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Features of the ratio of collagen I and III in patients with postoperative ventral hernias

Modern herniology presents innovational field of surgery. Among the causes of hernia development the disturbance of collagen metabolism plays a great role, and leads to heterogeneous maturation of the connective tissue and disorder of its structural characteristics. Investigation of the collagen contents of the connective tissue with the help of polarization microscopy reveals Collagen Type I to Type III ratio in skin and aponeurosis in patients with and without hernia disease. Presented investigation makes prognosis and prophylactics of hernia disease possible in early post operation period.

Keywords: Collagen Type I and Type III, post operation hernia, hernia disease, polarization microscopy.

Introduction. Nowadays treatment of abdominal anterior wall hernia disease is the actual problem of surgery. Among all surgical operations hernioplastics presents up to 32%, ventral hernias (VH) contribute 26%, 35% of the latter ones are urgently operated due to squeezing. 10 – 14% of all laparotomies are complicated in the late post operation period with VH of abdominal anterior wall. According to the opinion of certain authors [1,2,3,4,7] the rate of mediate VH continues to increase, presenting from 57 to 83% of whole quantity of anterior wall of the abdomen hernias. Despite of wide application of modern surgical techniques in clinical practice, the results of VH treatment cannot be accepted as satisfactory ones. It's well known that collagen metabolism disorders lead to the weakness of the connective tissue, and later to VH development. Collagen metabolism



investigation is possible with the help of monoclonal antibodies using, or special staining and polarization microscopy [1,5].

Most effective methods of surgical treatment of patients with VH are those with the synthetic materials using for plastics [4,6,7,8,9]. Although using of plastics without intention with application of synthetic materials has led to improvement of hernias treatment results, still there are certain problems of the select of the type of synthetic endoprothesis and of the method of surgical treatment [3,7]. Thus it is reasonable to forecast hernia disease and perform prophylactics of complications in early post operation and rehabilitation periods.

The purpose of investigation.

To estimate Collagen Type I to Collagen Type III ratio and Collagen contents in the skin and aponeurosis in patients with and without VH.

Tasks of investigation.

To confirm differences of Collagen Types Ratio (TC) in skin and aponeurosis in patients with and without VH.

To reveal correlation connections of TC ratio in skin and aponeurosis in patients with and without

To consider the information level of polarization microscopy for possible verification of hernia disease.

Materials and methods

In the presented work there were used data of examination and treatment of patients with uncomplicated VH, who were treated from 2010 till 2012 in clinics of surgical diseases № 1 of Kursk State Medical University, based in the Kursk Regional hospital.

95 patients were examined, and were divided into examinating and control groups. There were 30 males (31,6%), and 65 (68,4%) were females. Examinating group included 46 patients, among them – 37 (80,4%) were the patients with VH, 7 (18,9%) suffered from small hernias, 19 (51,4%) had middle ones, 7 (18,9%) had big ones, and 4 (10,8%) suffered from giant hernias. The rest 9 (19,6%) were patients with umbilical hernias, among them 4 (44,4%) patients suffered from hernias of middle sizes, and 5 (55,5%) – from small ones. There were 11 males (23, 9%), whose average age was 54 ± 9.9 , and females -35 (76,1%) of 56.8 ± 11.2 . The control group was presented by 49 patients without hernias and without clinical signs of connective tissue weakness. patients were treated in the department of general surgery of Kursk regional hospital with using of laparotomy, without hernias (for instance, after laparotomy cholecystectomy). Males were 19 (38,8%) with average age 62,8±13,3. Females were 30 (61,2%) with average age 54,5±13,8. Both



groups included two series of investigation – of skin and aponeurosis.

In laparotomy of both groups of patients skin and aponeurosis were taken. Tissues taken in operation were examined on qualitative contents of connective tissue collagen fibers. The cuts of tissues were stained with Sirius Red and examined in ordinary and polarization lights with the help of polarization microscope Altami Polar 2, magnification x100, x250 и x400, x630. Photographing of micro specimens were fulfilled with using of digital ocular camera Altami 3 Mpx., there were taken pictures of 10 fields of view with different magnification.

