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Effect of dissection on the mechanical properties of human ascending aorta and human ascending aorta aneurysm

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Purpose: The aim of the presented work is to determine (i) mechanical properties of the ascending aorta wall (DAA) and the wall of the ascending aortic aneurysm (DAAA), in which spontaneous dissection resulting from the evolving disease occurred, and (ii) the strength of the interface between the layers in the above-mentioned vessels. *Methods:* The mechanical tests were divided into two steps. In the first step the mechanical properties of the of DAA and DAAA walls were examined on the basis of uniaxial stretching until rapture. In the next step the mechanical parameters of the interface between layers of DAA and DAAA walls were determined by the peeling test. *Results:* Higher values of tensile strength (σ_{max}) and Young's modulus (E) were obtained for the DAAA group, to which the dissecting wall of the ascending aortic aneurysm was classified. For circumferential samples, the difference between the DAAA and DAA groups was 39% in the case of tensile strength and 70% in the case of the Young's modulus. *Conclusions:* Summarizing, the studies performed showed that the dissection process is different in the case of the ascending aortic aneurysm wall and the ascending aorta wall. The wall of the ascending aortic aneurysm is more susceptible to dissection, as evidenced by lower values of the mechanical parameters of the interface between the intima and the media-adventitia complex. The obtained results of mechanical properties tests confirm that dissection and aneurysm should be treated as separate disease entities that may coexist with each other.

Key words: *aneurysm, dissection, ascending aorta, blood vessel wall*

1. Introduction

One of the diseases of the vascular system is dissection, interpreted as the loss of the vessel wall cohesion, as a result of which blood flows between the layers. This results in the creation of two channels for blood flow: a true channel corresponding to the lumen of the blood vessel and a false channel [24] between the layers. Dissection is indicated as one of the most serious complications associated with aortic wall aneurysm, which develops as a result of degradation of the main structural components, important from the point of view of transferring mechanical loads, i.e., collagen, elastin and smooth muscle cells. These changes result in a decrease in the ability of the blood vessel wall to actively shrink under the influence of blood pressure and impaired ability of the vessel wall to resiliently return to the shape associated with diastolic pressure. During further development of the aneurysm, there is a balance disturbance between degradation and synthesis of collagen fibers. The increased degradation of these fibers, in particular collagen type I and III, leads to a reduction in the tensile strength of the vessel wall and is indicated as the main cause of the aneurysm rupture.

Dissection is a common complication associated with the development of the aneurysm, which is why the phenomena that can be indicated as potential initiating dissection are structural changes typical for the development of the aortic aneurysm. Clinical practice

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indicates that in most cases dissection is initiated within the middle layer, which, according to the current state of knowledge, is the result of "degeneration" of this layer, which involves structural changes in the extracellular matrix of the aortic wall, i.e., change in smooth muscle cell concentration and quantitative and qualitative changes of collagen and elastin fibers [11].

An essential element that determines the resistance to dissection are the mechanical properties of a vessel wall, individual layers, and in particular the mechanical properties of the connection between the layers. The authors who were the first to undertake a mechanical description of the dissection were Sommer et al. [19] and Tong et.al. [20]. Based on the research carried out, the authors showed that a interface between the intima and the media-adventitia complex is more prone to dissection. What is more, they showed that the mechanical properties of the interface between the vessel wall layers are directional and are higher in the longitudinal direction. Studies carried out by Kozuń [9], [10] have shown that the second stage of atherosclerotic disease leads to a decrease in the mechanical properties of the interface between the layers, and the least resistance to dissection occurs in the IV stage of atherosclerosis, which results, inter alia, from the formation of atherosclerotic plaque between the layers. The problem of aneurysms' dissection was undertaken by Pasta et al. [14]. The authors showed that (i) the aneurysm's wall is more susceptible to dissection and rupture, (ii) aneurysmal disease leads to isotropic behavior due to a more disorganized microstructure.

Summarizing, although dissection is an important problem from a medical, social and scientific point of view, and the number of people suffering from it is constantly increasing, the etiology and process of development of dissection have not been described so far. Predisposing factors include diet, genetic factors or a sedentary lifestyle that contributes to artery wall remodeling [4]. Most of the work related to the analysis of the dissection process concerns biological factors that can have a significant impact on the formation and development of this disease. These factors include qualitative and quantitative changes of elastin fibers, collagen and smooth muscle cells in the middle aortic wall layer, as well as the importance of increasing proteoglycans content in this layer. These structural elements determine passive and active mechanical properties of the vessel wall, and degenerative processes may lead to changes in the mechanical properties of the aortic wall, its individual layers and the connection between them, and thus, loss of integrity of the entire wall.

