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Age- and Sex-Specific Joint Biomechanics in Response to Partial and Complete Anterior Cruciate Ligament Injury in the Porcine Model

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Context: Pediatric anterior cruciate ligament (ACL) injury rates are increasing and are highest in female adolescents. Complete ACL tears are typically surgically reconstructed, but few guidelines and very limited data exist regarding the need for surgical reconstruction or rehabilitation for partial ACL tears in skeletally immature patients.

Objective: To evaluate the effects of partial (anteromedial bundle) and complete ACL transection on joint laxity and tissue forces under anterior and rotational loads in male and female stifle joints throughout skeletal growth in the porcine model.

Design: Descriptive laboratory study.

Setting: Laboratory.

Patients or Other Participants: We studied 60 male and female Yorkshire crossbreed pigs aged 1.5, 3, 4.5, 6, and 18 months (n = 6 pigs per age per sex).

Main Outcome Measure(s): Joint laxity was measured in intact, anteromedial bundle-transected, and ACL-transected joints under applied anterior-posterior drawer and varus-valgus torque using a robotic testing system. Loading of the soft tissues in the stifle joint was measured under each condition.

Results: Anterior-posterior joint laxity increased by 13% to 50% ($P < .05$) after anteromedial bundle transection and 75% to 178% ($P < .05$) after ACL transection. Destabilization after anteromedial bundle transection increased with age ($P < .05$) and was greater in late female than late male adolescents ($P < .05$). In anteromedial bundle-transected joints, the posterolateral bundle resisted the anterior load. In ACL-transected joints, the medial collateral ligament (MCL) contribution was largest, followed by the medial meniscus. The MCL contribution was larger and the medial meniscus contribution was smaller in male versus female specimens.

Conclusions: Partial ACL transection resulted in moderate increases in joint laxity, with the remaining bundle performing the primary ACL function. Destabilization due to partial ACL transection (anteromedial bundle) was largest in late adolescent joints, indicating that operative treatment should be considered in active, late-adolescent patients with this injury. Increased forces on the MCL and medial meniscus after ACL transection suggested that rehabilitation protocols may need to focus on protecting these tissues.

Key Words: knee, transection studies, sex factors

Key Points

- Partial anterior cruciate ligament (ACL) transection resulted in moderate increases in joint laxity, which increased throughout skeletal growth and were larger in late female adolescents compared with late male adolescents.
- The primary role of the ACL was supported by its remaining bundle after partial ACL transection, but loads were highest on the medial collateral ligament, followed by the medial meniscus, after complete ACL transection.
- In the ACL-deficient knee, loads placed on the medial collateral ligament were higher, whereas loads placed on the medial meniscus were lower in male than in female specimens.

Incidences of anterior cruciate ligament (ACL) injuries and reconstructions in pediatric patients are increasing at higher rates compared with adults.[1] Furthermore, injury rates appear to be sex dependent, such that female adolescent athletes experienced the highest risk of ACL injury.[2] It is well established that complete, or full-thickness, ACL tears often lead to major anterior and rotational knee instability.[3–5] An ACL injury can occur in conjunction with tears of the medial collateral ligament (MCL), lateral collateral ligament (LCL), or menisci, a clear example of how multiple tissues are loaded during movement and how injury to 1 tissue may alter the loading of other tissues.[6] In addition, although some groups advocated for nonoperative treatment in children to avoid growth-plate disturbances,[7] reports of secondary meniscal and chondral injuries presenting months after initial ACL injury are 2 to 12 times higher in pediatric patients who delay ACL reconstruction.[8,9] Therefore, early reconstruction is typically recommended for young athletes.[3]

