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Biochemical Study of Collagen in Adult Groin Hernias

Alain Pans, M.D., Ph.D.,* Adelin Albert, Ph.D.,† Charles M. Lapière, M.D., Ph.D.,‡ and Betty Nusgens, Ph.D.‡

** University Department of Abdominal Surgery, Clinique A. Renard, 1, rue A. Renard, 4040 Herstal, Belgium;*
†Department of Biostatistics; and ‡Laboratory of Connective Tissues Biology, CHU Sart Tilman, 4000 Liège, Belgium

Abstract

Background. Previous works have suggested that a defect in collagen fiber structure may play a role in inguinal hernia formation. These studies focused mainly on the rectus sheath or the skin, while only few reports dealt with the transversalis fascia. According to these findings and to our previous biomechanical and histological studies suggesting that a connective tissue pathology could play a role in the genesis of groin hernias, we performed a biochemical investigation of the collagen in the transversalis fascia and rectus sheath.

Materials and Methods. The samples were collected from 40 adult patients with uni- or bilateral hernias and from 20 control subjects without hernia (autopsies and organ donors). A constant area of tissue was taken by using a calibrator. The wet and dry weights per 100 mm^2 were determined and the total collagen concentration as well as its sequential extractibility in NaCl, acetic acid, and pepsin was measured. The ratios of α_1/α_2 chains (I) and of type I/III collagen were assessed by polyacrylamide gel electrophoresis.

Results. Samples collected in the control and patient sheaths showed an increased wet weight per 100 mm^2 in the patients. The wet and dry weights per unit area were increased in the patient fascias. The collagen concentration was increased in the indirect hernias. The fascias from the direct hernias (DH) presented a significantly increased collagen extractibility after pepsin digestion (5.6%), when compared to the control fascias (2.6%). The extractibility was 3.4% in the non-herniated (NH) sides. The qualitative study (ratios α_1/α_2 (I) and I/III collagen) showed no difference between the fascia groups.

Conclusions. The significant increase of collagen extractibility with pepsin in the DH fascias and at a lesser degree in the NH fascias suggests that molecular alterations of collagen could be involved in the genesis of groin hernias. This connective tissue pathology would express preferentially its effects in the inguinal region, since we have observed no major difference between the rectus sheaths of controls and those of patients,

Keywords : inguinal hernia ; etiology ; collagen ; biochemistry.

INTRODUCTION

Hernia repair is the most frequent operation, after appendectomy, in the United States [1]. The basic mechanisms of hernia formation, however, remain mostly unknown. Classically, inguinal hernias are considered the result of a multifactorial process linking predisposing anatomical and dynamic factors, such as enlargement of the weak inguinal area and increased intra-abdominal pressure [2]. In addition, histobiochemical factors, which have received little attention so far, are likely to play a key role in the genesis of groin hernias.

In light of the works of Peacock and Madden [3] and that of Wagh *et al.* [4-7], it appeared that hernia formation might be based on collagen metabolism anomalies. Hence, inguinal hernia could be considered a local manifestation of a more generalized collagen pathology. These studies about collagen disorder focused mainly on the rectus sheath [4 - 6] or the skin [8], while only few reports dealt with the transversalis fascia [9]. In this report [9], a slight decrease in lysine hydroxylation and a significant increase in the expression of matrix degrading enzyme, the matrix metalloproteinase-2, were detected in fascias from direct hernia. Our previous biomechanical and histological studies [10, 11] performed on the transversalis fascia and the rectus sheath in patients with inguinal hernia suggested that a connective tissue pathology could play a role in the genesis of groin hernias. In this work, we performed biochemical investigations of the collagen network in the transversalis fascia and the rectus sheath.

