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Original paper

Mechanical properties of cervical dura mater

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The aim of the study was to determine experimentally the stress as strain function as well as the orthotropy and heterogeneity of porcine dura mater of the cervical spinal cord. Material was divided into groups based on the place of collection, considering the dorsal side and ventral side, specifying the number of cervical vertebra, and the direction of tension of the sample – longitudinal or circumferential. Experimental studies were conducted with the MTS Synergie 100 testing machine. The tensile test was performed for each sample at a speed of 2 mm/min until the sample's break. There were determined the characteristics of stress as a function of strain in particular samples. Distribution maps of the stress and strain values at the characteristic points were then drawn (the beginning and the end of the linear range of the stress–strain characteristic and the point corresponding to the complete sample damage) for each set of samples, taking account of their collection place and direction of tension. The results confirmed the orthotropy of mechanical properties of dura mater. Stress and strain differed also in the value at the height of each vertebra and exhibited diversification on the ventral side compared to dorsal one.

Key words: *dura mater, mechanical properties, orthotropy, tensile test*

1. Introduction

Thanks to its unique biomechanical properties, dura mater protects the spinal cord against mechanical damage. Its mechanical properties depend mainly on the microstructure [1], [2]. The dura mater is a tissue rich in fibrous proteins such as collagen and elastin, which occur in intercellular matrix. Scanning electron microscope analysis shows that dura mater is composed of the bundles of many interconnected collagen fibres which run parallel and generally in the longitudinal direction, corresponding to the mater's basic load direction. There is an amorphous substance between them – thicker elastic fibres. The number of transversal fibres is small and limited primarily to the inner layers where the fibres run perpendicular to the longitudinal fibres [2]. Different internal structure [3], various at different heights, depends on the direction of the load – the more lateral or more oblique [1], [4].

Due to elastin fibres, many tissues of vertebrate bodies can reversibly twist, bend and strain. A characteristic feature of elastic fibres is their susceptibility to tension, so that they can achieve several times greater length and quickly recover their original shape and size after removal of the tensile force. However, if the limit of the tensile strength of fibres is exceeded, they are easily broken [5].

A typical stress–strain characteristic of soft tissue is shown in figure 3. Its nonlinearity was caused by the microstructure of biomaterial. The fibres immersed in intercellular matrix had a significant impact on the behaviour of tissue during tensile test. Elastin fibres were responsible for a linear portion of the first nonlinear section of characteristics. Then, when the plot begun to rise (the first non-linear range), elastin and

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collagen fibres acted together. A suitable force acted on the collagen fibres, which mainly determined the tissue behaviour, and the next linear segment appeared on the characteristics. When both types of fibres begun to undergo destruction the maximum stress ($\sigma_{\max}$) was reached, causing permanent damage to the tissue. The reduction of the force acting on the sample occurred due to an avalanche rupture of all the fibres [6].

The mechanical properties of collagen and elastin fibres were different in different directions, which was caused by their spatially complex network. Thus, the structural orthotropy caused mechanical orthotropy.

Probably the structural fibres are unevenly distributed along the entire length of the spinal cord. It is therefore possible that the mechanical properties are different not only in different directions, but also at different heights of the spinal cord. It can be assumed that there is heterogeneity of mechanical properties along the entire length of the spinal cord.

Wilcox et al. [7] tested the mechanical properties of animal dura mater. However, they diversified their samples based only on the stretching direction (circumferential and longitudinal). Similarly, Runza et al. [8] studied dura mater from thoraco-lumbar specimens of animal and human spines. However, they determined the dependence of tensile strength on mechanical properties of dura mater, taking into account only the stretching direction (circumferential and longitudinal) of the sample subjected to tensile force. In literature, there is little description of testing mechanical properties of dura mater, depending on the sampling site, which could be extremely useful from clinical point of view. In recent years, numerical modelling of the spine and spinal cord has attracted an increasing interest [9]–[13]. In some of those models, dural sac plays an important role as a supportive and load transferring structure [14]. As each complex numerical model should reflect real anatomical and biomechanical conditions, it is essential to improve the state of knowledge about mechanical properties of its components. The aim of our study was thus to determine experimentally the dependence of stress as strain function of porcine dura mater on its mechanical properties at different levels of the cervical spinal cord with a special emphasis put on the clinical application of the results achieved.