It should be emphasized, however, that there are no studies in the literature that would refer to mechanical parameters' determination of the aortic wall or the wall of the aortic aneurysm, in which dissection has already occurred. So far, none of the aortic dissections presented in the literature occurred spontaneously. Therefore, the results of the studies presented in this work only reflect the mechanical properties of the interface between the aortic wall layers or the aortic aneurysm wall, but do not provide sufficient knowledge necessary to describe the mechanism of dissection. Conducting analyzes on the research material of the dissection of the ascending aortic wall and dissecting wall of the ascending aortic aneurysm is important regarding explaining the mechanism of delamination, because, according to clinical data, acute aortic dissection is more common among patients who did not have an aneurysm earlier. This indicates that the aneurysm and dissection should be treated as two separate disease entities that may coexist with each other.

Therefore, the aim of the presented work is to determine (i) mechanical properties of the ascending aorta wall and the wall of the ascending aortic aneurysm, in which spontaneous dissection resulting from the evolving disease occurred, and (ii) the strength of the interface between the layers in the above-mentioned vessels.

2. Materials and methods

2.1. Materials

The research material was the ascending aorta ($n = 5$) and the ascending aorta aneurysm ($n = 4$). The vessels were collected during cardiac surgery performed at the Heart Surgery Clinic of the Wroclaw Medical University in patients who underwent replacement of the ascending aorta segment due to its dissection or due to the occurrence of a dissected aneurysm (Bioethical Commission approval number: KB-14/2019). The collected material was divided into two research groups: a DAA group, to which the dissecting ascending aorta was classified, and a DAAA group, which included dissecting ascending aorta aneurysm. The DAA group was a control group. All samples were taken from men (average age: 48 ± 12). In each case, the dissection was spontaneous (type A dissection according to Stanford classification) and occurred between the interface between the inner layer and the complex of the middle and outer layers. After collection, the test

material was immersed in physiological saline, frozen and stored until measurements were taken at a temperature of about –10 °C. Then the material was transferred to the Chair of Biomedical Engineering, Mechatronics and Mechanics Theory of Wroclaw University of Science and Technology in order to carry out mechanical properties' tests.

Preparation of samples for mechanical properties testing

Approximately 1.5 hours prior to testing, the samples were thawed at room temperature, i.e., 23 °C. Rectangular specimens with the length of 25 mm and width of 5 mm were cut out from each vessel and that was determined by the size of the punch. The number of specimens cut out from a single samples depended on its size and ranged from 3 to 6. The specimens were cut out in two directions, i.e., in the longitudinal and circumferential direction in relation to the axis of the main blood vessel. A detailed methodology for specimen preparation is described in [9]. Finally, 44 specimens were prepared for the mechanical properties tests, of which 33 specimens were cut out in a circumferential direction and 11 in the longitudinal direction (Table 1). During the specimen cutting, the presence of spontaneous dissection of the vessel wall between the intima and the media-adventitia complex (I-MA) was found. On the basis of macroscopic assessment, pathological changes typical for the development of atherosclerosis, i.e., calcium deposits characteristic for calcification processes in the blood vessel wall were also found [8].

Table 1. Number of samples prepared for mechanical properties testing

Research group	No. of samples	
	circumferential direction	longitudinal direction
DAA	17	6
DAAA	16	5
Summa	33	11

DAA – dissected ascending aorta,
DAAA – dissected asceding aorta aneurysm.

Until the mechanical properties were tested, the samples were stored in physiological saline (0.9% NaCl).

2.2. Methods

Examination of mechanical properties of DAA and DAAA walls.

The mechanical properties of the specimens were determined based on the uniaxial tensile test carried out under quasi-static conditions using the MTS Synergie 100 testing machine. The length of the specimen section was 20 mm. The test consisted of two stages, i.e., prestreching (i) and uniaxial stretching test (ii). Prestreching consisted of three times loading and relieving the sample of 10% of the length of its measuring section (2 mm). At the end of this stage, the proper uniaxial stretching test started until the specimen was broken. This test is widely used in the literature in the study of mechanical properties of soft tissues [7]. Both steps were carried out at a loading speed of 2 mm/min. During the tests, changes in force in the displacement function were recorded.