However, the decision regarding surgical reconstruction for partial ACL tears in young patients is less clear. Partial ACL tears account for an estimated 10% to 27% of diagnosed ACL injuries[10] and result in more moderate knee instabilities.[4] Of interest, this figure likely underestimates the frequency of partial ACL tears due to challenges in early clinical detection.[11] Partial tears can occur primarily in 1 of the functional bundles of the ACL—the anteromedial (AM) or posterolateral (PL) bundle—and the distributions of partial ACL tears are reported as approximately 60% AM bundle tears and 40% PL bundle tears.[12,13] As summarized by McClincy and Hayworth,[3] partial ACL tears can be treated nonoperatively, with standard surgical reconstruction, or with selective bundle reconstruction in which the injured bundle is reconstructed while the uninjured bundle is left intact. No consensus currently exists regarding treatment approaches for partial ACL tears in young patients, and only 2 published studies have examined treatment options for partial ACL tears in skeletally immature patients.[12,14] In each, the likelihood of progression to a complete ACL tear was higher in adolescent patients than in children[12] or adults,[14] indicating that age may be an important factor in treatment considerations.

Factors such as changes in joint laxity and load placed on other tissues after partial ACL injury are critical to consider when making treatment decisions regarding surgical intervention and rehabilitation. Typically, a side-to-side difference of $>$3 mm in an anterior drawer test is considered indicative of an ACL tear.[5,15] Partial ACL tears may not always exceed the 3-mm side-side difference in an anterior drawer test,[3,4] yet subsequent meniscal injuries have been reported in 20% of young, active patients.[14] Furthermore, preclinical data from a skeletally mature ovine model suggested that partial ACL tears resulted in a greater progression of osteoarthritis compared with healthy control animals.[16] The effect of partial ACL tears on the knee joint in skeletally immature patients remains largely uncharacterized.

Previous researchers[17,18] showed that complete ACL injury led to increased joint laxity and in situ forces on other passive stabilizers, such as the MCL, LCL, and medial meniscus in skeletally mature cadaveric knee joints using 6 degrees-of-freedom robotic testing systems. Such systems can apply loads to joints while measuring translations and then repeat the translations after dissection of tissues to measure in situ tissue forces.[19,20] These types of data are generally impossible to obtain from human pediatric patients due to the paucity of cadaveric specimens. Therefore, models of the pediatric ACL injury are necessary. Earlier investigators[21] using a skeletally immature female porcine model found an age-dependent effect of partial ACL injury on joint laxity. However, sex differences in injury rates in skeletally immature patients point to the need to compare the effects of ACL injury between sexes. Furthermore, recent work from our group[22] in a porcine model demonstrated sex differences in normal ACL bundle function during simulated clinical examinations, indicating that the AM bundle became functionally dominant over the PL bundle during early adolescence in male specimens but during late adolescence in female specimens. These differences in bundle contributions may influence joint biomechanics in response to ACL injuries, particularly partial ACL tears concentrated in 1 bundle.

The objective of our study was to evaluate the effects of partial and complete ACL injury on joint laxity and tissue forces under applied anterior and rotational loads in male and female stifle joints throughout skeletal growth in the porcine model. We hypothesized that partial and complete ACL injuries would lead to increased joint laxity and increased loads placed on other soft tissues in an age- and sex-dependent manner. To test this hypothesis, we biomechanically tested stifle (knee) joints from male and female pigs at 5 ages, ranging from early youth to late adolescence, using a robotic testing system. Joint laxity and tissue forces were measured under applied loads in intact, AM bundle-transected, and ACL-transected joints.

METHODS

The animals were cared for by the North Carolina State University Swine Educational Unit, and the experimental protocols were approved by the North Carolina State University Institutional Animal Care and Use Committee. Hind limbs were collected from a total of 60 male and female Yorkshire crossbreed pigs at early youth (1.5 months), youth (3 months), early adolescence (4.5 months), adolescence (6 months), and late adolescence (18 months) immediately after euthanasia ($n = 6$ per age per sex).[23] Stifle joints were isolated, wrapped in saline-soaked gauze, and stored at –20°C. The data reported here were part of a larger study in which we had previously supplied data regarding the effect of partial and complete ACL injury on female joints[21] and comparing healthy joint function in males and females.[22,24]