Estimation of Collagen Types ratio (TC) was based on differences of color scale, characteristic for each type of collagen: Type I Collagen is red, Type III Collagen is green. Estimation of Collagen Type I to Collagen Type III ratio was performed with using of program complex Altami Studio 3.0 and ImageJ 1,47a, on the base of examination of color histogram of the certain focus in each field of view. Determination of color spectrum was carried out on the base of histogram of each color. Absolute indexes of red and green colors, got with the help of visualprogram complex for each field of view, were converted into comparative ones with accounting of standard deviation. Then it was accounted the level of TC ratio. Analysis of the results was performed with the help of embedded computer functions of supplement Microsoft Exel-2010, Statistica 6.0.

Results and discussion

Collagen Type I to Type III ratio in the skin of 1 group patients is 1,06±0,1, because of this green color predominates in spectrum. Examination of aponeurosis histological specimens showed that Collagen Type I contents is 52,35±2,97%, and Collagen Type III contributes 47,65±2,97% (Picture 1). Collagen Type I to Type III ratio in aponeurosis of patients with hernias is 1,11±0,15, this is much less than in patients without VH. Examination of the skin histological specimens of the 2 group patients (control) revealed that the contents of Collagen Type I is 69,07±2,66%, and Collagen Type III contents is 30,93±2,66%. Collagen Type I to Type III ratio in skin is 2,26±0,29. Polarization microscopy of the histological specimens of the aponeurosis of control group patients showed the contents of Collagen Type I is 69,11±2,68%, and Collagen Type III is 30,89±2,68% (Picture 2). Index of Collagen Type I to Type III ratio is 2,27±0,3, which is more than in patients of examinating group.

Thus, patients with hernias present reliable significant lowing of Collagen ratio both in skin and in aponeurosis in comparison with patients without hernias. The contents of Collagen Type I in the skin of patients with small hernias is 51,46± 1,34%, and Collagen Type III is 48,531±1,34%. Collagen Type I and Type III in aponeurosis of patients with hernias of middle sizes is 51,4±2,32%

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and 48,6±2,32%.

In examination of skin specimens of patients with big hernias we revealed, that contents of Collagen Type I is 50,7±0,8% and Collagen Type III is 49,3±0,83%. Within examinating group there were 4 patients with giant hernias, in whom average content of Collagen Type I is 50,4±0,82%, and of Type III is 49,6±0,82%. Comparative analysis of the results showed that within control and examinating groups there are no significant differences between series (comparison of the results of skin and aponeurosis examination). Due to this corresponding conformity of Collagen Types ratio in skin and aponeurosis was revealed.

Thus, index of Collagen Types ratio in skin and aponeurosis in patients of each group is the same. Examination of Collagen Type I and Type III ratio in skin of patients with and without hernias revealed significant differences of collagen contents. Collagen Types ratio in patients without hernias is 2,26, whereas in patients with hernias it is significantly lower - 1,06 ($p \le 0,001$).

Collagen Types ratio in aponeurosis of patients without hernias is 2,27, but in patients with hernias it is 1,12. Collagen Type I and Type III ratio in the skin and aponeurosis of patients without hernias is 2,26 and 2,27 correspondently, it shows absence of significant differences and presence of strong correlation association (Spirman's coefficient is 0,71) (Picture 3).

In patients with hernias Collagen Type I to Collagen Type III ratio in skin is 1,06, and in aponeurosis it is 1,12, it also presents absence of significant differences and presence of strong correlation association (Picture 4). Comparison of Collagen Type I and Collagen Type III indexes in examinating and control groups showed significant differences between them (p≤0,001). Decrease of Collagen Type I and Type III percentage is one of the causes of post operation hernias development.