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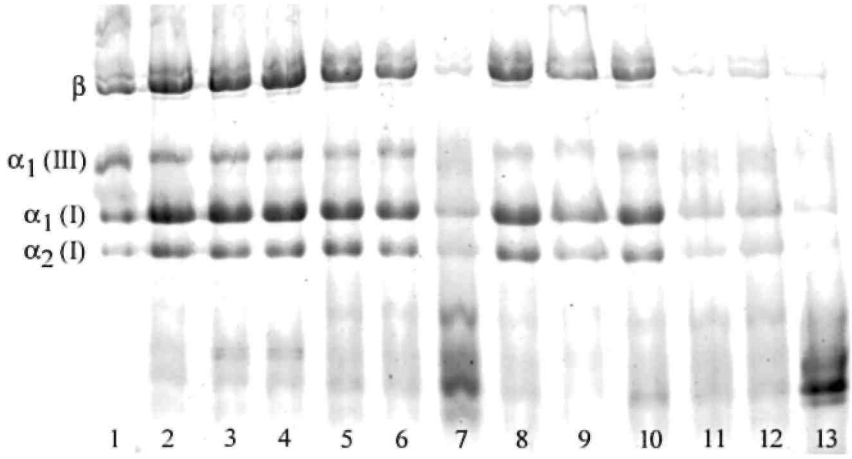
FIG. 1. *Example of collagen electrophoresis in SDS-polyacrylamide gel. Lane 1: control collagen from human skin. Lanes 2, 5, 8: patient rectus sheaths. Lanes 3, 6, 9: fascias from the nonherniated side. Lane 4: fascia from femoral hernia. Lanes 7, 10: fascias from indirect hernia. Lanes 11, 12: left and right control rectus sheaths (autopsy). Lane 13: left control fascia (autopsy).*



METHODS

This study was approved by the Institutional Ethics Committee of the University Hospital. In our institution, groin hernias are generally treated through a midline, preperitoneal approach, with insertion of a mesh on each side, in accordance with unilateral or bilateral Stoppa's procedure [12, 13]. A bilateral repair was carried out in all patients. Either the hernia was bilateral or the prophylactic repair was decided preoperatively with the patient's consent, because of the high incidence of clinically asymptomatic, controlateral hernia and bilateral disease during a patient's lifetime [14].

Once the preperitoneal dissection was accomplished, adequate exposure of the posterior wall of the inguinal canal was obtained. Biopsies of a constant surface area were taken with a specially designed area gauge from the transversalis fascia (13 × 18 mm), on the left and right sides as well as from the left anterior rectus sheath (10 × 20 mm). In the control group, left and right tissue samples were taken using the same procedure from autopsies within 24 h of death (n = 10) and organ donors (n = 7). A sample of the left rectus sheath alone was taken from three surgical patients without hernia. In these three surgical patients, the rectus sheath only was sampled because removal of a part of the posterior inguinal wall could conceivably cause an inguinal hernia.

Patients receiving steroid therapy and those who had previous subumbilical midline laparotomy or inguinal hernia repair were not included in the study. The same criteria were applied to control subjects. In addition, subjects with a history of recent abdominal surgery or abdominal aortic aneurysm were not included.

Measurement of Collagen Concentration and Extra ctibility

Each fresh sample was weighed (wet weight) and stored at -20°C. The thawed tissue specimens were cleaned of remaining blood with distilled water, lyophilized, and weighed. Dried tissue samples were crushed in liquid nitrogen and again lyophilized. Two aliquots were weighed, one for measuring total collagen concentration after 6 NHC1 hydrolysis according to the technique of Bergman and Loxley [15], while the second one was used for measuring collagen extractibility. Aliquots of dry powdered tissue were sequentially extracted for 48 h with 1 ml neutral buffered saline solution containing proteases inhibitors (1 M NaCl, 0.05 M Tris-hydroxymethyl-aminomethane, 0.02 M EDTA, 0.1 mM phenylmethanesulfonyl fluoride and 0.5 mM *N*-ethylmaleimide), then for 24 h with 1 ml 0.5 M HAc brought to pH 2.0 with HC1, and finally with 1 ml of a 100 µg/ml pepsin solution in 0.5 M HAc, pH 2.0. Extracts were collected by centrifugation at 7000 rpm for 30 min. All procedures were performed at 4°C. The collagen content of each extract was determined by hydroxyproline measurements according to the technique of Bergman and Loxley [15].