The joints were thawed at room temperature and imaged using a 7.0-T MAGNETOM scanner (Siemens Healthineers AG) with a double-echo steady-state sequence (DESS, flip angle = 25°, repetition time = 17 milliseconds, echo time = 6 milliseconds, voxel size = $0.42 \times 0.42 \times 0.4$ mm; no gap between slices). Joints were stored at –20°C after imaging. The ACL and MCL were manually segmented from magnetic resonance images using commercial software (version S-2021.06; Simpleware ScanIP, Synopsys) and exported as surface models. Using a custom MATLAB code (The MathWorks, Inc), we rotated the models to align the primary axis vertically. The cross-sectional area (CSA) was calculated perpendicular to the primary axis and averaged over the central 50% of the model.

In preparation for biomechanical testing, the joints were thawed at room temperature. Soft tissue surrounding the joint capsule was removed, and the femoral and tibial diaphyses were fixed in custom molds using a fiberglass-reinforced epoxy (Everglass; Evercoat). Once fixed, the joints were wrapped in saline-soaked gauze and stored at –20°C. They were thawed at room temperature 1 day before biomechanical testing.

Biomechanical testing was performed using a 6 degrees-of-freedom robotic testing system (models KR 300 R2500, KR C4, 130; KUKA Robotics) equipped with a universal force-moment sensor (model Omega160 IP65; ATI Industrial Automation). Systems were integrated using the simVITRO software package knee module (Cleveland Clinic). For each joint, the femur was fixed to a testing platform, and the tibia was fixed to the robotic system using

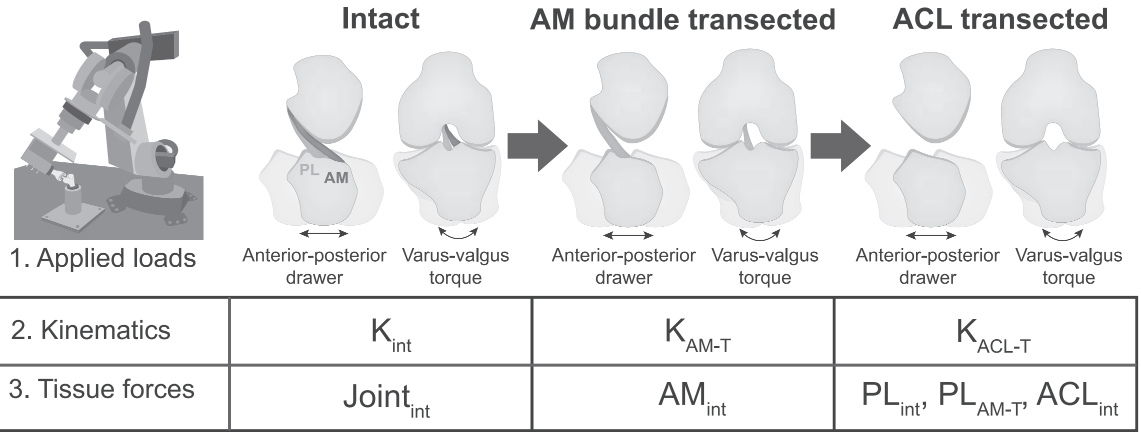

Figure 1. Robotic testing methods. Anterior-posterior and varus-valgus loads were applied to joints in the intact, anteromedial bundle-transected (AM-T), and anterior cruciate ligament-transected (ACL-T) states. For each loading condition, kinematics were recorded. Then kinematics were replayed while recording forces. Soft tissue force contributions under each set of kinematics were calculated, as described in Supplemental Table S1. Notably, force contributions from the AM bundle, posterolateral (PL) bundle, and ACL in the intact state and the PL bundle in the AM-T state were calculated from testing, as indicated. Subscripts represent the kinematic state.

custom clamps. Joints were lightly wrapped in saline-soaked gauze throughout testing. A joint coordinate system was established using a 3-dimensional digitizer (model G2X MicroScribe; GoMeasure3D, Inc).

