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The anatomy of the anterolateral structures of the knee — A histologic and macroscopic approach☆

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ABSTRACT

Background: The anatomy of the anterolateral structures of the knee is complex and still controversial. The aim of our study was to analyze this anatomy by histologic and macroscopic evaluation, with a particular emphasis on the anterolateral ligament (ALL).
Material and methods: Twenty-three cadaveric knee joints were dissected followed by a qualitative and quantitative anatomic analysis of the anterolateral knee structures. Histology and comparison of different anterolateral structures was performed in addition.
Results: The ALL was identified in all of the dissected cadaveric knee specimens. It runs in an oblique course from its proximo-dorsal insertion at the distal femur into a ventro-distal direction to the anterolateral tibia. The femoral insertion site was found to be posterior and slightly proximal to the lateral femoral epicondyle and the femoral attachment of the lateral collateral ligament (LCL). The femoral insertion of the ALL overlapped the LCL in all dissected knees. The tibial insertion site was midway between Gerdy's tubercle (GT) and the tip of the fibular head (FH). In 15 of the dissected 23 knee joints, thin attachments to the lateral meniscus were observed. Histology confirmed differences in the composition of the anterolateral knee joint capsule, the ALL and the iliotibial band (ITB).
Conclusions: The ALL occurs as a regular separate anterolateral ligamentous structure. It is distinguishable from the ITB and the anterolateral joint capsule in both embalmed and non-embalmed specimens. Histology of the ALL indicates typical ligamentous tissue which clearly differs from the anterolateral knee joint capsule and the thicker ITB.
Level of evidence: Level II, descriptive anatomic study.

1. Introduction

Persistent dynamic rotational laxity is a well-known problem following anatomic anterior cruciate ligament (ACL) reconstructions [1]. In those cases, an additional lesion of the anterolateral structures of the knee may be suspected [2–6]. The anatomy of

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the anterolateral soft tissue structures of the knee are complex and the subject of controversy [4,7–12]. The anterolateral joint capsule, the lateral meniscus, the anterolateral ligament (ALL) as a separate ligamentous structure and the iliotibial band (ITB) are proposed as relevant anterolateral knee joint stabilizers. In 1879, Segond described a "pearly, fibrous band" as an anterolateral ligamentous structure. Vincent et al. [13] and Claes et al. [14] rediscovered this structure and named it the ALL. Further studies [15–21] confirmed these findings by identifying a separate anterolateral knee ligament with distinct femoral and tibial insertion sites. Nevertheless, others rejected this theory and denied the presence of a separate ALL, but rather described an anterolateral complex consisting of the iliotibial tract and its deep capsulo-osseous layer, the so-called "Kaplan fibers", and the anterolateral joint capsule [22–26]. The results of our pilot study revealed that the ALL could be found as a distinct, separate anterolateral ligamentous structure of the knee [27]. The aim of our present study was to describe a reproducible dissection technique of the anterolateral knee joint structures for embalmed and non-embalmed specimens, to analyze the anatomy of the anterolateral structures of the knee by histologic and macroscopic evaluation and to verify the existence of the ALL. Our hypothesis was that the ALL is a separate anterolateral ligamentous structure which is distinguishable from the surrounding tissues by a histologic as well as macroscopic approach.

2. Material and methods

2.1. Specimens

The anterolateral knee joint structures were dissected in 20 paired human cadaveric knee joints of formalin fixed (embalmed) body donors (6 male, 4 female, aged between 54 and 81 years). Three knees (2 left, 1 right) were taken from two female alcohol-fixed body donors (aged 85 and 90 years) early post-mortem (non-embalmed). The mean age of the body donors at the time of death was 73 [54–90] years. Previous surgical procedures of the knee joint were excluded. A total of 23 cadaveric knees were examined. The specimens were taken from the body donation program of Saarland University Medical Center, Institute of Anatomy and Cell Biology.

2.2. Dissection technique

The anterolateral knee joint structures were all dissected in a standardized dissection technique which was based on the recommendations by Claes et al. [14] and Daggett et al. [20]. It was used for embalmed as well as non-embalmed specimens. The relevant anatomic landmarks (patella, tibial tuberosity, Gerdy's tubercle [GT], fibular head [FH], lateral joint line, lateral femoral epicondyle, ITB) were first marked on the skin. Afterwards a curved skin incision was done starting distally, connecting GT, the lateral femoral epicondyle, and the ITB. A cutaneous flap was created anteriorly as well as posteriorly and the subcutaneous fat and soft tissue was removed to visualize the ITB, extensor apparatus, patella, patella tendon, tibial tuberosity, GT, FH and the short head of biceps femoris. The ITB was cut transversely 6–8 cm proximal to the lateral femoral epicondyle and was then carefully reflected distally. The biceps femoris was reflected posteriorly off the fibular head. After a partial capsulectomy the ALL was identified in knee flexion and internal rotation of the tibia. The detailed stepwise dissection technique is shown in Figure 1.