Based on the results obtained, the examined tissues were described as incompressible hyperslastic material which undergoes large deformations. For each sample, the normal component of the Green strain tensor and the normal component of the Cauchy stress were calculated. On the basis of the obtained stress–strain curves mechanical parameters were determined, i.e., tensile strength (σ_{max}) – defined as the maximum stress, maximum strain (ε_{max}) obtained at the point corresponding to the strength and the maximum tangent modulus of elasticity (E). The maximum elastic modulus was taken from the maximum slope prior to failure.

Research on mechanical properties of the interface between DAA and DAAA wall layers

The mechanical properties of the interface between the layers of the tested specimens were determined on the basis of a peeling test at a 2 mm/min load speed, using the MTS Synergie 100 testing machine. The load was applied perpendicularly to a plane of the specimen dissection (T-peeling test configuration) [9] [10]. This test stage was only carried out for circumferential specimens. Due to the fact that in each of the investigated specimens, dissection occurred spontaneously between the intima and the media-adventitia complex (I-MA), only this combination was tested for mechanical properties. Before testing, each of the two parts of the spontaneously dissected specimen (Fig. 1A) were mounted to the holders of the testing machine, as shown in Fig. 1B. It should be emphasized that spontaneous dissection occurred only in the initial section of the specimen.

Based on the carried out tests, the dependence of force on displacement was obtained, and then the width of the specimen was taken into the force/width-displacement curves and the mechanical parameters such as maximum force (F_{max}), stiffness coefficient of the tested interface (k), mean value of force obtained

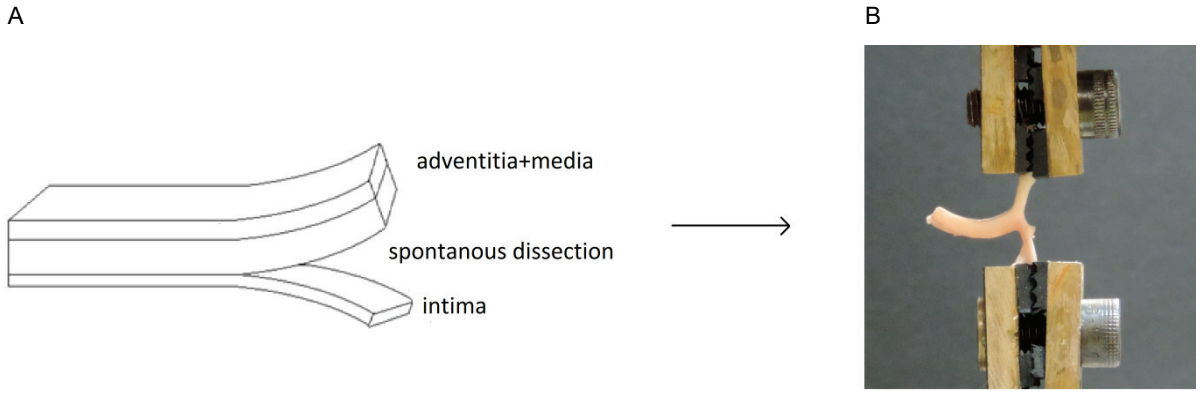

Fig. 1. Schematic illustration of the peeling test: A) Diagram of a delaminated specimen. In each case, dissection occurred spontaneously between the intima and media-adventitia complex (I-MA); B) The specimen was mounted in the holders of the testing machine. The load was applied perpendicularly to the plane of dissection (T-peeling test configuration)

during dissection (F_{aver}) and dispersed energy during dissection (W) [9], [10], [19], [20].

Both stages of the mechanical properties tests were carried out at constant temperature (24 °C) and the specimens were wetted with saline solution. Saline (40÷50 µl) was given per specimen 4 times a minute, from the upper handles.

Statistics

The obtained values of mechanical parameters were treated as random variables, for which statistical analysis was performed. This analysis included; (i) verification of the distribution normality of the analyzed variables and (ii) verification of the statistical significance of the differences obtained between the measurement groups and the studied directions. The distribution normality of the analyzed variables was tested using the Shapiro–Wilk test (α = 0.05). Statistical significance between the studied groups was verified using the student's t-test. The values of mechanical parameters are presented as mean and standard deviation (X_{mean} ± SD).