To simulate clinical examinations, we applied anterior-posterior (AP) drawer forces followed by varus-valgus (VV) moments to the tibia at 60° of flexion (Figure 1; see Supplemental Table S1, available online at http://dx.doi.org/10.4085/1062-6050-0565.21.S1). Maximum applied forces (20, 40, 80, 100, and 140 N for AP drawer forces) and VV moments (1, 2, 4, 5, and 7 N·m) were scaled to femoral cortical CSA for each joint age group (1.5-, 3-, 4.5-, 6-, and 18-months old, respectively). Loads were selected to account for 7-fold increases in bone size from 1.5 to 18 months, and prior work[25] confirmed that these loads reached the linear region of the force-translation curve for the ACL. During loading, flexion and internal rotations were fixed, and loads in the remaining 3 degrees of freedom were minimized. Paths under the applied loads were recorded for the intact joint and then the kinematics were replayed while forces were recorded. Next, the joint capsule was opened, and the AM bundle of the ACL was transected. Preliminary data showed that the joint capsule accounted for minimal force (see Supplemental Table S2) and the bundle interaction was negligible under anterior drawer testing in intact knees. Loads were applied to the AM bundle-transected (AM-T) joint while recording positions. Both the intact and AM-T kinematic paths were replayed while forces were recorded. The PL bundle was then transected, and loads were applied to the ACL-transected (ACL-T) joint while recording positions. The intact, AM-T, and ACL-T kinematic paths were replayed while recording forces. Next, the MCL was transected, and the 3 sets of kinematic paths were replayed while recording forces. This process was repeated for the LCL, posterior cruciate ligament (PCL), medial meniscus, and lateral meniscus.

Data from robotic testing were analyzed in MATLAB. The maximum AP tibial translation (APTT) and VV rotation were calculated for each joint in the intact, AM-T, and ACL-T states. Using the principle of superposition,[20] the force contribution of each soft tissue was calculated as a percentage of the applied anterior force in the knee joint under maximum anterior translation for each kinematic state (see Supplemental Table S1).

Statistical analysis was performed using Prism (version 9.3.0; GraphPad). We examined APTT and VV rotation for the male and female joints individually using 2-way analysis of variance (ANOVA) with age and injury state as main effects (injury state as a repeated measure) and Tukey post hoc tests to compare injury states within each age group (overall $\alpha = .05$). To compare the effect of injury between sexes, the differences in APTT and VV rotation between the injured and intact states were calculated for each joint. We assessed these δ values using 2-way ANOVA with age and sex as main effects and Sidak post hoc tests to compare sexes within each age group (overall $\alpha = .05$). Force contributions under anterior drawer testing were evaluated for significance using 1-sample *t* tests comparing means to 0 for the AM and PL bundle in the intact state, the PL bundle in the AM-T state, and the MCL and medial meniscus in the ACL-T state for each age group by sex ($\alpha = .005$ to adjust for multiple comparisons). Differences in these tissue contributions across age and sex were compared using 2-way ANOVA with age and sex as main effects (overall $\alpha = .05$). Linear regressions were performed to assess the relationship between the intact APTT and ACL CSA and between the change in APTT after ACL transection and MCL CSA.

RESULTS

Joint laxity generally increased after AM bundle transection and ACL transection in male and female pigs throughout growth, as is seen in the average force-translation curves (Figures 2 and 3). Transection of the AM bundle led to only moderate (13%–50%) increases in APTT (Figure 4A; see Supplemental Table S3), whereas complete ACL transection unsurprisingly led to substantial (75%–178%) increases in APTT (Figure 4B; see Supplemental Table S3). Age produced no clear effect on overall APTT in the intact joint. However, the instabilities caused by both AM bundle transection (Figure 4C) and complete ACL transection (Figure 4D) increased throughout skeletal growth (*P* values $<$ .001 with main effects of age for both;