2.3. Macroscopic analysis

Macroscopy included a qualitative and quantitative anatomic description of the ALL.

2.3.1. Qualitative anatomic description

The qualitative anatomic description of the ALL was done with respect to its femoral and tibial insertion sites, shape, course and relation to the lateral collateral ligament (LCL) and the lateral femoral epicondyle.

2.3.2. Quantitative anatomic description

The quantitative anatomic description of the ALL included its length, width (at femoral insertion site, joint line and tibial insertion site), thickness (at joint line) as well as distance to GT and distance to the FH. All measurements were performed with a digital caliper (accuracy of 0.01 mm).

2.4. Histologic examination

For histology, three different tissue samples were taken from the specimens: part of the anterolateral knee joint capsule, the ALL and the ITB. All tissue samples were postfixed in 4% formalin for two weeks, dehydrated in graded concentrations of ethanol and embedded in paraffin. Sagittal sections 7-μm thick were stained by HE and MG (HE = hematoxylin–eosin stain, MG = May–Grünwald stain) according to Romeis [28]. The qualitative description compared the three different samples with respect to thickness, content of dense connective tissue, fiber orientation, presence of cellular material and synovial membrane as well as number and size of blood vessels. A standardized magnification of 50-fold and 200-fold was applied for microscopic evaluation of the samples. The microscopic evaluation and comparison of the samples was performed with a Zeiss Axiophot microscope by an experienced senior anatomist and histologist.

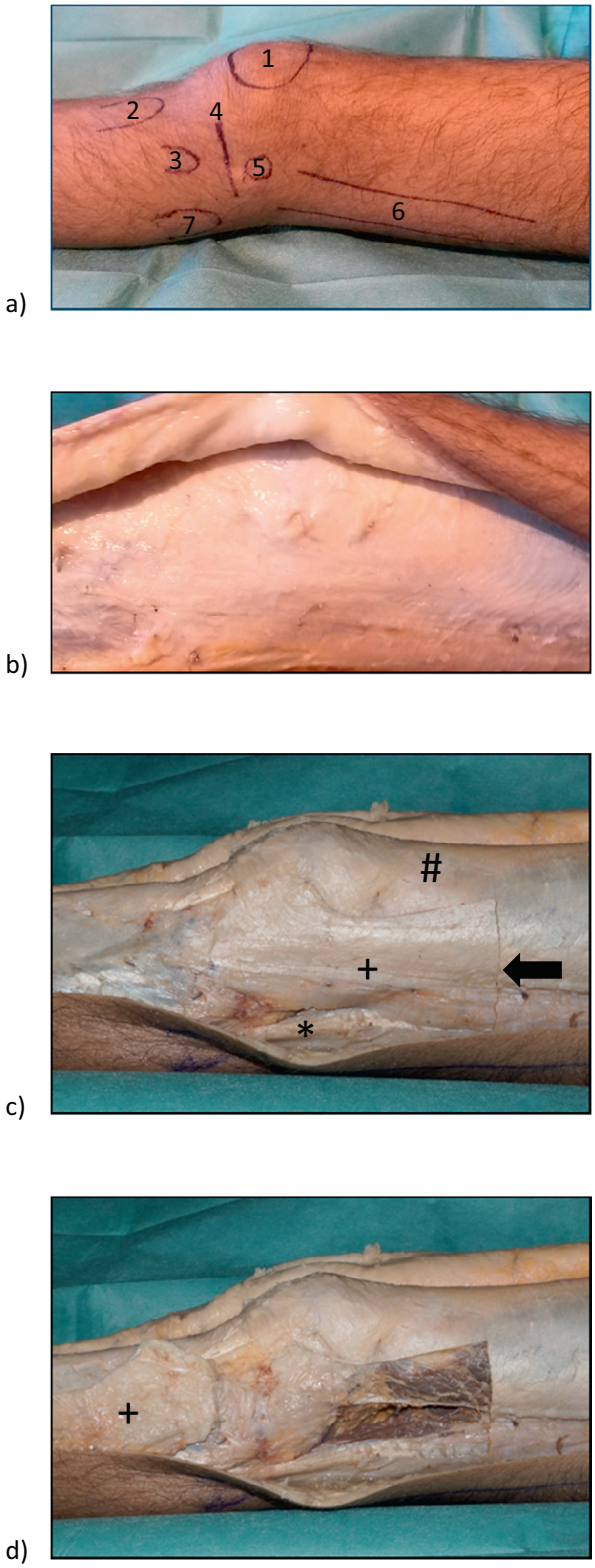
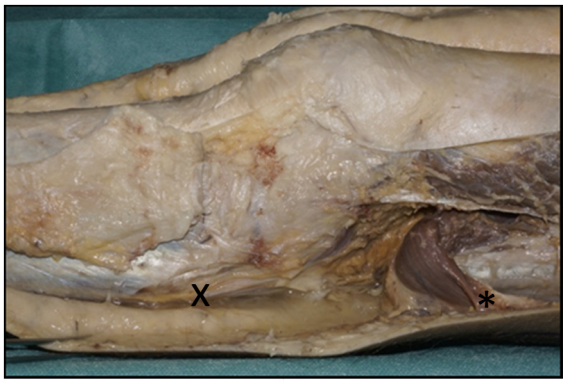


