Number of fiber bundles in the fetal anterior talofibular ligament

Mutsuaki Edama1 · Tomoya Takabayashi1 · Hirotake Yokota1 · Ryo Hirabayashi1 · Chie Sekine1 · Sae Maruyama1 · Mayuu Syagawa1 · Ryoya Togashi1 · Yuki Yamada1 · Hiroki Otani2

Received: 14 July 2021 / Accepted: 5 August 2021 / Published online: 11 August 2021 © The Author(s), under exclusive licence to Springer-Verlag France SAS, part of Springer Nature 2021

Abstract
Purpose For the anterior talofibular ligament (ATFL), a three-fiber bundle has recently been suggested to be weaker than a single or double fiber bundle in terms of ankle plantarflexion and inversion braking function. However, the studies leading to those results all used elderly specimens. Whether the difference in fiber bundles is a congenital or an acquired morphology is important when considering methods to prevent ATFL damage. The purpose of this study was to classify the number of fiber bundles in the ATFL of fetuses.

Methods This study was conducted using 30 legs from 15 Japanese fetuses (mean weight, 1764.6 ± 616.9 g; mean crown-rump length, 283.5 ± 38.7 mm; 8 males, 7 females. The ATFL was then classified by the number of fiber bundles: Type I, one fiber bundle; Type II, two fiber bundles; and Type III, three fiber bundles.

Results Ligament type was Type I in 5 legs (16.7%), Type II in 21 legs (70%), and Type III in 4 legs (13.3%).

Conclusion The present results suggest that the three fiber bundles of the structure of the ATFL may be an innate structure.

Keywords Lateral ankle ligament injury · Gross anatomy · Ankle inversion braking function · Number of fiber bundles

Introduction
Injury to the lateral ligaments of the ankle is one of the most frequent sports injuries in both competitive and recreational sports. Roughly 70% involve injuries to the anterior talofibular ligament (ATFL) alone [8, 19, 24]. The morphological features of the lateral ligaments of the ankle are thought to be heavily involved in these injuries, and many anatomical studies of the ATFL in particular have been reported [11].

The main morphological features of the ATFL that have been investigated are the number of fiber bundles, fiber bundle length, fiber bundle width, and fiber bundle angle. However, differences in the number of fiber bundles have yet to be clarified. ATFL types with one, two, or three fiber bundles have been reported [12, 21], but other reports have suggested that types with three bundles do not exist [2–4, 13, 16, 20, 23, 25]. On the contrary, many recent reports using a large number of specimens have described three fiber bundles [5, 7, 9, 10].

The ATFL is reported to serve a major function in ankle plantarflexion and inversion control [15]. During plantarflexion, the superior fiber bundle tenses and the inferior fiber bundle relaxes, while during dorsiflexion, the superior fiber bundle relaxes and the inferior fiber bundle tenses [22]. In earlier studies, the ATFL was mainly described as the two fiber bundle type, and functions of the superior and inferior fiber bundles differed. In other earlier studies, however, fiber bundle length and total fiber bundle width were the same even when the number of fiber bundles differed, so functional differences were considered unlikely [22]. In all of these reports, the effects of differences in ATFL number of fiber bundles on function remained speculative. In recent studies, three-dimensional reconstructions of single, double, and triple fiber bundles of ATFL have been created. These were used to simulate and calculate ATFL strain during dorsiflexion (20°) and plantarflexion (30°) on the talocrural joint axis and inversion (20°) on the subtalar joint axis. The results suggested that an ATFL with three fiber bundles was weaker than those with single or double fiber bundles in...
terms of ankle plantarflexion and inversion braking function [6].

However, all specimens used in those previous studies were from elderly individuals. Whether the difference in fiber bundles represents a congenital or an acquired morphology is important when considering the prevention of ATFL damage. The purpose of this study was, therefore, to classify the number of fiber bundles of the ATFL in fetuses.

Materials and methods

Cadavers

The ligaments investigated were obtained from 30 legs of 15 Japanese fetuses (mean weight, 1764.6 ± 616.9 g; mean crown-rump length, 283.5 ± 38.7 mm; 8 males, 7 females). All specimens were fixed and preserved in 10% formalin and placed in water for 24 h before dissection. The human fetuses used in the present study belong to the Kyoto Collection of Human Embryos [14, 18] and all were Japanese. They were obtained through collaborating obstetricians and in accord with the Japanese Eugenic Protection Law (the present Law for the Protection of Mothers’ Bodies), mainly from healthy parents, rather than due to any clinical reasons of either embryos/fetuses or parents. Therefore, these embryos/fetuses can be considered to be representative of the normal Japanese intrauterine population. There were no congenital disorders in the central nervous system in the present cases. Mean gestational age of specimens was 23 weeks (range 21–25 weeks; all in the second trimester) as calculated from the crown-rump length [17]. This study was approved by the ethics committee at our institution (approval no. 4881).