Collagen Electrophoresis

An aliquot of the pepsin extract was neutralized with 1 M Tris base and brought to 33% ethanol in the cold. The precipitated collagen was collected by centrifugation, lyophilized, and subjected to SDS-PAGE according to the technique of Laemmli [16], with a late reduction to discriminate type I and type III collagens. After staining with Coomassie blue, the intensity of the bands was recorded with a LKB Ultroscan XL LASER densitometer (Fig. 1). The ratio of α_1 and α_2 chains of type I collagen was calculated. The percentage of type I and type III collagens and of β chains was measured.

Statistical Analysis

Results are expressed as means ± SD. Statistical analysis was performed by chi-square test for qualitative findings, analysis of variance, Mann-Whitney test, and Wilcoxon test (paired data) for quantitative variables. The simultaneous effect of age, sex, and category of the tissue samples on the biochemical variables was studied by general linear model. Results were considered to be significant at the 5% critical level ($P \leq 0.05$).

RESULTS

The present series involved 40 patients (87.5% male), with a mean age of 56 ± 14.5 years. Hernias were classified according to the Nyhus classification [17] (Table 1). Twenty-nine fascias from the nonherniated sides (9 with a direct hernia on the other side, 14 with an indirect hernia, 2 with a type IIIb hernia, and 4 with a femoral hernia on the other side) were available for the study. The mean age of the 20 control subjects (60% male) was 60.5 ± 18.4 years. The only significant difference between the control subjects and patient characteristics was the gender ($P = 0.015$).

TABLE 1 *Distribution of Patients and Herniated Fascias According to the Nyhus Classification*

Type	Description	Sides	No. of patients/ fascias
II	Indirect without posterior wall defect	Unilateral	14/14
		Bilateral	3/6
III	*Posterior wall defect*		
IIIa	Direct	Unilateral	9/9
		Bilateral	4/8
IIIb	Indirect	Unilateral	2/2
IIIc	Femoral	Unilateral	4/4
	Mixed bilateral hernias	Bilateral	2/4
IIIa-IIIb			2/3
Total			40/50

Control Samples

No significant biochemical difference was observed between the left and right sides of the rectus sheaths and fascias (data not shown). Accordingly, the data of left and right sides were averaged for each control subject. The sheaths were compared with the fascias in a paired manner (Table 2). The hydration percentage was similar in these two structures. The collagen concentration per milligram dry weight was very significantly higher in the rectus sheaths than in the fascias ($P < 0.001$). On the other hand, the percentages of extractible collagen with, respectively, NaCl, HAc, and pepsin were significantly increased in the fascias ($P < 0.05$). Regarding the qualitative collagen composition, there were a significant decrease of the percentage of the β chains ($P < 0.05$) and a tendency toward a decrease of the type I/III collagen ratio in the fascias ($P = 0.08$), when compared with the rectus sheaths.

The comparison of the three types of control subjects (autopsies, organ donors, and surgical patients) revealed an increased collagen content per milligram dry weight in the group of organ donors, when compared with the group of autopsies (respectively, 739 ± 95 µg versus 626 ± 123 µg for autopsies). This observation prompted us to study the age and sex influence on the different biochemical variables for the control sheaths and fascias. In the rectus sheaths, the difference of the collagen content was related to the difference of the mean age between the two groups (respectively, for organ donors 42 ± 15 and 71.5 ± 12 years for autopsies). The collagen

concentration decreased with age (r = -0.52, P = 0.02). In the control fascias, an effect of age (P = 0.007) and gender (P = 0.003) was found on the collagen content per milligram dry weight. The collagen content of the fascias was higher in females (558 ± 121 μg) than in males (426 ± 128 μg). As in the control sheaths, it decreased with age (in males, collagen content = 650 - 4.17 × age; in females, collagen content = 831 - 4.17 × age). Accordingly, it was necessary to take age and gender into account to make a valid comparison between the control subjects and the patients.