Figure 1. Dissection technique: stepwise dissection of the anterolateral structures of a left knee (right = proximal, left = distal), a) marking of the anatomic landmarks (1 patella, 2 tibial tuberosity, 3 GT, 4 anterolateral joint line, 5 lateral femoral epicondyle, 6 ITB, 7 FH) b) skin incision, elevating a cutaneous flap, c) dissection of ITB (+), extensor apparatus (#) and short head of biceps femoris (*); ITB was cut transversely 6–8 cm proximal to the lateral femoral epicondyle (arrow), d) ITB (+) is carefully reflected distally, e) biceps femoris (*) is reflected posteriorly off the FH (x), f) identification of the ALL (arrow, red needles mark the femoral and tibial insertion of the ALL) with knee in flexion and internal rotation of the tibia.



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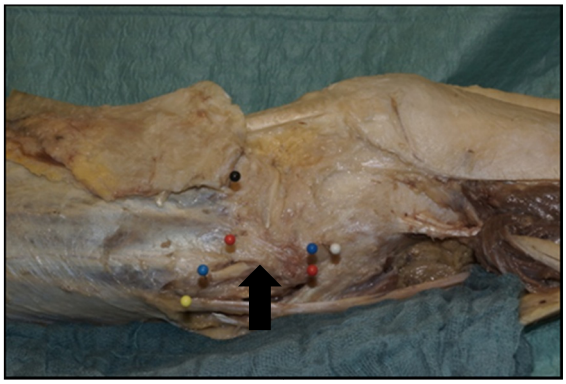

f)

Figure 1 (*continued*).

2.5. Statistical analysis

Statistical analysis was performed using Microsoft Excel 2010® (Redmond, WA, USA) and IBM SPSS Statistics® Version 22 (Armonk, NY, USA). Descriptive statistics were performed for the quantitative analysis including mean value, standard deviation and min/max values.

3. Results

3.1. Dissection technique

The dissection technique was reproducible in all specimens and resulted in an adequate exposure of the anterolateral knee joint structures especially the ALL. It was applicable for both embalmed and non-embalmed specimens. It was easier to separate the different tissue layers of the anterolateral knee structures and to position the knee joints in flexion and internal rotation in non-embalmed fresh cadaveric specimens than in embalmed specimens. Furthermore, in non-embalmed fresh cadaveric specimens, it was less difficult to distinguish the ALL from the surrounding anatomical structures such as the deep layer of the ITB and the anterolateral joint capsule as well as to identify the femoral insertion site of the ALL to embalmed specimens.

3.2. Macroscopic analysis

3.2.1. Qualitative anatomic description

A distinct, separate extra-articular anterolateral ligamentous structure as previously described and named the ALL was identified in all 23 dissected knees. The ALL was separated in both embalmed as well as non-embalmed specimens. It was distinguishable from the anterolateral joint capsule and the deep layer of the ITB. It had an oblique course from the distal femur proximodorsal into a ventro-distal direction to the anterolateral tibia. The femoral insertion site was found to be posterior and slightly proximal to the lateral femoral epicondyle and the femoral attachment of the LCL. The femoral insertion of the ALL overlapped the LCL in all dissected knees. The tibial insertion site was midway (49.2%) between GT and the tip of the FH. In 15 of the dissected 23 knee joints (65%), thinner attachments to the lateral meniscus were found. Internal rotation of the tibia led to tensioning of the ALL. Figures 2 and 3 show the macroscopic anatomy of the anterolateral knee structures of an embalmed and non-embalmed specimen respectively.