Methods

In the ATFL dissection procedure, the skin, subcutaneous tissue, and crural fascia were removed, and the ATFL was carefully dissected (Fig. 1). The ATFL was then classified by the number of fiber bundles: Type I, one fiber bundle; Type II, two fiber bundles; and Type III, three fiber bundles.

Results

Classification by number of ATFL fiber bundles

The ligament type was Type I in 5 legs (16.7%), Type II in 21 legs (70%), and Type III in 4 legs (13.3%) (Fig. 2).

Discussion

This study examined differences in the number of ATFL fiber bundles in the fetus. To the best of our knowledge, no previous anatomical studies have reported on the number of ATFL fiber bundles in the fetus.

The classification based on differences in ATFL fiber bundles was Type I in 5 legs (16.7%), Type II in 21 legs (70%), and Type III in 4 legs (13.3%). Previous studies have reported Type I in 23–100% [3–5, 7, 9, 10, 12, 13, 16, 20, 21, 23, 25], and Type II in 0–75% [3–5, 7, 9, 10, 12, 13, 16, 20, 21, 23, 25], and Type III in 2–100% [2–5, 7, 9, 10, 12, 13, 16, 20, 21, 23, 25], and absent in 5% [16, 20]. Findings
have clearly been inconsistent. One major reason for this is thought to be differences in sample size, with 4–152 legs being examined [2–5, 7, 9, 10, 12, 13, 16, 20, 21, 23, 25] in the previous studies (Table 1). The results of the present study were similar to reports using a large number of specimens. The three bundles in the structure of the ATFL are already visible as early as the second trimester and resemble those seen in adults. The present results, therefore, suggest that the three fiber bundles in the ATFL may represent innate structures.

The ATFL is reported to serve a major function in ankle plantarflexion and inversion control [15]. One previous study [6] created three-dimensional reconstructions of the single, double, and triple fiber bundles of ATFL. The results suggested that a structure with three fiber bundles yielded an ATFL that was weaker than single or double fiber bundles in terms of ankle plantarflexion and inversion braking function. Our findings indicate that the three fiber bundles of the structure of ATFL may represent innate structural characteristics of the ligament. We, therefore, suggest that the presence of three fiber bundles in the ATFL may represent a risk factor for ATFL injury. In the future, we believe it will be necessary to perform biomechanical research using our basic data with in vivo samples, which may lead to development of the prevention of the ATFL injuries.

Some limitations to this study need to be considered. First, this study investigated only fetal ATFL in the second trimester. Investigations of fetal ATFL in the first trimester have also been attempted, but dissections proved difficult. Second, this study focused solely on differences in the number of ATFL fiber bundles and did not include quantitative analyses. Third, this study did not investigate bone morphology information such as the radius of curvature of the talus [1].

### Table 1 Summary of studies assessing ATFL morphology

| Study                     | Number of feet | Type I (%) | Type II (%) | Type III (%) | Absence (%) |
|---------------------------|----------------|------------|-------------|--------------|-------------|
| Burks et al. [2]          | 39             | Inferior band seen in some specimens |            |              |             |
| Milner et al. [12]        | 26             | 38         | 50          | 12           | 0           |
| Delfaut et al. [4]        | 4              | 25         | 75          | 0            | 0           |
| Taser et al. [20]         | 42             | 93         | 2           | 0            | 5           |
| Ugurlu et al. [21]        | 22             | 23         | 59          | 18           | 0           |
| Raheem et al. [16]        | 20             | 25         | 70          | 0            | 5           |
| Neuschwander et al. [13]  | 8              | 25         | 75          | 0            | 0           |
| Yildiz et al. [25]        | 46             | 75.6       | 24.4        | 0            | 0           |
| Clanton et al. [3]        | 14             | 50         | 50          | 0            | 0           |
| Wenny et al. [23]         | 17             | 100        | 0           | 0            | 0           |
| Edama et al. [5]          | 81             | 33         | 57          | 10           | 0           |
| Kakegawa et al. [9]       | 60             | 23.3       | 70          | 6.7          | 0           |
| Edama et al. [7]          | 110            | 31         | 60          | 9            | 0           |
| Kobayashi et al. [10]     | 152            | 43.4       | 54.6        | 2            | 0           |
| Present study             | 30             | 16.7       | 70          | 13.3         | 0           |
Conclusion

This study examined differences in the number of ATFL fiber bundles in fetuses. ATFL fiber bundles were classified as Type I in 5 legs (16.7%) Type II in 21 legs (70%), and Type III in 4 legs (13.3%). These results suggest that the three fiber bundles of the structure of the ATFL may be an innate structure.