2. Material and method

The animal-derived material was used for our experimental studies. The soft tissue of porcine origin is a good model showing the behaviour of human soft tissue, which was described by Runza et al. [7] and also by Sparrey et al. [15], [16]. Nine freshly collected porcine spines were frozen. Then, after thawing at room temperature, the spinal cord was prepared. The dura mater was dissected from the spinal canal by means of microsurgical instruments. The purification of the material was intended to gently remove unnecessary structures, such as blood vessels and nerve roots, for they would not affect the measurement. The properly prepared samples were stored in 0.9% normal saline at 4 °C for no longer than 24 h. This practice affected neither the structure of maters nor any of their mechanical properties [8].

For all the vertebral columns tested the sampling algorithm was the same. Seven fragments from C1 to C7 were excised from all the cervical sections. Then the dura mater was removed, cut up on the side, so that the ventral and dorsal sides remain intact. The next step was to cut the samples in the longitudinal and circumferential directions from every side (ventral and dorsal). Thus, 4 samples were obtained from each level. Schematic of sampling is shown in figure 1.

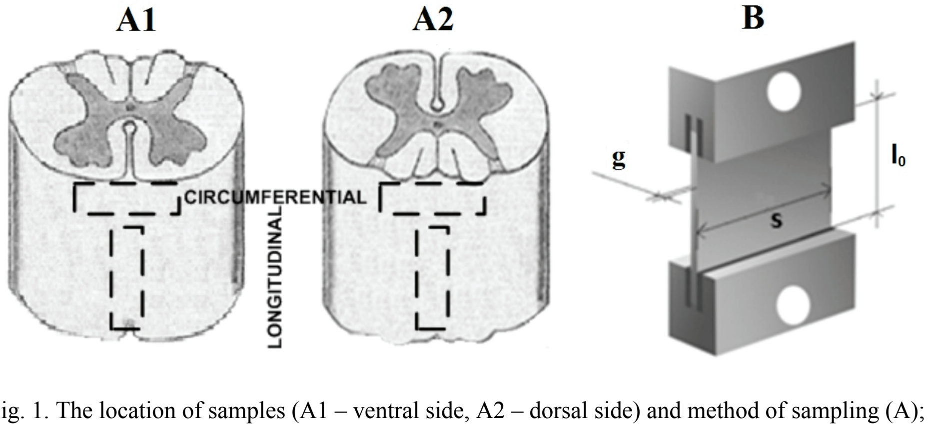

Fig. 1. The location of samples (A1 – ventral side, A2 – dorsal side) and method of sampling (A); the method of dimensioning (B)

In total, 28 samples were collected from every cervical spine specimen. In order to analyze the mechanical properties of dura mater, more than 250 samples were measured during all studies. Material was divided into groups, depending on:

- the site of the sample taking, i.e., the height of vertebra from C1 to C7,
- sampling site with the distinction between the dorsal and ventral sides,
- the direction of the sample tension: circumferential or longitudinal.

Some results had to be omitted, since they differed considerably from the others and could therefore interfere with reality. Mostly it was due to the mater's sliding out of the handles and due to rupture near the place of mounting.

Fixing samples in the testing machine was very difficult because of their small size. Therefore, specially designed clamps were prepared. The fragments of susceptible rubber were glued to the ends of the clamps to prevent the sample slipping. This fulfilled its role in the vast majority of cases. Before performing the tensile test the sample dimensions were measured (figure 1). Its thickness g was measured using the Mitutoyo CCP OK3 device (Mitutoyo Co., USA), while the width s and the length l_0 of the active sample were measured using callipers. The samples had almost the same geometric dimensions: 8.1 ± 0.4 mm in length, 5.3 ± 0.3 mm in width and 0.08 ± 0.01 mm in thickness.