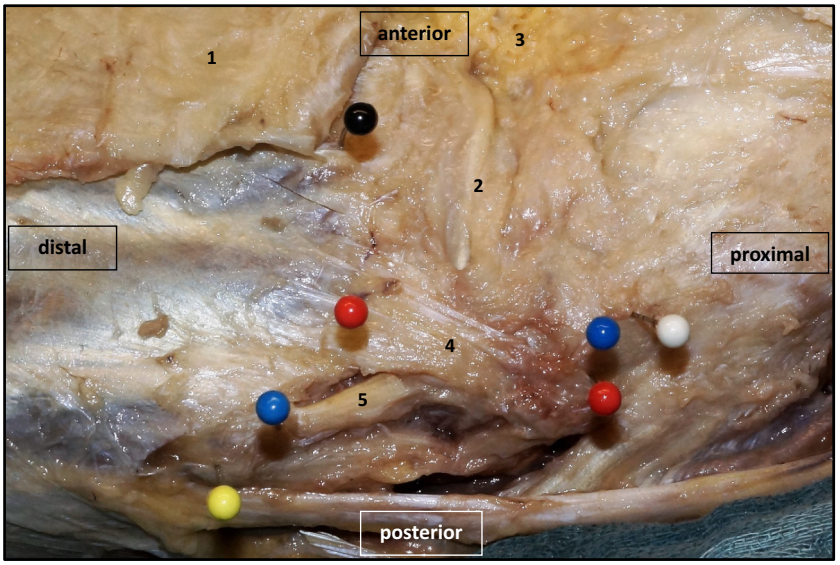

Figure 2. Lateral view of the anterolateral structures of a left knee (embalmed specimen, male). Needle black: GT, needle yellow: FH, needle white: Lateral femoral epicondyle, needles red: Femoral and tibial insertion of the ALL, needles blue: Femoral and tibial insertion of the LCL, 1 ITB (cut and reflected distally), 2 Lateral joint line, 3 Hoffa fat body, 4 ALL, 5 LCL.

3.2.2. Quantitative anatomic description

The results of the quantitative anatomic analysis are presented in Table 1.

Figure 4 shows a schematic drawing of the anterolateral knee structures based on the results of the macroscopic analysis.

3.3. Histologic examination

Histology of the anterolateral joint capsule revealed a thin connective tissue structure (fibrous membrane) with a few and randomly dense packed collagen fibrils. Below this, loose connective tissue (subsynovial membrane) with many small blood vessels was visible followed by a synovial membrane consisting of type A and type B synoviocytes (Figure 5). Sections of the ALL presented a thin ligamentous structure with organized densely packed collagen fibrils. The ALL was thicker and consisted of more connective tissue than the anterolateral joint capsule. In ALL, fibers had a parallel orientation and were arranged slightly wavelike.

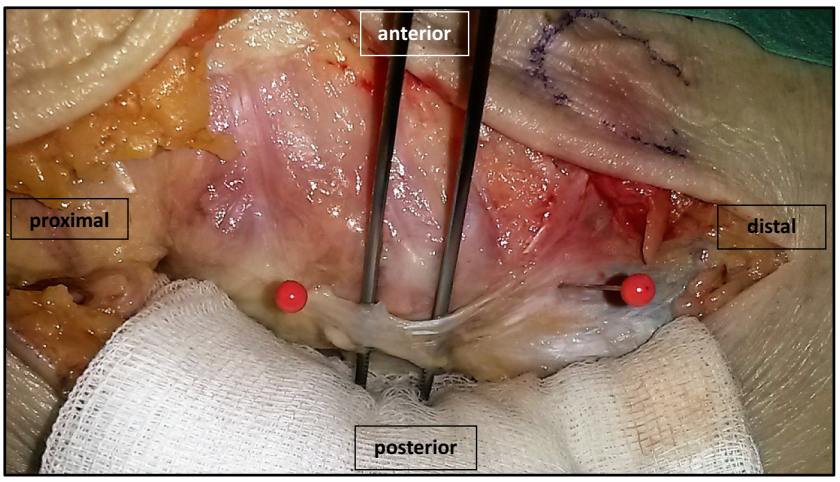

Figure 3. Lateral view of the anterolateral structures of a left knee (non-embalmed specimen, female), forceps placed between anterolateral joint capsule and ALL (femoral and tibial insertion of the ALL marked by red needles).

Table 1
Macroscopic evaluation – quantitative analysis (all values in mm, SD = standard deviation, Max = highest value, MIN = smallest value, ALL = anterolateral ligament, GT = Gerdy's tubercle, FH = fibular head).

	Mean value	SD	Min	Max
ALL length (in extension of the knee)	39.13	7.26	28.64	51.18
ALL width (femoral insertion)	10.20	0.89	8.48	12.66
ALL width (joint line)	8.99	1.47	4.53	12.33
ALL width (tibial insertion)	9.91	1.43	6.49	13.17
ALL thickness (joint line)	2.06	0.34	1.34	2.67
Distance ALL – GT	17.61	2.43	14.14	23.86
Distance ALL – FH	18.16	3.36	11.94	23.71

Only a few cells were visible and sporadically occurring blood vessels (Figure 6). The ITB was always a thick ligamentous tissue structure of bigger size than the ALL. In the ITB, collagen fibrils run in thick bundles of parallel organized collagen fibrils. Comparable to the ALL only a few cells could be observed between the connective tissue fibers. The ITB showed denser connective tissue

