level of the sixth lumbar vertebra. From this point, the single ventral spinal artery continued caudally. A communicating branch connected together with the bilateral spinal branches at the level of the fifth lumbar artery and sent cranially thin branches joining the ventral spinal artery (**Figure 23**). In all the cases, the artery of Adamkiewicz was joining the ventral spinal artery.

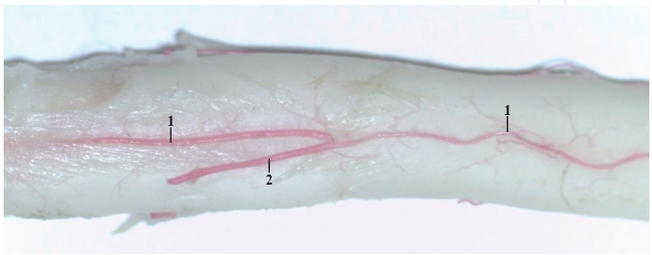

Figure 21. Left-sided localization of artery of Adamkiewicz. (1) Ventral spinal artery and (2) artery of Adamkiewicz. Dissected specimen, ventral view, magnification 8x.

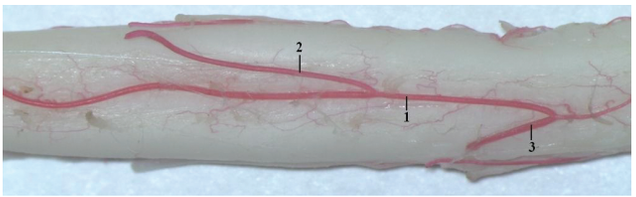

Figure 22. Doubled artery of Adamkiewicz. (1) Ventral spinal artery, (2) right-sided artery of Adamkiewicz, and (3) left-sided artery of Adamkiewicz. Dissected specimen, ventral view, magnification 8x.

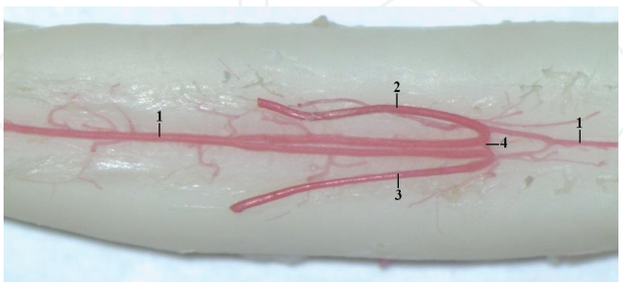

Figure 23. Doubled artery of Adamkiewicz. (1) Ventral spinal artery, (2) right-sided artery of Adamkiewicz, (3) left-sided artery of Adamkiewicz, and (4) communicating branch. Dissected specimen, ventral view, magnification 12.5x.

In 60% of cases, we found two irregular longitudinal dorsal spinal arteries located in lateral dorsal grooves (**Figure 24**). The dorsal branches of spinal branches were joined to the dorsal spinal arteries. Three irregular longitudinal dorsal spinal arteries receiving the dorsal branches

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of spinal branches were present in 40% of cases (**Figure 25**). The third dorsal spinal artery runs along the median dorsal groove. The occurrence of individual dorsal branches is shown in **Table 6**. In the cases of the presence of two irregular longitudinal dorsal spinal arteries, they were formed only by the fusion of the small cranially and caudally directed branches originating from the dorsal branches. The left-sided dorsal branches in the thoracic spinal cord were present in 56.8% and the right-sided in 43.2% of cases. The left-sided dorsal branches in the lumbar spinal cord were present in 65.5% of cases and right-sided in 34.5% of cases. Along the entire thoracolumbar spinal cord, the left-sided dorsal branches were present in 60.3% of cases and the right-sided in 39.7% of cases.

Figure 24. The presence of two longitudinal dorsal spinal arteries. (1) Dorsal spinal artery. Dissected specimen, dorsal view, magnification 12.5x.

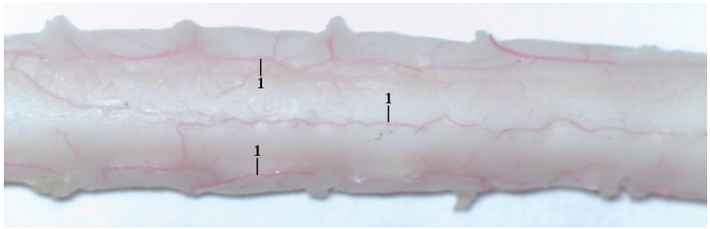

Figure 25. The presence of three longitudinal dorsal spinal arteries. (1) Dorsal spinal artery. Dissected specimen, dorsal view, magnification 12.5x.