TABLE 2 *Comparison of the Control Fascias and Rectus Sheaths*

	Sheaths	Fascias
	(mean± SD)	
Wet weight[a] (n = 17)	88.7 ± 13.3	42.9 ± 8.9[e]
Dry weight[a] (n = 17)	22.6 ± 4.2	11.2 ± 3.4[e]
Hydration % (n = 17)	74.4 ± 4.3	74.3 ± 4.1
NaCl collagen extractibility % (n = 16)	0.09 ± 0.04	0.11 ± 0.07[c]
HAc collagen extractibility % (n = 16)	0.13 ± 0.08	0.23 ± 0.23[d]
Pepsin collagen extractibility % (n = 16)	1.73 ± 0.81	2.58 ± 1.44[d]
Collagen content[b] (n = 16)	670 ± 127	480 ± 139[e]
α_1/α_2 (I) ratio (n = 7)	2.0 ± 0.3	2.2 ± 0.5
Type I collagen % (n = 1)	75.1 ± 6.9	70.6 ± 6.3
Type III collagen % (n = 1)	24.9 ± 6.8	29.4 ± 6.3
β chains % (n = 1)	38.6 ± 4.4	34.1 ± 4.6[c]

[a] Expressed as mg/100 mm^2.
[b] Expressed as μg/mg dry weight.
[c] P< 0.05.
[d] P < 0.01.
[e] P < 0.001.

Samples from Treated Patients with Hernia

Rectus sheaths. We compared the patient sheaths (n = 40) and the control sheaths (n = 37), taking into consideration different factors, such as age, gender, and body mass index (BMI). The qualitative study of the collagen (ratio $\alpha_1(I)/\alpha_2(I)$, β chains, type I and III collagens) concerned 24 patient and 19 control sheaths. The only statistically significant difference between the two groups was an increased wet weight per unit area in the patients (Table 3). The statistical analysis also showed that the dry weight per unit area was significantly (P < 0.05) lower in the females than in the males (data not shown). The collagen content per milligram dry weight significantly decreased with age (r = -0.3, P < 0.01). The same negative correlation was observed for the percentage of the collagen solubi-lized by pepsin (r = -0.3, P < 0.01).

Fascias. As for the sheaths, the control and patient fascias were compared with a general linear model, taking into account the type of fascia (control, non-herniated, indirect hernia, direct hernia, type IIIb hernia, and femoral hernia), the age, and the gender (Table 4). Globally in the patients, we observed an increased wet and dry weight per 100 mm^2. The fascias from the direct hernias presented a significant increase of the collagen extractibility with pepsin, when compared with the control fascias (5.2 versus 2.6%). The extractibility percentage of the fascias from the indirect hernias and the nonherniated sides was intermediate (respectively, 3.3 and 3.4%). The collagen content per milligram dry weight was significantly increased in the indirect hernias, when compared with the control fascias. In the older subjects, we observed a decrease of the hydration, the collagen content per milligram dry weight, and the collagen extractibility with HAc and pepsin. In females, the collagen content per milligram dry weight was significantly higher than in males (521 ± 148 μg versus 499 ± 162 μg, P < 0.01). The qualitative collagen analysis did not show any significant difference between the patients and the controls. The time elapsed between the operation and the occurrence of the direct or indirect hernias influenced some biochemical parameters. In the direct hernia group, we observed a significant negative correlation for the percentage of extracted collagen with HAc (r = -0.6, P = 0.02) and of β chains (r = -0.8, P = 0.006). In the indirect hernia group, a negative correlation was observed between the dry weight/100 mm^2 (r = —0.6, P = 0.009) and hernia duration.

***TABLE 3** Comparison of the Control and Patient Rectus Sheaths*

	Patients	Controls
	(mean ± SD)	(mean ± SD)
Wet weight[a]	108 ± 26	88.4 ± 15[c]
Dry weight[a]	26.7 ± 8.5	22.5 ± 4.8
Hydration *%*	75.4 ± 3.6	74.5 ± 4.4
NaCl collagen extractibility *%*	0.08 ± 0.05	0.08 ± 0.04
HAc collagen extractibility *%*	0.10 ± 0.06	0.13 ± 0.08
Pepsin collagen extractibility *%*	2.2 ± 1.9	1.8 ± 0.9
Collagen content[b]	650 ± 114	679 ± 137
α_1/α_2 (I) ratio	2 ± 0.25	2 ± 0.32
Type I collagen *%*	78.3 ± 7.3	73.4 ± 7.3
Type III collagen *%*	21.7 ± 7.3	26.6 ± 7.3
β chains *%*	37.1 ± 5.5	36.6 ± 6.6

[a] Expressed as mg/100 mm^2.
[b] Expressed as µg/mg dry weight.
[c] $P < 0.05$.