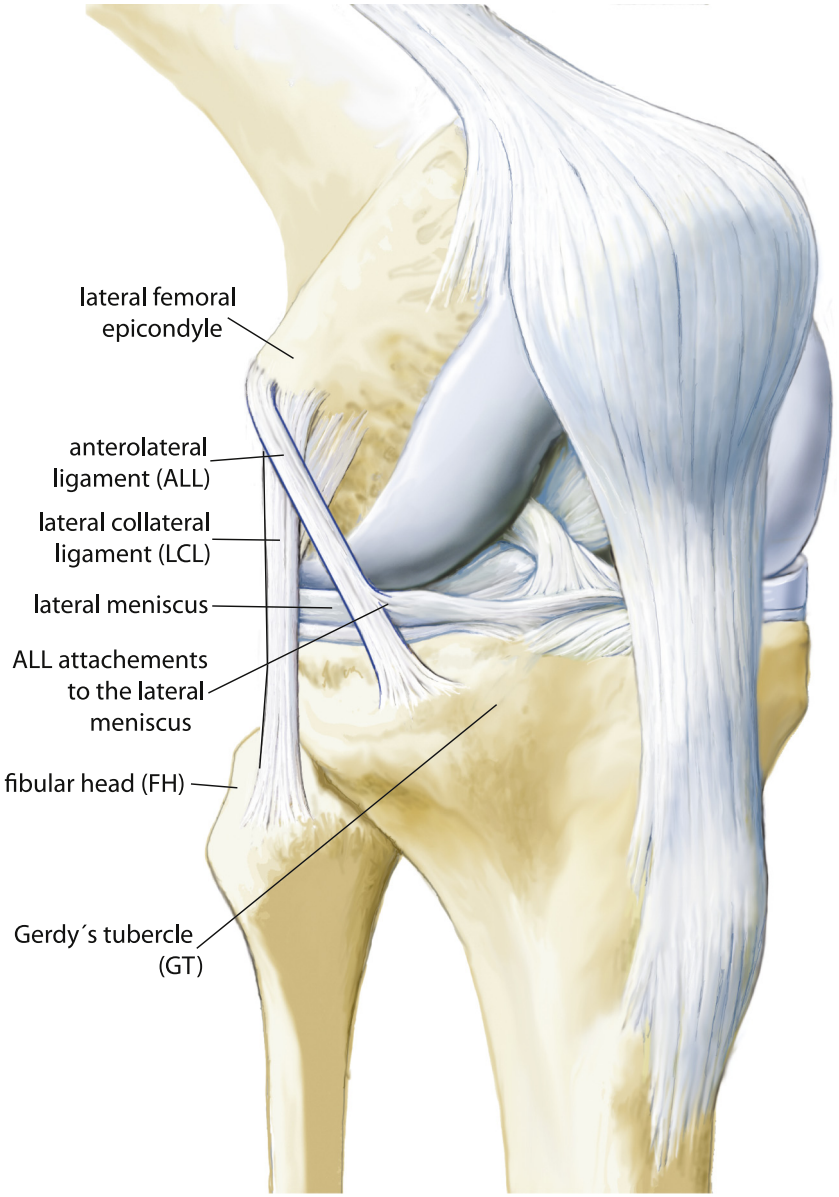

Figure 4. Schematic drawing of the anterolateral knee structures.