The uniaxial tensile test was performed with the MTS SYNERGIE 100 machine (MTS Systems, Inc., USA) at a speed of 2 mm/min until a complete rupture of the sample at ambient temperature of 24 °C.

The dependence of the force F (N) on the displacement Δl (mm) was obtained as a result of the tensile test. Based on the data obtained the stress–strain characteristic was determined.

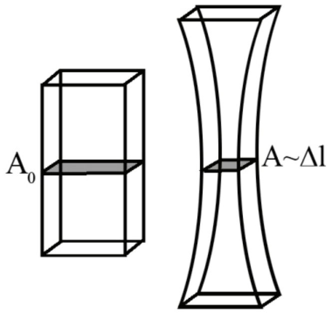

Fig. 2. Diagram showing the change in cross-section of the sample during tension

Since the biomaterial is characterized by the strain greater than 15%, the theory of great strains was used to explain the results. Taking into account the change in the cross-section of the sample (figure 2) during the stretching, the surface was calculated according to the formula [17]:

$$A = \frac{l_0 s g}{l_0 + \Delta l},$$

where:

A (mm^2) – cross-sectional area,
l_0 (mm) – initial length of the sample,
Δl (mm) – displacement,
s (mm) – width,
g (mm) – thickness.

Thereafter, the stress–strain characteristics for each sample were determined, depending on the vertebra's height from C1 to C7, the direction of sample tension (the circumferential and longitudinal) from the ventral and dorsal sides.

3. Results

The characteristics representing a given group of samples were averaged and in this way the averaged characteristics were obtained for each of them.

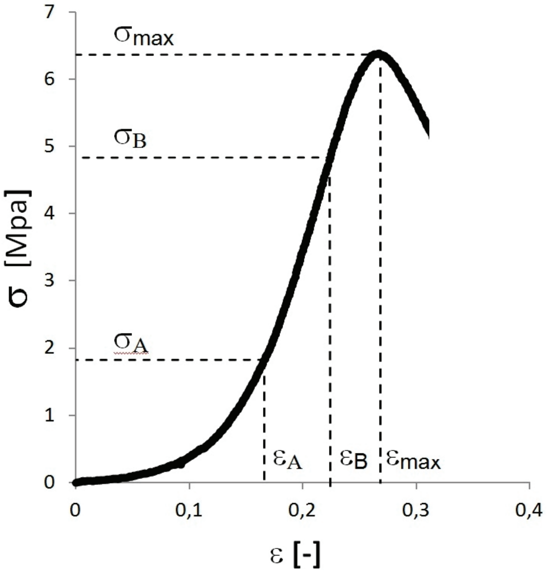

Fig. 3. Example characteristic of stress–strain with the selected characteristic points

The coordinates of the following specific points were determined from the curve representing the stress as the function of strain (figure 3):

(ε_A, σ_A) – the beginning of the linear range;
(ε_B, σ_B) – the end of the linear range;