3. Results

Research of mechanical properties of DAA and DAAA

Regardless of the analyzed direction, higher values of tensile strength (σ_{max}) and Young's modulus (E) were obtained for the DAAA group, to which the dissecting wall of the ascending aortic aneurysm was classified. For circumferential samples, the difference between the DAAA and DAA groups was 39% in the case of tensile strength and 70% in the case of the Young's modulus. These differences were statistically significant (α = 0.05). For longitudinal samples, the mean values of all analyzed mechanical parameters were also higher in the case of the DAAA group, although the differences obtained between the research groups are not statistically significant.

In the case of dissecting ascending aorta (DAA group), higher values of tensile strength and maximum strain were obtained for circumferential specimens. These values were respectively: σ_{max} = 0.141

Table 2. Mean values of mechanical parameters, i.e., tensile strength (σ_{max}), maximum strain (ε_{max}) and Young's modulus obtained for the dissecting ascending aorta (DAA) wall and the dissecting wall of the ascending aortic aneurysm (DAAA) in two directions

	DAA group		DAAA group	
	peripheral direction	longitudinal direction	peripheral direction	longitudinal direction
σ_{max} [MPa]	0.141* ± 0,060	0.055** ± 0,010	0.231* ± 0.100	0.496** ± 0.180
ε_{max} [–]	0.076 ± 0,045	0.056** ± 0,020	0.065 ± 0.020	0.149** ± 0.029
E [MPa]	1.13*** ±0.29	1.80*** ± 0.78	3.75* ± 1.02	5.27** ± 2.53

*, **, *** statistically significant differences (α = 0.05).

± 0.060 MPa and ε_{max} = 0.076 ± 0.045, and were higher by 60% and 26% compared to longitudinal specimens. These differences are not statistically significant. The longitudinal specimens were characterized by the higher value of Youngs modulus (E = 1.8 ± 0.8 MPa), while for circumferential specimens the value of this parameter was E = 1.1 ± 0.3 MPa. In the case of the Young's modulus, the differences between the directions are statistically significant (α = 0.05). In the case of dissecting ascending aortic aneurysm wall, the mean values of all analyzed mechanical parameters were higher for longitudinal specimens, although the differences between the two directions are not statistically significant (α = 0.05).

Tests of mechanical properties of the interface between DAA and DAAA layers

In Fig. 2 the delamination curves obtained for the DAA and DAAA groups are shown. In both cases, the jagged plateau region corresponding to the propagation of dissection characterizes oscillations in obtained values of force. The oscillation is often referred to in the rubber mechanics as "unstable" or stick-slip "tearing". In the case of dissection of the aneurysm wall, oscillations in the dissection values obtained in the process are, however, lower compared to the oscillations obtained for the dissection of the aortic wall. For the dissecting wall of the aneurysm, the difference between the maximal and the minimum force value ranges from 20% to 34%, whereas for the dissecting aortic wall the range is from 70% to 82%.

Higher values of all the analyzed mechanical parameters of the connection between the inner layer and the middle and outer layers were obtained in the case of the delaminating wall of the ascending aorta (DAA group). These differences are: 54% for maximum force, 68% for stiffness of the tested connection, 36% for medium strength and 49% for dispersed energy during dissection. In the case of stiffness and medium strength, the differences obtained are not statistically significant (α = 0.05) (Table 3).