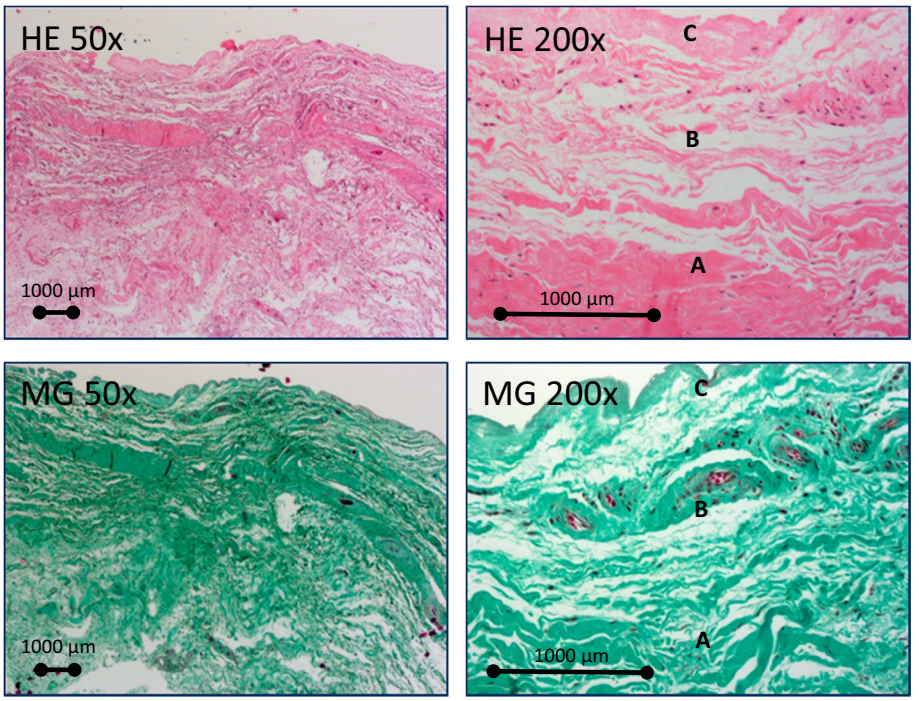

Figure 5. Histology: Anterolateral knee joint capsule (HE = hematoxylin–eosin stain, MG = May–Grünwald stain). Note the fibrous membrane (thin connective tissue structure with a few and randomly dense packed collagen fibrils; A), below the subsynovial membrane (loose connective tissue with many small blood vessels; B) and followed by the synovial membrane (C).

than the anterolateral joint capsule and the ALL. In addition, fatty tissue was visible between the connective tissue strands (Figure 7). The histologic analysis of the tissue samples confirmed the expected differences in histological composition of the anterolateral knee joint capsule, the ALL, and the ITB.

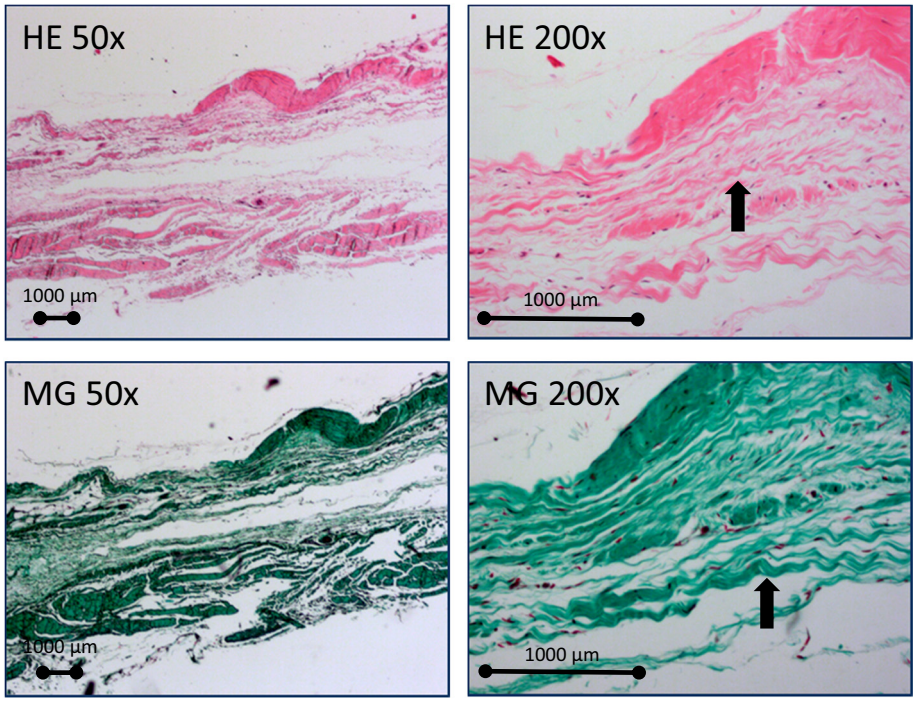

Figure 6. Histology: Anterolateral ligament (ALL) (HE = hematoxylin–eosin stain, MG = May–Grünwald stain). Note the organized densely packed collagen fibrils of this thin ligamentous structure and the parallel orientation of the slightly wavelike arranged fibers (arrow).