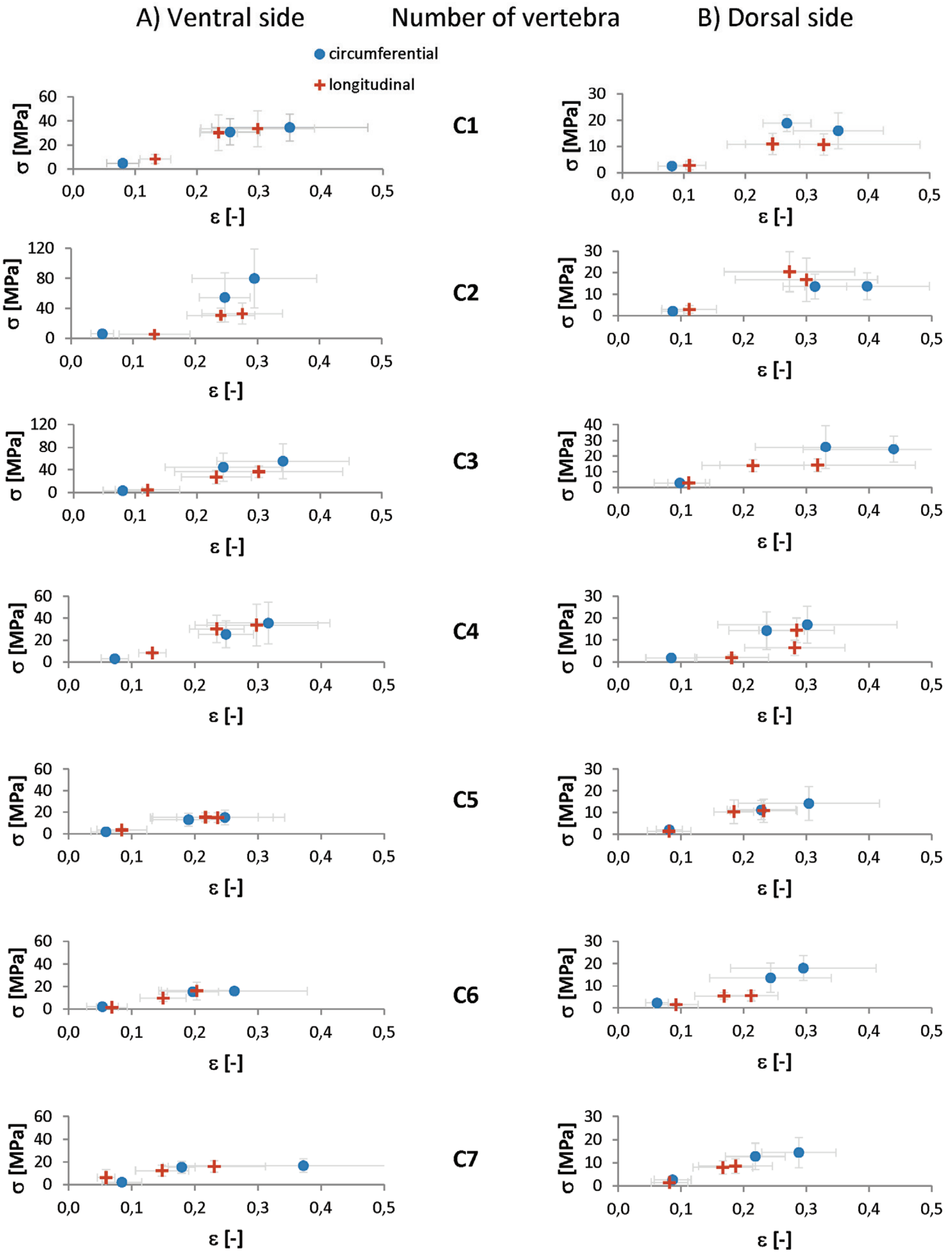

Fig. 4. The coordinates of characteristic points for each vertebra: A) ventral side, B) dorsal side with standard deviation. The circumferential tension (circle) and the longitudinal tension (cross)

($\varepsilon_{\max}$, $\sigma_{\max}$) – the points representing the maximum stress.

Thus, the range of nonlinearity was established, mainly due to the stress of elastin fibres [(0, ε_A); (0, σ_A)], the range of linearity due to the physiological stress [(ε_A, σ_A); (ε_B, σ_B)], and the range of nonlinearity caused by overloading the material [(ε_B, σ_B); ($\varepsilon_{\max}$, $\sigma_{\max}$)].

The curves were plotted based on the data obtained (figure 4A and 4B). They show the differences in the coordinates of specific points at the height of each vertebra. The data series were compared: the circumferential tension (circle) and the longitudinal tension (cross) on each individual chart. Figure 4A shows the results for the ventral side, figure 4B for the dorsal one.

The differences in the values of stress and strain at characteristic points between the samples stretched circumferentially and longitudinally are clearly visible on the charts. Typically, the stress causing permanent damage to the dura mater has a higher value for the samples stretched circumferentially compared to those stretched longitudinally. To deform the dura mater in the circumferential direction a greater force was needed than to deform it in the longitudinal direction. Such a dependence was observed both for the ventral and dorsal sides.