***TABLE 4** Comparison of the Control and Patient Fascias*

	Controls	NH	IH	DH	IIIb	FH
	(All values expressed as mean ± SD)					
Wet weight[a]	42.9 ± 12.6[c] (n = 34)	66.1 ± 22.3 (n = 29)	58.2 ± 25.7 (n = 20)	63.2 ± 25.8 (n = 19)	53 ± 3.1 (n = 3)	79.9 ± 14.4 (n = 8)
Dry weight[a]	11.2 ± 4.4[c] (n = 34)	16.9 ± 7.9 (n = 29)	13.8 ± 6.1 (n = 19)	14.7 ± 10.1 (n = 19)	11.3 ± 2.5 (n = 3)	20.3 ± 7.9 (n = 8)
Hydration %	74.3 ± 5.3 (n = 34)	74.3 ± 7.4 (n = 29)	76.3 ± 3.8 (n = 19)	77.5 ± 6.3 (n = 19)	78.4 ± 5.9 (n = 3)	74.8 ± 8 (n = 8)
NaCl collagen extractibility %	0.11 ± 0.08 (n = 33)	0.15 ± 0.11 (n = 29)	0.15 ± 0.11 (n = 18)	0.12 ± 0.08 (n = 17)	0.13 ± 0.07 (n = 3)	0.11 ± 0.08 (n = 8)
HAc collagen extractibility %	0.22 ± 0.22 (n = 33)	0.19 ± 0.15 (n = 29)	0.22 ± 0.17 (n=18)	0.23 ± 0.12 (n = 17)	0.23 ± 0.1 (n = 3)	0.16 ± 0.1 (n = 8)
Pepsin collagen extractibility %	2.6 ± 1.5 (n = 33)	3.4 ± 2.2 (n = 29)	3.3 ± 1.7 (n = 18)	5.2 ± 2.7[d] (n = 17)	3.9 ± 2 (n = 3)	2.5 ± 1.4 (n = 8)
Collagen content[b]	484 ± 151 (n = 33)	500 ± 179 (n = 29)	573 ± 114[d] (n = 18)	509 ± 188 (n = 19)	475 ± 142 (n = 3)	445 ± 113 (n = 8)
α_1/α_2 (I) ratio	2.2 ± 0.5 (n = 15)	2.4 ± 0.3 (n = 14)	2.2 ± 0.3 (n = 12)	2.4 ± 0.3 (n = 13)	1.8 ± 0.05 (n = 2)	2.5 ± 0.4 (n = 5)
Type I collagen %	70.6 ± 7.6 (n = 15)	73.6 ± 9.2 (n=14)	71.7 ± 7 (n = 12)	71.2 ± 10.5 (n = 13)	69.2 ± 3.7 (n = 2)	76.2 ± 4.4 (n = 5)
Type III collagen %	29.4 ± 7.6 (n = 15)	26.4 ± 9.2 (n=14)	28.3 ± 7 (n = 12)	28.8 ± 10.5 (n = 13)	30.8 ± 3.7 (n = 2)	23.8 ± 4.4 (n = 5)
β chains %	33.3 ± 5.7 (n = 15)	34.8 ± 5.2 (n = 14)	33.9 ± 7.1 (n = 12)	34.6 ± 5 (n = 13)	32.8 ± 8.6 (n = 2)	32.7 ± 8.2 (n = 5)

Note. NH, nonherniated sides; IH, indirect hernias; DH, direct hernias; IIIb, inguino-scrotal hernias (see Table 1, Nyhus classification); FH, femoral hernias.
[a] Expressed as mg/100 mm^2.
[b] Expressed as µg/mg dry weight.
[c] $P < 0.001$.
[d] $P < 0.01$ vs the other groups after adjusting for age and sex.