Acknowledgements The authors wish to sincerely thank those who donated their bodies to science so that anatomical research could be performed. Results from such research can potentially improve patient care and increase humanity’s overall knowledge. Therefore, these donors and their families deserve our highest gratitude. We also thank the technical staff members and Satoru Hirano for helping in cadaver management. This study was supported by a Grant-in-Aid for Scientific Research (19K11358) from the Japanese Society for the Promotion of Science (JSPS).

Author contributions ME and TT contributed to the study design and data collection, and drafted the manuscript; HY, RH contributed to the data analysis and made critical revisions to the manuscript; CS, SM, RT, and YY made critical revisions to the manuscript; HO supervised the study, contributed to the analysis and interpretation of data, and made critical revisions to the manuscript. All authors read and approved the final manuscript prior to submission.

Funding None.

Availability of data and materials The data that support the findings of this study are available from the corresponding author upon reasonable request.

Declarations

Conflict of interest The authors declare that they have no conflict of interest.

Ethics approval and consent to participate Informed consent was obtained from the families of all subjects. All methods were carried out in accordance with the 1964 Declaration of Helsinki, and all cadavers used in the present study were legally donated for research purposes to the Kyoto University, Kyoto, and later transferred to the Shimane University, Izumo, Japan.

Consent for publication Not applicable.