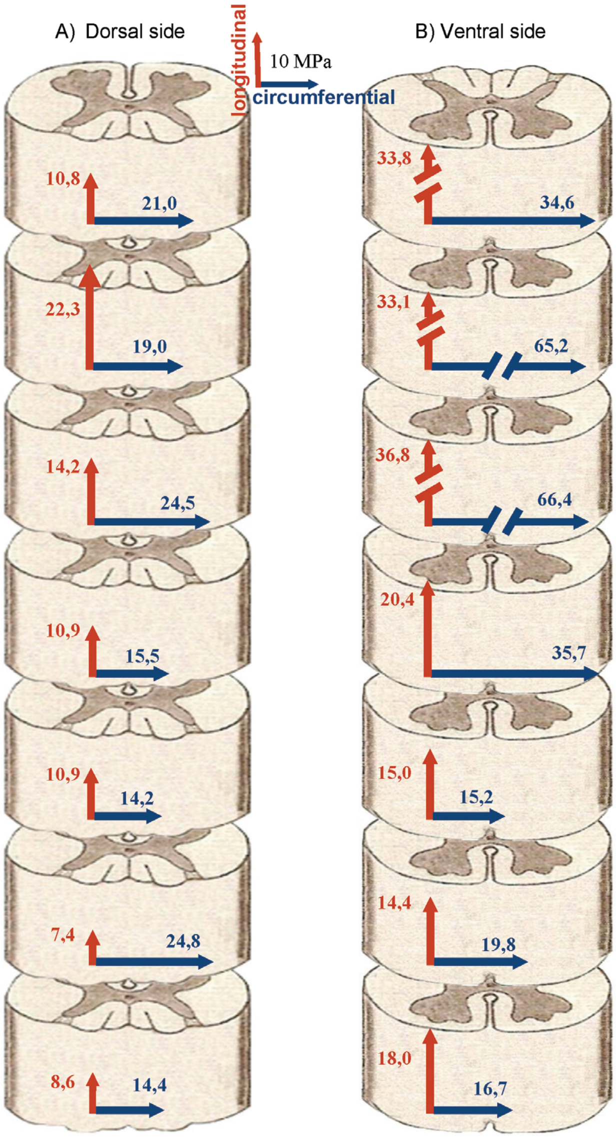

Fig. 5. Map of stress distribution at the point $\sigma_{\max}$ (MPa): A) dorsal side, B) ventral side