Due to information above, we can consider, that changes of Collagen contents of the connective tissue precisely decrease of Collagen fibers Type I to Type III ratio is one of the factors of VH etiology and pathogenesis. Collagen Types ratio in the contents of the connective tissue in skin and aponeurosis has strong correlation association both in control and examinating groups. Usage of the presented method of Collagen Types indexes estimation in the connective tissue of the anterior wall of abdomen gives surgeons the opportunity to choose well-founded method of VH treatment.

Conclusions:

Collagen Types ratio in aponeurosis of patients with VH is significantly less, than in patients without VH.

Collagen Types ratio in skin and aponeurosis has strong correlation association in control and



examinating groups.

Polarization microscopy has high information level in investigation of Collagen Types ratio disorders in the structures of the anterior wall of abdomen.

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Table 1 Characteristics of Collagen Type I and Type III in skin of patients of examinating and control groups

	Series 1 – Skin (N=95)		
Groups	Collagen Type I	Collagen Type III	Collagen Type I and
Groups	Conagen Type I	Conagen Type III	Type III ratio
Examinating	51,48±1,83*	48,52±1,83*	1,06±0,1 1
Patients with VH (N= 46)	**	**	
Control	60.07+2.66*	20.02 2.66*	2.26+0.201
Patients without VH	69,07±2,66*	30,93±2,66*	2,26±0,29 ¹
(NI— 40)	**	**	
(N=49)			

^{*-} p≤0,001, comparison of indexes between groups of patients.

Characteristics of Collagen Type I and Type III in aponeurosis of patients of examination and control groups

*- p \(\) 0,001, comparison of indexes between groups of patients.

p_0,001, comparison of macros between groups of patients.				
	Series 2 – Aponeurosis (N=73)			
	Collagen Type I	Collagen Type III	Collagen Type I and	
			Type III ratio	
Examinating	52,35±2,97*	47,65±2,97*	1,11±0,15 ¹	
Patients with VH (N= 46)	**	**	1,11±0,13	
Control	(0.11+ 3 .60*	20.00+2.00*		
Patients without VH	69,11±2,68*	30,89±2,68*	$2,27\pm0,3^{-1}$	
	**	**		
(N=49)				

^{** -} $p \ge 0.5$, no significant differences between series in group.

 $^{^{1}}$ – Spirman's coefficient = 0,71, comparison of indexes between series.



- ** $p \ge 0.5$, no significant differences between series in group.
- 1 Spirman's coefficient = 0,71, comparison of indexes between series.



Fig. 1. Microscopic picture of the section of aponeurosis in patient with VH. Polarization microscopy. Sirius Red. X400.

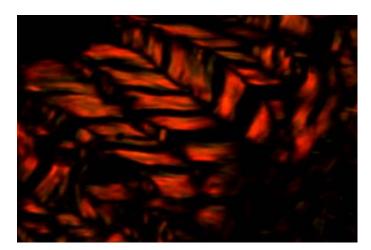


Fig. 2. Microscopic picture of the section of aponeurosis in patient without VH. Polarization microscopy. Sirius Red. X400.



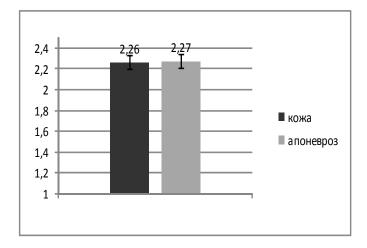


Fig. 3. Collagen Type I to Type III ratio in skin and aponeurosis of patients without VH.

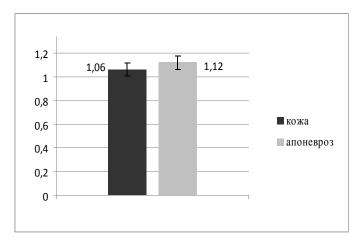


Fig.4. Collagen Type I to Type III ratio in skin and aponeurosis of patients with VH.