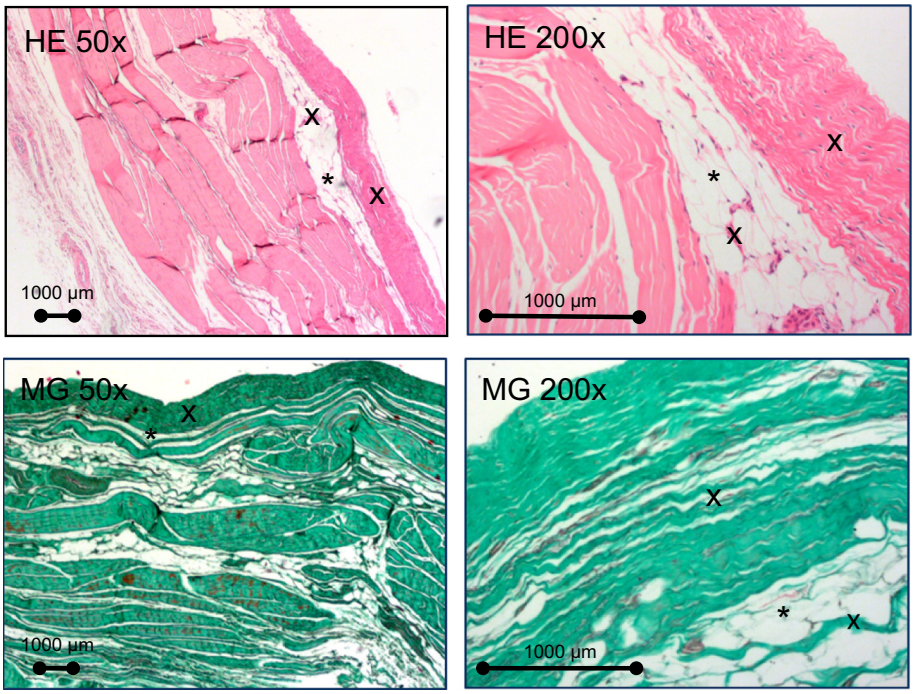

Figure 7. Histology: Iliotibial tract (ITB) (HE = hematoxylin–eosin stain, MG = May–Grünwald stain). Note that the collagen fibrils are parallel organized and run in thick bundles. Only a few cells can be observed between the connective tissue fibers. It shows denser connective tissue than the anterolateral joint capsule and the ALL. In addition fatty tissue (*) was visible between the connective tissue strands (x).

4. Discussion

Our study confirmed the presence of an ALL in 23 knees from 12 cadavers. The ALL was found as a separate ALL in all specimens. It was distinguishable from the ITB and the anterolateral joint capsule in both embalmed and non-embalmed specimens. The histological findings of the ALL support the view of a separate ligament which was clearly histologically distinguishable from the anterolateral knee joint capsule and the ITB.

The number of 23 human cadaveric knee specimens seems acceptable with respect to other recent anatomic studies of the anterolateral aspect of the knee joint. Lower or comparable numbers of knee specimens could be found in most anatomic studies [13,14,16,17,29–32]. Nevertheless, some studies included higher numbers of specimens [21,33]. Daggett et al. [21] analyzed the femoral origin of the ALL in a considerably higher number of 52 specimens. Neri et al. [34] dissected 84 fresh-frozen cadaveric knees.

Following the discussion in the literature about the best conditions for dissections of the ALL, the present study analyzed the anterolateral structures of the knee joint in embalmed as well as non-embalmed specimens. The ALL was found under both conditions, but the dissections revealed some advantages of the latter. Indeed, it was easier to separate the different soft tissue layers of the anterolateral knee structures, to distinguish the different anatomic structures, and to position the knee joints in flexion and internal rotation. Most of the reported studies presented the results of an anatomic dissection of the anterolateral aspect of fresh or fresh-frozen knee specimens [31,32,34,35]. Vincent et al. [13] analyzed the anterolateral knee structures in 10 fresh cadaveric knees. Other studies dissected the anterolateral aspect of the knee in embalmed specimens [14,21,33].

The technique which was used to dissect the anterolateral knee was based on the method previously described by Claes et al. [14] and Daggett et al. [20]. It was successful and feasible in all dissected knees, regardless whether embalmed or not embalmed. The key point of this technique is a careful and precise dissection at the thinner part of the ITB while reflecting the ITB distally in order to avoid an accidental resection of the ALL fibers [20]. Parker et al. [36] described another dissection technique for stiff, embalmed specimens in which the quadriceps tendon is incised and the lateral inferior genicular vessels are used to identify the ALL. With our technique the quadriceps tendon could be preserved.

In the present study, the ALL was identified in all dissected specimens. This is in line with the majority of other recent studies. Helito et al. [17] confirmed the presence of the ALL in all of the 20 human cadavers they dissected. Claes et al. [14] found the ALL in 97% of the knee specimens they analyzed. Further studies also reported a frequency of the ALL of more than 90% [13,16,37]. In contrast, the ALL was identified in only 45.5% of the dissected knee joints in the study of Runer et al. [33], in 50% of the knee specimens in the study of Stijak et al. [30], and in 60% of the dissected knee cadavers in the study of Roessler et al. [35]. Finally, other studies could not confirm the presence of a discrete ALL [10,23,38]. Shea et al. [39] analyzed the presence of the ALL in pediatric cadaveric specimens and described the ALL to be an inconsistent structure in the pediatric population. The inconsistency of the

presence of an ALL in these studies might be related to different dissection techniques that are connected to the identification of the femoral insertion of the ALL. Apart from anatomical studies, the presence of the ALL was also confirmed by ultrasound [40–42], MR imaging [29,43,44] as well as arthroscopic examination [45].