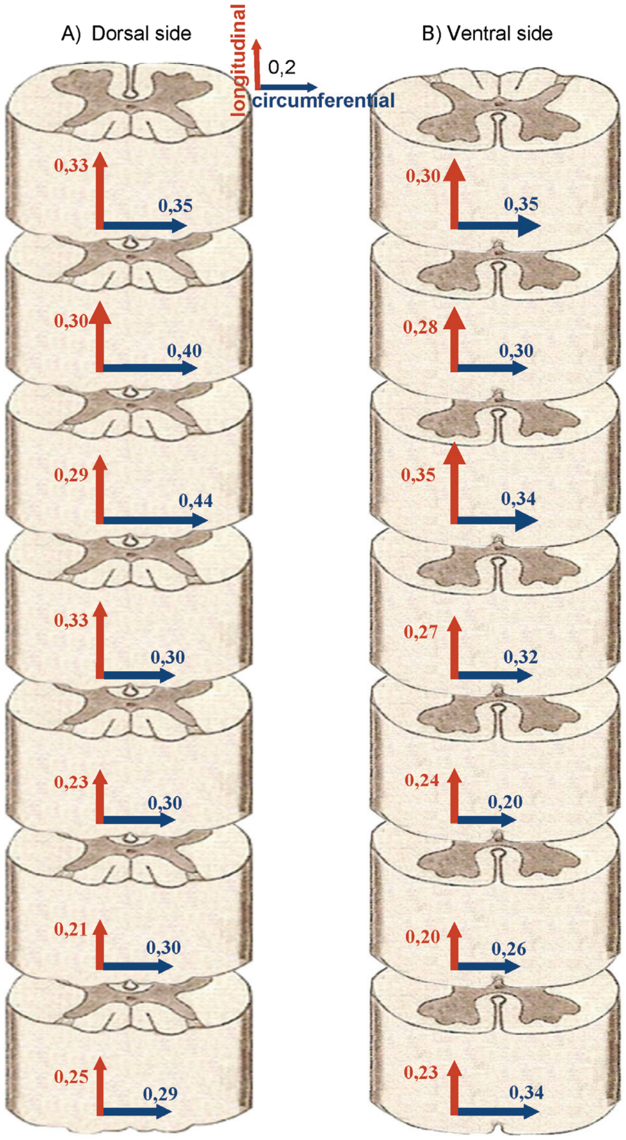

Fig. 6. Map of strain distribution at the point $\varepsilon_{\max}$ (–): A) dorsal side, B) ventral side

The map of the distribution of mechanical properties at the characteristic points *A*, *B*, and MAX was designed to accurately describe how the values differ primarily at the height of each section of the cord, for both the ventral and dorsal sides. In this way, the differences in the values of these parameters, depending on the direction of the samples tension: longitudinal, consistent with the cord long axis, and circumferential, were also illustrated.

The separate lists for the points ε_A, ε_B, $\varepsilon_{\max}$, σ_A, σ_B, $\sigma_{\max}$ were created to compare the state of stress and strain throughout the whole cervical spinal cord and its ventral and dorsal sides. The distributions of $\sigma_{\max}$ (figure 5) and $\varepsilon_{\max}$ (figure 6) for both investigated sides of the spinal cord were shown. Based on such sets, the values of these coordinates were analysed.

The coordinate ε_A: The distribution of strain at the dura mater's point ε_A was diversified along the length of the whole of the cervical spinal cord. Sample failed within the physiological load range at the strain higher in the case of longitudinal tension than circumferential tension, and on the ventral and dorsal sides. The dorsal side strain in the longitudinal direction was found in the range of 0.09–0.18 (–), in the circumferential direction in the range of 0.06–0.10 (–); on the ventral side, respectively, 0.06–0.13 (–) and 0.05–0.10 (–). Thus, on the dorsal side the strain was greater than that on the ventral one.

The coordinate ε_B: The distribution of strain at the point *B*, at which the range of tissue overload began, was very diversified, especially in the circumferential direction on both sides; the strain values were slightly higher for the dorsal side than those for the ventral one. In the longitudinal direction on the dorsal side, the strain values increased to 0.28 (–) in the section of C1–C4, followed by a decrease in the sections C5 and C6 to 0.17 (–) and finally in the section C7 they again reached a maximum value equal to 0.28 (–). In the circumferential direction for the dorsal side, the strain ranged from 0.22 (–) to 0.33 (–), and its maximum value was reached at the height of the section C3. On the ventral side the strain, characterising the end of the linear range of physiological loads, was larger in the circumferential direction and ranged from 0.19 to 0.27 (–); for the longitudinal direction, 0.15–0.24 (–).

The coordinate $\varepsilon_{\max}$: The percentage of strain causing the permanent damage to the sample was higher in the circumferential direction than in the longitudinal one regardless of side, from which the sample had been taken. Analysing the individual strain values in the longitudinal direction for each level, we observed that its distribution was very diversified on both sides of the spinal cord. For the dorsal side the lateral strain was the greatest for the level C3, and decreased on the adjacent vertebrae. For the vertebra C5 of ventral side the deflection $\varepsilon_{\max}$ was the smallest, and away from it – it grew.

The coordinate σ_A: The stress (σ_A) characterising the starting of the range of physiological loads acting on the dorsal side was smaller than on the ventral one, both in circumferential and longitudinal directions. So greater force had to be used to stretch the sample in the circumferential direction than in the longitudinal one, regardless of sampling place. On the dorsal side significantly higher values were observed for the first 3 sections in the longitudinal direction, the difference in values was 3.4 (MPa). However, in the circumferential direction along the entire length of spinal cord they were in the range of 2.3 ± 0.5 MPa. On the ventral side with the longitudinal tension, the largest stress occurred in the section C1, and was almost five times higher than that in the section C6, where the stress reached its minimum. The force needed to stretch the mater from the ventral side in the circumferential direction varied widely, because the stress ranged from 2.2 MPa to 9.4 MPa.