4. Discussion

4.1. Cervical spinal cord

Based on our results, it can be concluded that the blood supply of the cervical spinal cord in rabbit and guinea pig has high variability. In contrast with our findings, only uniform origin of the ventral spinal artery in both species was described [16, 17]. Cervical spinal cord injury was studied in several species of experimental animals. The dogs, rats, pigs, rabbits, and guinea pigs belong to the most used species. The arterial arrangement of the cervical spinal cord in the dog was studied in detail with pointing on the variations in formation of the ventral spinal artery and the frequency of occurrence of spinal branches [18]. The rat was also studied in details, but the results of several studies differ [17, 19–21]. In pigs, only variations and the

presence of extrasegmental arteries of the spinal cord blood supply were described [22, 23]. The frequency of occurrence of the spinal branches in our study was higher on the left than on the right side, opposite to the dog [18].

The arterial blood supply to the cervical spinal cord in monkeys, dogs, rabbits, and rats was studied by [24]. The origin of the ventral spinal artery was not recorded, and the ventral spinal artery was described as paired vessel. In our specimens, the ventral spinal artery was in a form of a single trunk with different types of origin in the place of fusion of bilateral vertebral arteries. In this work, the origin of dorsal spinal arteries from the posterior inferior cerebellar artery was described. In our specimens, we did not find the origin of the dorsal spinal arteries.

Some reports described the similarity of the arterial blood supply to the cervical spinal cord in rabbits, guinea pigs, and humans [16, 17]. Based on our study, we can conclude that there is partly different arterial pattern compared with human. The fusion of basilar artery is in human without gap [25]. In rabbits and guinea pigs, we found different types and numbers of gaps. In humans, the anterior spinal artery (homologue to the ventral spinal artery in animals) is formed by the fusion of the anterior spinal branches arising from the vertebral arteries [26]. In rabbits and guinea pigs, we found three different types of origin of ventral spinal artery in the place of fusion of vertebral bilateral arteries. In rabbits, we found the right-sided ventral branches joining the ventral spinal artery in 46.2% and left-sided in 53.8% of cases, and in guinea pig, the right-sided ventral branches were present in 41.8% of cases and the left-sided in 58.2% of cases. Only two or three ventral branches joining the anterior spinal artery were described in humans [27].

In rabbits and guinea pigs, we found on the dorsal surface high variability in the arrangement of the dorsal spinal arteries (in human, the posterior spinal arteries). The posterior spinal arteries in human are normally continuous rostral to caudal and supply the posterior third of the spinal cord [28]. The frequency of occurrence of individual dorsal and ventral branches in rabbits and guinea pigs was greater than in the case in humans.

According to our results, it can be concluded that the higher resistance to ischemic damage by the interruption of ventral and dorsal spinal arteries was because of the presence of dorsal and ventral branches reaching the cervical spinal cord in almost every segment. Rabbits and guinea pigs are often used as an experimental model for the study of spinal cord injury. The cervical spinal cord served as experimental model for the study of several types of damage [13, 29, 30].

4.2. Thoracolumbar spinal cord

Based on our results, it can be concluded that the blood supply to the thoracolumbar spinal cord in rabbit and guinea pig has high variability. The anatomical arrangement with regard to the origin of segmental dorsal intercostal and lumbar arteries has a very important role during operations of thoracoabdominal aneurysms [31]. Correctly performed reimplantation of segmental arteries decreases the risk of spinal cord ischemia, which can also lead to the paraplegia [32–34]. Till now in rabbits, the segmental arteries were described as paired branches originating independently from the dorsal surface of descending aorta [35, 36]. In guinea pigs, the presence of dorsal intercostal arteries with the origin from the supreme

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intercostal artery and the costocervical trunk was very variable. It varies from four to seven arteries on each side. Twelve pairs of dorsal intercostal arteries were present, and the remaining arteries were direct branches with the origin from the thoracic aorta [37]. In guinea pigs, the dorsal intercostal and lumbar arteries were described as paired branches arising independently from the dorsal surface of descending aorta [35, 36]. In guinea pigs, two types of origin of seven pairs of lumbar arteries were found: an independent origin and origin by means of a common trunk of the arteries at the same level [38].