References

1. Bonnel F, Toullec E, Mabit C, Tourné Y (2010) Chronic ankle instability: biomechanics and pathomechanics of ligaments injury and associated lesions. Orthop Traumatol Surg Res 96:424–432. https://doi.org/10.1016/j.otsr.2010.04.003
2. Burks RT, Morgan J (1994) Anatomy of the lateral ankle ligaments. Am J Sports Med 22:72–77
3. Clayton TO, Campbell KJ, Wilson KJ, Michalski MP, Goldsmith MT, Wijdicks CA, LaPrade RF (2014) Qualitative and quantitative anatomic investigation of the lateral ankle ligaments for surgical reconstruction procedures. J Bone Jt Surg Am 96:e98. https://doi.org/10.2106/jbjs.m.00798
4. Delfaut EM, Demondion X, Bouny N, Cotten H, Mestdagh H, Cotten A (2003) Multi-fasciculated anterior talo-fibular ligament: reassessment of normal findings. Eur Radiol 13:1836–1842. https://doi.org/10.1007/s00330-003-1853-4
5. Edama M, Kageyama I, Kikumoto T, Nakamura M, Ito W, Nakamura E, Hirabayashi R, Tokabayashi T, Inai T, Onishi H (2018) Morphological features of the anterior talofibular ligament by the number of fiber bundles. Ann Anat 216:69–74. https://doi.org/10.1016/j.aanat.2017.11.001
6. Edama M, Takabayashi T, Inai T, Kikumoto T, Ito W, Nakamura E, Hirabayashi R, Ikezu M, Kaneko F, Kageyama I (2019) The effect of differences in the number of fiber bundles of the anterior tibial ligament on ankle braking function: a simulation study. Surg Radiol Anat 41:69–73. https://doi.org/10.1007/s00276-018-2133-y
7. Edama M, Takabayashi T, Inai T, Kikumoto T, Ito W, Nakamura E, Hirabayashi R, Ikezu M, Kaneko F, Kageyama I (2019) Relationships between differences in the number of fiber bundles of the anterior talofibular ligament and differences in the angle of the calcaneofibular ligament and their effects on ankle-braking function. Surg Radiol Anat 41:675–679. https://doi.org/10.1007/s00276-019-02239-2
8. Fong DT, Hong Y, Chan LK, Yung PS, Chan KM (2007) A systematic review on ankle injury and ankle sprain in sports. Sports Med 37:73–94. https://doi.org/10.2165/00007256-200707010-00006
9. Kakugawa A, Morii Y, Tsuchiya A, Sumitomo N, Fukushima N, Morizumti T (2019) Independent attachment of lateral ankle ligaments: anterior talofibular and calcaneofibular ligaments—a cadaveric study. J Foot Ankle Surg 58:717–722. https://doi.org/10.1053/j.jfas.2018.12.009
10. Kobayashi T, Suzuki D, Kondo Y, Tokita R, Katayose M, Matsumura H, Fujimiyami M (2020) Morphological characteristics of the lateral ankle ligament complex. Surg Radiol Anat 42:1153–1159. https://doi.org/10.1007/s00276-020-02461-3
11. Matsui K, Takao M, Tochigi Y, Ozeki S, Glazebrook M (2017) Anatomy of anterior talofibular ligament and calcaneofibular ligament for minimally invasive surgery: a systematic review. Knee Surg Sports Traumatol Arthrosc 25:1892–1902. https://doi.org/10.1007/s00167-016-4194-y
12. Milner CE, Soames RW (1997) Anatomical variations of the anterior talofibular ligament of the human ankle joint. J Anat 191(Pt 3):457–458
13. Neuschwander TB, Indresano AA, Hughes TH, Smith BW (2013) Footprint of the lateral ligament complex of the ankle. Foot Ankle Int 34:582–586. https://doi.org/10.1177/1071107012466851
14. Nishimura H, Takanohi, Tanimura T, Yasuda M (1968) Normal and abnormal development of human embryos: first report of the analysis of 1,213 intact embryos. Teratology 1:281–290. https://doi.org/10.1002/tera.1420010306
15. Ozeki S, Kitaoaka H, Uchiyama E, Luo ZP, Kaufman K, An KN (2006) Ankle ligament tensile forces at the end points of passive circumferential rotating motion of the ankle and subtalar joint complex. Foot Ankle Int 27:965–969
16. Raheem OA, O’Brien M (2011) Anatomical review of the lateral collateral ligaments of the ankle: a cadaveric study. Anat Sci Int 86:189–193. https://doi.org/10.1007/s12565-011-0109-7
17. Sahota DS, Leung TY, Leung TN, Chan OK, Lau TK (2009) Fetal crown-rump length and estimation of gestational age in an ethnic Chinese population. Ultrasound Obstet Gynecol 33:157–160. https://doi.org/10.1002/ujog.6252
18. Shita K (2018) Study of normal and abnormal prenatal development using the Kyoto collection of human embryos. Anat Rec (Hoboken) 301:955–959. https://doi.org/10.1002/ar.23790
19. Sittler M, Ryan J, Wheeler B, McBride J, Arciero R, Anderson J, Horodynski M (1994) The efficacy of a semirigid ankle stabilizer
to reduce acute ankle injuries in basketball. A randomized clinical study at West Point. Am J Sports Med 22:454–461. https://doi.org/10.1177/036354659402200404

20. Taser F, Shafiq Q, Ebraheim NA (2006) Anatomy of lateral ankle ligaments and their relationship to bony landmarks. Surg Radiol Anat 28:391–397. https://doi.org/10.1007/s00276-006-0112-1

21. Ugurlu M, Bozkurt M, Demirkale I, Comert A, Acar HI, Tekdemir I (2010) Anatomy of the lateral complex of the ankle joint in relation to peroneal tendons, distal fibula and talus: a cadaveric study. Eklem Hastalik Cerrahisi 21:153–158

22. van den Bekerom MP, Oostra RJ, Golano P, van Dijk CN (2008) The anatomy in relation to injury of the lateral collateral ligaments of the ankle: a current concepts review. Clin Anat 21:619–626. https://doi.org/10.1002/ca.20703

23. Wenny R, Duscher D, Meytap E, Weninger P, Hirtler L (2015) Dimensions and attachments of the ankle ligaments: evaluation for ligament reconstruction. Anat Sci Int 90:161–171. https://doi.org/10.1007/s12565-014-0238-x

24. Woods C, Hawkins R, Hulse M, Hodson A (2003) The Football Association Medical Research Programme: an audit of injuries in professional football: an analysis of ankle sprains. Br J Sports Med 37:233–238. https://doi.org/10.1136/bjsm.37.3.233

25. Yıldız S, Yalcin B (2013) The anterior talofibular and calcaneofibular ligaments: an anatomic study. Surg Radiol Anat 35:511–516. https://doi.org/10.1007/s00276-012-1071-3

Publisher’s Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.