The coordinate σ_B: The value of stress, which triggered nonphysiologic strain of the dura mater, was smaller for the dorsal side than for the ventral one; a similar relation was found for both tensile directions. Stress on the dorsal side in the longitudinal direction was the greatest for the section C2, its value significantly differed from the other values and exceeded 12 MPa, i.e., the value higher than the minimal one for the section C6 equal to 8 MPa. One cannot clearly declare whether lateral or longitudinal strain is larger. However, on the ventral side the lateral stress was greater, for example, in the section C3 it was almost twice that value. In the longitudinal direction, the greatest force had to be used to deform the dura mater in the sections C1 and C2 on the ventral side. Also a decreasing tension was visible in the longitudinal direction with an increase in the vertebra number for the ventral side. It should also be noted that in the circumferential direction, the greatest stress terminating the range of physiological load σ_B as well as the stress starting the range σ_A occur in the section C3.

The coordinate $\sigma_{\max}$: There may be a correlation between the stress value $\sigma_{\max}$ and the stress values permanently damaging the tissue. To stretch the sample in the circumferential direction until it was destroyed the greater force was necessary compared to the force used in the longitudinal tensile direction on both the dorsal and ventral sides. However, there was not any constant relationship between the value of a circumferential stress and a vertebra's number. For example, analysing the ventral side along the cord length, it was visible that the stress increased in the C1–C3 sections, then it decreased in the C3–C5, and once more increased and decreased. On the dorsal side, the higher the vertebra's number, the less the force necessary for a permanent damage to the dura mater subjected to longitudinal tension. It could be concluded that a similar phenomenon was observed on the ventral side.

Totalling the obtained values of characteristic points, it was noted that along the entire length of the cervical portion the dura mater was subject to greater stress in the circumferential direction. Dura mater was deformed at lower values of forces acting longitudinally. This dependence was observed for the samples collected from the ventral and from the dorsal sides. The only exceptions were the samples of the section C2 (dorsal side) and the section C7 (ventral side). However, the distribution of stress values was very diverse, both across the entire cervical portion and in different directions. The stresses causing the permanent damage to the structure were much higher on the ventral side of the dura mater than these on the dorsal one. Typically, the ventral side was able to withstand greater loads without any damage to its structure. However, the dispersion of values was so great that a specific relationship could not be found.

4. Discussion

The structure of the dura mater is adapted to the loading it has to withstand. The dura mater consists of the fibres arranged mainly in the direction of load acting on them. With every movement of the head and the cervical spine the dural sac also moves. In the extreme forward bending, the accompanying movement of the dura mater occurs, right up to the lower part of chest [18]. In the flexion movement, the dura mater is pulled upward, and returns to its previous place during straightening. Similar but weaker shifts occur in the lateral and rotational movements of the cervical spine [1]. The vertebra C2 is primarily responsible for the head rotation, and therefore at the height of that segment the highest stress and strain occur. In the circumferential direction, these values were as follows: 80 ± 39 MPa, 0.3 ± 0.2 (–) for ventral side and 14 ± 6 MPa, 0.4 ± 0.1 (–) for dorsal side. In the longitudinal direction, these values were: 33 ± 14 MPa, 0.3 ± 0.1 (–) for ventral side and 17 ± 10 MPa, 0.3 ± 0.1 (–) for dorsal side. At the height of the lowest vertebrae the rotational movement is negligible, that is why the mechanical properties are reduced below the vertebra C2. Different values of stress and strain occurring at the height of each cervical vertebra may indicate that the fibres' texture is varied. Similarly, spinal ventral side is more responsible for the vertebra stability than the dorsal one, so it seems that the ventral side should be stronger. This assumption has been confirmed by the results of our studies. Both stress and strain distributed throughout the cervical spine had higher values for the ventral side than for the dorsal one.