In the study of ischemic injury in the thoracolumbar spinal cord, dogs, rats, pigs, rabbits, guinea pigs, and mice were used as experimental animals. In dogs, high variability in the density of arteries forming the spinal arterial ring and in the spinal branches was described [18]. In rats, the results in the study of arterial supply to the thoracolumbar spinal cord were very different [17, 19–21]. The dorsal spinal arteries were found in number of two [39] or as less constant [19]. In pigs, the studies were concentrated on the extrasegmental blood supply to the thoracolumbar spinal cord [22]. In mice, the spinal cord blood supply was partially described [10, 40].

Only one work dealing with arterial arrangement of the thoracolumbar spinal cord in rabbit and guinea pig was published [17]. But in this work, the artery of Adamkiewicz, the place of its origin, and any other variations were not described. We found variable arrangement of the artery of Adamkiewicz in rabbit and guinea pig, but in both species it was present in all cases. In guinea pig, doubled artery of Adamkiewicz with origin from spinal branch of the third or fourth lumbar artery was found [16]. In our study, the artery of Adamkiewicz was single or doubled with variable level of origin. In dogs, the artery of Adamkiewicz was found only in one half of the studied specimens. In rats, the presence of artery of Adamkiewicz is questionable. Some authors described its presence in all cases [17, 19, 20, 41], but some authors doubt its presence [21, 42]. In pigs, the artery of Adamkiewicz was not described [12, 13]. In mice, it was found in all cases [10] and also in humans [43].

The vascular arrangement of the dorsal spinal arteries in rabbits and guinea pigs was very variable. The dorsal spinal arteries in guinea pigs were described as two smaller anastomotic chains of arteries, running in the lateral dorsal grooves [16]. In our study, the number of dorsal spinal arteries varied from two to three. In humans, the posterior spinal arteries (homologue to the dorsal spinal arteries in animals) were found as normally trunks continuing in the cranial to caudal direction [28]. In dogs, four dorsal spinal arteries were described [18]. In rats, two much less constant dorsal spinal arteries with irregular connections between each other were found [19]. In mice, two spinal arteries [10] or only one single artery were described [40].

Our results indicate high variability in the presence of dorsal and ventral branches supplying the rabbit and guinea pig thoracolumbar spinal cord. On the left side, they occurred in higher numbers. The segmental arteries reaching the spinal cord ensured the blood supply of the ventral and dorsal surface of the respective segments of thoracolumbar spinal cord. In rabbits, the absence or irregularity of dorsal and ventral branches supplying the thoracic spinal cord was higher than that of branches supplying the lumbar spinal cord. The higher risk of ischemic damage to the thoracic spinal cord in rabbit was concluded. In guinea pigs, we found higher absence or irregularity of dorsal and ventral branches supplying the lum-

bar spinal cord, which allowed us to assume higher risk of irreparable ischemic damage to the lumbar spinal cord.

Based on results of the study, it is possible to conclude that the more appropriate model for the experimental study of ischemic injury of the thoracic spinal cord is the guinea pig and of the lumbar spinal cord is the rabbit, due to a lower incidence of variations of arterial arrangement in the corresponding spinal cord region. This implies that the thoracic spinal cord in guinea pig and lumbar spinal cord in rabbit are the most similar in their arterial arrangement to the homosegmental blood supply of human spinal cord.

5. Conclusion

The principles of blood vessel distribution to the spinal cord can be explained by the studies of arterial arrangements of several animals used as experimental models. In general, these studies can provide the additional information about the vascularization schema of the central nervous system [19].

For the prediction of functional results of neurological injuries and disorders, animal models from which the rodent models have a special place were used. Several clinical symptoms described in human patients are very parallel to the symptoms observed in rodents. The analysis of therapeutic approaches and behavioral sequel will help to determinate the limitations and strengths of animal models. It is very important to respect each aspect before an experimental study is started [44]. It is important to assess goals and expectations of the experiment before choosing a model. The understanding of the arterial arrangement to the spinal cord plays a very important role in avoiding the spinal cord ischemia or infarction during surgical interventions to the spine [45]. The presence of the artery of Adamkiewicz and nearly regular segmental blood supply to the thoracolumbar spinal cord in all studied animals is responsible for the use of rabbit and guinea pig as a simple model of ischemic damage to the thoracolumbar spinal cord.

The determination of appropriate species in the experiments of spinal cord injury requires the detailed study of the spinal cord arteries in all species used in this research area. The biased or erroneous outcomes can be caused by the presence of variation in arterial arrangement.

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