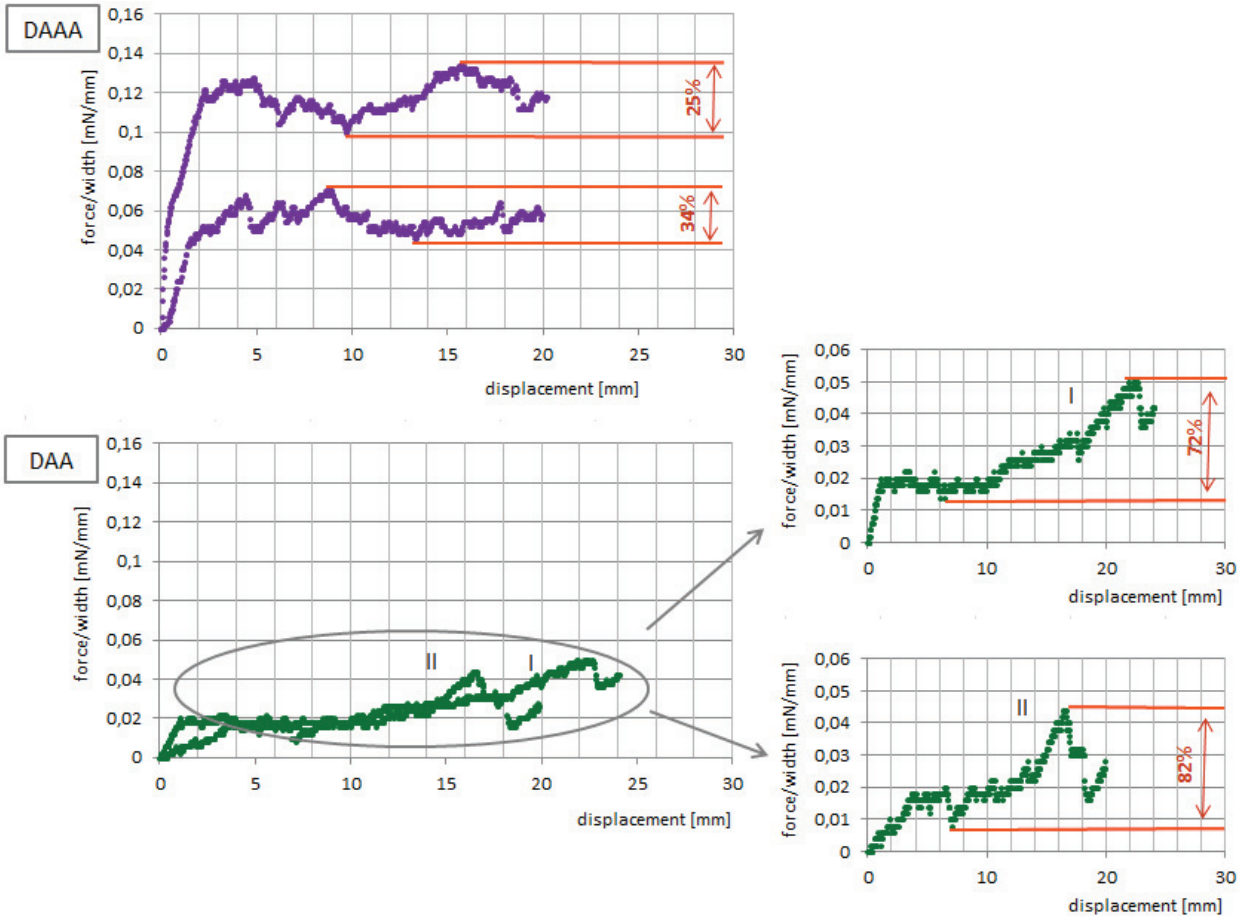

Fig. 2. Sample force/width *vs.* displacement curves obtained for the I-MA interface of the dissected ascending aortic aneurysm (DAAA) and dissected ascending aorta (DAA). In each case, the region corresponding to the propagation of dissection is characterized by oscillations in the obtained values of force. Therefore, for all curves, the difference between maximum and minimum peak of force values was determined

Table 3. Mean values of mechanical parameters, i.e., maximum force (F_{max}), stiffness coefficient (k), mean force (F_{aver}) and dissipation energy (W) obtained during dissection of the interface between the intima and the media-adventitia complex the wall of the ascending aorta (DAA) and the wall of the ascending aorta aneurysm (DAAA)

	DAA group	DAAA group
F_{max} [mN/mm]	118 ± 36*	54 ± 26*
k [N/mm]	0.77 ± 0.31	0.25 ± 0.09
F_{aver} [mN/mm]	33 ± 16	21 ± 9
W [mJ/cm^2]	43 ± 16*	22 ± 11*

* statistically differences significant ($\alpha = 0.05$).

4. Discussion

The presented work concerns the problem of dissection of the blood vessel wall which is now more and more often treated in clinical practice as a separate disease entity. The dissecting aneurysms of ascending aorta and dissecting wall of the ascending aorta were examined. In each case, the mechanical parameters of the entire vessel wall (uniaxial tensile test) and interface between the intima and the media-adventitia complex (I-MA) (peeling test) were determined. It should be emphasized that dissection occurred spontaneously between the abovementioned layers, and was not forced for the needs of research, which has not been presented in the literature so far. On the basis of the determined mechanical parameters, the dissection properties have been characterized by the underlying aortic dissection.