DISCUSSION

Up to now, there are very few biochemical studies focusing on the transversalis fascia [9, 18]. However, its study is fundamental to explore the mechanisms leading to inguinal herniation, because its disruption in the weak inguinal area is responsible for groin hernias. Abnormalities of the skin or rectus sheath in these patients can be considered potential markers of systemic connective tissue pathology. It is the reason why we also analyzed the rectus sheath in this study. We want also to point out the localization of the fascia samples. In order to study regional factors that could modify the expression of a systemic disease, the best control transversalis fascia must

be sampled from the posterior wall of the inguinal canal. For ethical reason taking such a sample in healthy subjects is not acceptable because of the risk of inducing subsequent herniation; therefore we used two types of controls (autopsies and organ donors). The nature of these control samples seemed appropriate. Indeed, the feasibility of this type of biochemical study from autopsy samples has already been demonstrated on the aortic wall [19]. The organ donors being generally younger than the autopsied subjects, it was worthy to analyze the influence of age and sex on the investigated biochemical parameters. Another original aspect of this work is to have evaluated the modifications of the rectus sheath and the transversalis fascia in relation with aging and gender.

We found that the collagen content per milligram dry weight of the sheaths was lower in the females than in the males, while the percentage of collagen extracted with pepsin decreased with age. This observation has been already described in skin [20]. A further observation was the significant difference between the control sheaths and the fascias. The collagen content was higher in the sheaths, but the collagen extractibility with NaCl, HAc, and pepsin was significantly higher in the fascias, suggesting a lower level of collagen cross-linkings in this thin anatomical structure. The qualitative collagen composition also seemed to be different, with a tendency toward an increase in type III collagen in the fascias.

When comparing the patient and control sheaths, the only statistically significant difference was an increased wet weight per unit area in the patients, the collagen content per milligram dry weight being similar in the two groups. These observations are not in agreement with the results of Wagh *et al.* [4], who found a decreased wet weight of the rectus sheaths in the hernia patients, with, however, no difference in the dry weight. In their series, the collagen concentration per milligram dry weight was significantly decreased in the direct hernia group, suggesting an increase of the noncollagen proteins. Surprisingly, the amount of glycoproteins and mucopolysaccharides was not significantly altered compared to controls.

When compared to control fascias, the fascias from the hernia patients globally presented an increased wet and dry weight per 100 mm^2. We have no obvious explanation for this observation. This increase of wet and dry weight in the patient fascias might be perhaps related to the conditions of sampling: although great care was taken to standardize the procedures, the dissection of the posterior wall of the inguinal canal could have been more extensive in the organ donors and the autopsied subjects, with a stronger traction on the abdominal wall to place the area gauge onto a plane surface. Consequently, if the posterior wall was more stretched, the thickness of the sample could have been reduced.