It was shown that in the lumbar spine, where the greatest stresses occurred in the longitudinal direction, the collagen fibres were oriented in the direction of acting loads. RUNZA et al. [8] have shown the values of maximum stress ($\sigma_{\max}$) in bovine samples from lumbar region and subjected to longitudinal tension were as follows: ≈11÷22 MPa for fresh samples, ≈14÷15 MPa for the samples frozen for 96 h at –4 °C and ≈17÷23 MPa for the samples stored for 96 h at 4 °C. The values of maximum stress ($\sigma_{\max}$) in the samples from cervical region stored in a physiological NaCl solution at 4 °C no longer than 24 h at longitudinal tension obtained by us are the following: 7÷22 MPa for dorsal side and 14÷37 MPa for ventral side. Runza et al. have also shown the values of maximum strain of bovine samples ($\varepsilon_{\max}$) subjected to longitudinal tension. These values ranged from ≈90 to 120% for fresh samples, from ≈80 to 100% for the samples frozen for 96 h at –4 °C and from ≈100 to 115% for the samples stored for 96 h at 4 °C. The results presented in our study ranged from 21 to 33% for dorsal side and from 20 to 35% for ventral side. The differences in the results obtained by us and other researchers may arise from the limitations of the methods. The cervical spine was frozen, the dura mater examined after thawing and stored in 0.9% NaCl solution at 4 °C until it was placed in the holders of the testing machine. Perhaps the samples kept in the air during testing dried, which could have an impact on the results obtained. Only one article dealt with the marginal effect of freezing the dura mater on its mechanical properties. It is possible that the storage of cervical spine at temperatures below 0 °C has a greater influence on the mechanical properties than Runza et al. have shown. Those scientists, however, did not explain the origin of the samples, did not specify where the material had been taken from [8]. Other studies showed the viscoelastic properties of the dura mater of animal origin. They depend exclusively on the sample tensile direction – longitudinal or circumferential [7].

In this paper, a completely different approach to the problem under consideration has been proposed. Based on the structure of the spine and the loads transferred to every section it can be concluded that the spinal cord strain may vary despite the constant stress. Taking account of the fact that the characteristics of dura mater can vary depending on the vertebra's number and location of the dura mater in relation to the frontal plane (ventral or dorsal side), a detailed breakdown of the material, depending on the sampling site, was made. In these studies, a special attention has been given to the choice of the sampling places, remembering the position in relation to the frontal plane and the height of the vertebra, from which the material was prepared. The tensile test was performed in the circumferential and longitudinal directions relative to the cord axis, just as the tests of RUNZA et al. [8] and WILCOX et al. [7], [11].

The dura mater is composed of several tens of layers. Their number, however, is different at different heights of the vertebra, which is reflected in different mechanical properties [1], [4]. These assumptions have been confirmed by the results of our work.

The experimental analysis allows us to continue the research in the field of numerical analysis. Given the fact that the dura mater is composed of multiple layers of collagen and elastin fibres, it is possible to perform numerical tests based on the number of layers and their relative arrangement one to each other. Choosing the appropriate geometric parameters of the numerical model may lead to the model of the ideal mapping of the characteristics obtained experimentally. This model could be used in numerical tests of such a structure as the whole spinal cord.

Animal tissues could be obtained and preserved under optimal conditions, which, for obvious reasons, was not possible for human samples. We decided to use porcine spinal cord specimens. Additionally, as is shown in the literature, the porcine spinal cord should be not less suitable than human specimens in biomechanical testing [15], [16].

5. Conclusions

The properties of individual elements of the tissue structure, in this case, the dura mater, affect its behaviour under load and allow the non-linear stress–strain characteristics to be plotted.

The samples are divided into groups, depending on:

- the place of collection from all the cervical portion and breakdown of vertebrae from C1 to C7,
- the place of collection – from the dorsal or ventral side,
- the direction of tension – longitudinal (consistent with the long axis of the cord) and circumferential (peripheral).

This division enabled us to define the detailed mechanical properties of the dura mater with special consideration of the sample origin.

Based on the data obtained, the orthotropy of mechanical properties of the dura mater was confirmed. As expected, the mechanical properties depend both on the direction of the sample tension and on the sampling site. Therefore, it appears that the unification of the structure tested is a serious shortcoming of the traditional approach.

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