In the case of dissecting the aneurysm wall as well as the dissecting aortic wall, the delemination curves revealed an oscilation (Fig. 2). This indicates that in both cases dissection does not propagate at a steady rate. It arrests and reinitiates at somewhat irregular intervals. A larger range of oscillations obtained in the case of a dissecting aortic wall indicates that in this case the necessary force to drive the dissection varies more widely. This is confirmed by the results obtained by Pasta et. al. [14]. According to the authors, such course of the curve results from damage to a large number of elastin fibers during propagation of dissection and their high extensibility during dissection testing. Consequently, this leads to an increase in the peel test magnitude [14]. The obtained results showed that the wall of the ascending aortic aneurysm was characterized by a lower value of the maximum force and energy dispersed during the dissection process of the examined interface, therefore, patients with ascending aortic aneurysm are more prone to dissection. In the case of dissecting the ascending aortic aneurysm wall, higher tensile strength values were obtained, which, in turn, indicates that a greater risk of disruption of the vessel wall continuity occurs in the case of the dissecting wall of the ascending aorta. However, in the performed studies, the diameter of the aneurysm was not taken into account, which makes it impossible to determine the degree of its advancement. However, based on the obtained values of mechanical parameters, in particular tensile strength and Young's modulus, it can be concluded that the tested specimens showed the second stage of aneurysm development. Directional analysis of mechanical properties showed no statistically significant differences between the values of mechanical parameters obtained for circumferential and longitudinal specimens of the dissecting wall of the ascending aortic aneurysm. In the case of dissecting the aortic wall, only in the case of the Young's modulus, the differences between the circumferential and longitudinal directions are statistically significant. The obtained results showed that both in the case of the ascending aortic aneurysm wall as well as the ascending aorta wall, the dissection leads to isotropic behavior of the blood vessel wall. Similar conclusions were provided by Pasta et al. [14] for the ascending aortic aneurysm wall. However, the mechanical parameters obtained by him are higher than the values obtained in the presented work. This may result from a different degree of severity of the disease process and, what is associated with this, another degree of severity of structural changes in the vessel wall.

The change in the mechanical properties of both the dissected wall of the ascending aorta and the dissected wall of the ascending aortic aneurysm, as well as the interface between the layers in both vessels result from degenerative changes of the main structural components, which is associated with the development of the disease process. The first phenomenon occurring in the process of aneurysm development are, according to the literature, structural changes in the middle aortic wall layer, which consist in reduction of the concentration of elastin fibers (up to 63%–92% [20]) and smooth muscle cells (up to 74.5% [12]). There are, however, opposing theories about the etiology and mechanism of the dissection development in literature. According to Tsamis [21], the quantitative and qualitative changes of elastin fibers in the middle layer are responsible for the formation of dissection. Sariola et al. [17] emphasizes that the formation and development of

the disease is caused by the increase in the collagen fiber content in the middle layer, which leads to fibrosis of the aortic wall and thus, the increase of its stiffness, which increases the susceptibility to dissection. Borges et al. [3] also emphasizes the importance of collagen fibers in the dissection process, although, according to the author, before the onset of the disease, the content of these structural components decreases in the middle layer. This theory is confirmed by the work of Wu et al. [25]. Roccabianca et al. [15] presents a new approach to the initiation and development of dissection. The authors suggest that proteoglycans are responsible for the development of dissection. In a well-shaped middle layer, the content of these compounds ranges from 2% to 5%. Increasing the content of proteoglycans above 5% and their accumulation in the middle layer leads to the formation of cysts causing stress concentration and local decrease of the mechanical strength of this layer, which may cause integrity loss of the middle layer and initiate the formation of dissection [15].

5. Conclusions

The studies performed showed that the dissection process is different in the case of the ascending aortic aneurysm wall and the ascending aorta wall. The wall of the ascending aortic aneurysm is more susceptible to dissection, as evidenced by lower values of the mechanical parameters of the interface between the intima and the media-adventitia complex. The dissecting ascending aorta wall, in turn, is more vulnerable to disruption of continuity, as indicated by lower failure properties. Indication of the reasons for this type of changes in mechanical properties requires a wide structural analysis of dissecting vessels, i.e., the wall of dissecting the ascending aorta aneurysm as well as the wall of the dissecting ascending aorta. These studies have not been presented in this work, which is a limitation of the presented analysis. Referring to the currently existing theories related to the development of dissection, structural studies should include quantitative and qualitative analyzes of the main structural components of the blood vessel wall, i.e., collagen, elastin and smooth muscle fibers, as well as the proteoglycans content in the middle layer. The obtained results of mechanical properties tests confirm the latest literature reports that dissection and aneurysm should be treated as separate disease entities that may coexist with each other.

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