Nevertheless, we can conclude that there is no obvious atrophy of the transversalis fascia in the hernia pathology. We even observed an increase of the collagen content per milligram dry weight in the indirect hernias. This increase is surprising and its explanation remains unclear. Since we did not observe a similar increase in the other groups of fascias, this observation could be more specific for the indirect hernias. But this phenomenon was not observed in the very large indirect hernias (type IIIb). Furthermore, it is well-known that collagen metabolism is regulated by mechanical forces [21]. The distribution of the mechanical forces, induced by the intra-abdominal pressure, onto the posterior wall of the inguinal canal, could be different in the direct and indirect hernias, related to the presence of the indirect sac beyond the posterior wall. Therefore, the increase in collagen concentration in the indirect hernias could reflect a remodeling process of the connective tissue, present mainly at the beginning of the hernia, as a negative correlation was observed between the dry weight/100 mm^2 and the hernia duration in the indirect hernia group. Sequential extraction of collagen in solvents of increasing dissociation capacity (NaCl < acetic acid < pepsin) is a procedure permitting some estimation of the state of maturation of the molecules in the fibers. Extractibility is high in the skin of actively growing animals [22] ; it diminishes with aging in the human skin [20, 23]. It is largely increased in animals treated with an inhibitor of lysyl oxidase, the enzyme involved in the formation of the covalent aldehyde cross-links in the mature fibrils of collagen [24]. The main characteristic of the fascias from the direct hernia was a significant increase in collagen extractibility with pepsin, when compared to the control fascias (5.2 versus 2.6%). The extractibility of the fascias from the indirect hernias and the nonherniated sides was intermediate (respectively, 3.3 and 3.4%). Three hypotheses can explain this increased extractibility. The first one would be an enhanced collagen turnover rate producing an increased proportion of immature collagen. The second one might be an increased partial breakdown of the collagen by enzymes, such as the matrix metalloproteinases (MMP). Based on immuno-morphological data, an increased expression of MMP-2 was observed in the fascias from direct hernias, when compared to the indirect hernias [9]. No data on its state of activation are available so far. The third one would be a defective maturation and formation of the covalent cross-links. This is, however, less likely since the proportion of β chains (dimers of α chains co-valently cross-linked) did not vary in the different types of hernia.

In our study, no important biochemical alteration was observed in the fascias from the femoral hernias, but the

number of samples was small. The qualitative collagen investigations did not reveal any significant difference between the control and patient fascias, in particular in the ratio of α_1 and α_2 chains of type I collagen and in the proportion of collagen type I and type III. The observed ratios are those usually found in normal connective tissues. Our results are in contradiction with those of Klinge *et al.* [18], who found a reduction of the collagen I/III ratio, due to an increased amount of collagen type III in the hernia fascias, and surprisingly a complete absence of α_2 (I) chains, while these chains clearly exist in our study and are present in a normal ratio. The patterns obtained by Western blot used in their study were heavily loaded, influencing therefore their quantitative interpretation.

With regard to the age and sex influence on the biochemical parameters, we observed, in the fascias, a decrease in collagen content per milligram dry weight with aging as well as a decreased extractibility of collagen in HAc and with pepsin. This is in accordance with the data of the literature reported for skin [20, 23] and the well-known increase of collagen cross-linkage with age, resulting in diminished extractibility. In females, we found an unexpected higher collagen content per milligram dry weight compared to the male population. The reason for this is unknown but this finding could explain, at least partially, why the groin hernias are less frequent in females than in males.

In conclusion, the increased collagen extractibility in the fascias from the nonherniated sides suggests that a connective tissue pathology may be implicated in the genesis of groin hernias. This hypothesis is further supported by our previous studies clearly showing bio-mechanical and structural histological alterations of the nonherniated side [10, 11]. The transversalis fascia from the nonherniated side could correspond to a preclinical stage of hernia disease. This pathology seems to mainly affect the fibrillar network of collagen polymers although, to our knowledge, the elastic fibers have not been biochemically investigated. In our bio-mechanical study [10], extensibility (maximum distension) and biological elasticity (capacity of the stretched tissue to return to its initial position) were significantly increased in the fascias from the direct hernias and the nonherniated sides. The correlation between the collagen extractibility with pepsin and the values of extensibility and elasticity recorded for all the fascias ($n = 108$) yielded a very significant correlation ($r = 0.36$, $P < 0.001$) with maximum distension but not with elasticity. This is also in favor of a defective organization of the collagen fibers. According to our studies, this connective tissue pathology would express preferentially its effects in the inguinal region, since we did not observe any major biomechanical, histological, and biochemical difference between the rectus sheaths of controls and those of patients. This regional expression of a more generalized alteration of collagen metabolism could be linked to genetic factors, currently unknown, and to the important mechanical stresses occurring in the inguinal region. These genetic factors could play a role in the increased type III collagen synthesis found in skin fibroblast cultures from patients with hernias [8]. Further studies are needed to understand this complex metabolic problem, and clinical investigations are required to find clinical signs of this suspected systemic collagen pathology, which would allow to propose a systematic bilateral repair in patients at risk.

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