The ALL was found to be a separate anterolateral ligamentous structure which is distinguishable from the ITB and the anterolateral joint capsule in both embalmed and non-embalmed specimens. It showed an oblique course from the distal femur proximo-dorsal into a distal and ventral direction to the anterolateral tibia. The femoral insertion side was posterior and slightly proximal to the lateral femoral epicondyle and the femoral attachment overlapping the LCL. Other recent studies analyzing the anatomy of the anterolateral knee joint described the femoral insertion site of the ALL inconsistently. The present results were comparable to the findings of Daggett et al. [21], Dodds et al. [15], and Helito et al. [46]. Vincent et al. [13] described the location of the ALL femoral origin directly anterior to the popliteus tendon. Claes et al. [14] and Helito et al. [17] reported that the ALL femoral insertion was found ventral to the origin of the LCL. In the study of Caterine et al. [16], the ALL femoral insertion site was described ventro-distal to the LCL origin in 11 specimens and proximo-dorsal to the LCL origin in 8 specimens. Most recent studies described consistently that the tibial insertion site is located approximately midway between GT and the tip of the FH [14–17,30,33]. The results of the present study confirmed these findings. The identification of the insertion sites of the ALL was more challenging at the femoral insertion because it was more difficult to separate the thin ALL fibers from the thicker ITB in this part of the knee joint. In 15 of 23 knee joints, we found additional attachments to the lateral meniscus. They were thinner compared to the main part of the ligament. Helito et al. [47] and Kosy et al. [32] also described an additional meniscal insertion of the ALL. Our results were in line with the consensus of the ALL Expert Group [1].

The quantitative analysis of the ALL showed a mean length in knee extension of 39.13 mm, a mean width at the height of the joint line of 8.99 mm, and a mean thickness at the height of the joint line of 2.06 mm. The findings of previous studies were highly variable and depended on knee flexion and rotation angle as well as the exact knee position in which the measurements of the ALL were performed. Furthermore, the measurements might also be influenced by the dissection techniques and the inconsistent definition of the femoral and tibial insertions of the ALL. The findings for the ALL length are comparable to the results of Claes et al. [14], Helito et al. [17], Runer et al. [33], Caterine et al. [16], Kosy et al. [32], and Roessler et al. [35]. Dodds et al. [15] reported a considerably longer ALL length whereas Vincent et al. [13] found a shorter mean length of the ALL. Concerning the ALL width at joint line, the present results were comparable to those of Vincent et al. [13] and Stijak et al. [30]. The mean ALL width at the femoral and tibial insertion that was found for this study is almost similar to the results of Claes et al. [14] and Roessler et al. [35]. We found an ALL thickness of 2.06 mm being comparable to the findings of Vincent et al. [13]. Helito et al. [17] reported a clearly thicker ALL with 2.7 mm, whereas the majority of previous studies described a thinner ALL <2 mm [14,16,30,32,33,35].

Histology confirmed the morphological differences between the anterolateral knee joint capsule, the ALL, and the ITB. The ALL demonstrated organized dense connective tissue with a parallel and wavelike orientation of collagen fibrils characteristic for a ligamentous structure. Helito et al. [17,46] described similar histologic findings with the presence of dense connective tissue, arranged fibers, and little cellular material. Caterine et al. [16] reported dense, regularly organized collagenous bundles in histologic sections of the ALL and a dense network of peripheral nervous innervation. Zens et al. [48] found a unique crimping pattern for the ALL using polarization microscopy and therefore proved the existence of a ligamentous structure. Macchi et al. [49] also analyzed the ALL histologically and described a band of dense connective tissue mainly composed of collagen type I.

A study limitation is that it does not provide any biomechanical data. The quantitative analysis of the ALL was not performed at different knee flexion angles. The number of non-embalmed specimens ($n = 3$) is quite low.

The strength of the present study is that it is both a macroscopic and a microscopic approach to verify the presence of the ALL. In contrast to other recent studies, both embalmed and non-embalmed cadaveric specimens have been analyzed by the same technique which was feasible for the purpose. An acceptable number of dissections were performed to provide reliable results. A distinct quantitative and qualitative anatomic analysis of the ALL provided precise and relevant data concerning the insertion sites, length, width, and thickness of this ligament. This might be helpful during surgical restoration of the anterolateral soft tissue structures of the knee.

5. Conclusion

The analysis of the anterolateral knee anatomy reveals the ALL as a regular separate anterolateral ligamentous structure. It is distinguishable from the ITB and the anterolateral joint capsule in both embalmed and non-embalmed specimens. Histology of the ALL indicates typical ligamentous tissue which clearly differs from the anterolateral knee joint capsule and the thicker ITB.

Author's contributions

All authors read and approved the final manuscript.

Competing interests

The authors report no relationships that could be construed as a conflict of interest.

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Ethics committee letter

The specimens were taken from the body donation program of Saarland University Medical Center, Institute of Anatomy and Cell Biology. The bodies were donated by individuals who, prior to death, had given informed consent for their use for scientific and educational purposes. Therefore, an Ethics Committee agreement was not obtained for this study.

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