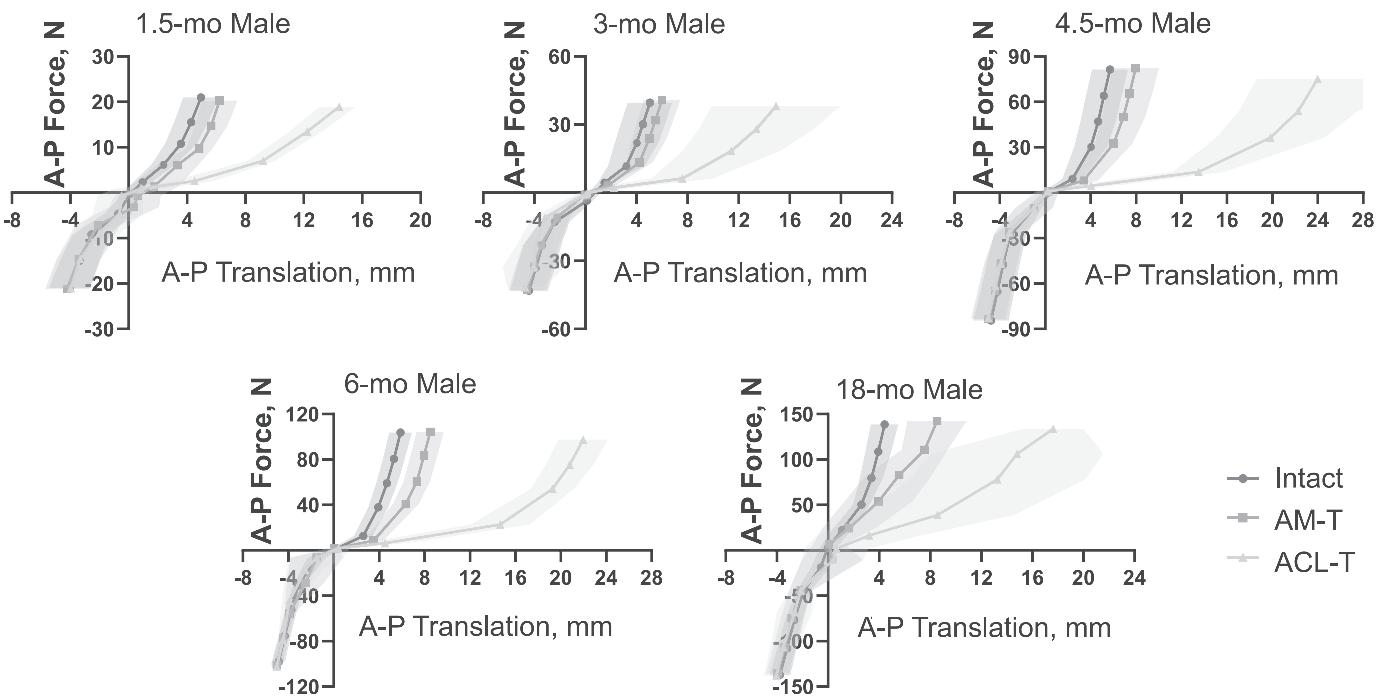

Figure 2. Mean anterior-posterior (A-P) force-translation curves for male joints in each age group under intact, anteromedial bundle-transected (AM-T), and anterior cruciate ligament-transected (ACL-T) states. Shaded regions represent the standard deviation in joint translation, interpolated between data points.

see Supplemental Table S4). The APTT due to AM bundle transection increased 2.6-fold in males and 3.9-fold in females from early youth (1.5 months) to late adolescence (18 months). Meanwhile, APTT after ACL transection increased approximately 2-fold in both males and females from early youth (1.5 months) to late adolescence (18 months). Clinically, >3-mm side-side asymmetry is considered indicative of an ACL tear,[5,15] and this difference was exceeded only in 6-month female (3.1 mm) and 18-month male and female specimens (3.6 mm and 5.1 mm, respectively). In terms of differences due to sex, the change in APTT after AM bundle transection was similar between males and females from early youth to adolescence ($P = .4$–1.0 between sexes at 1.5–6 months) but 33% greater in

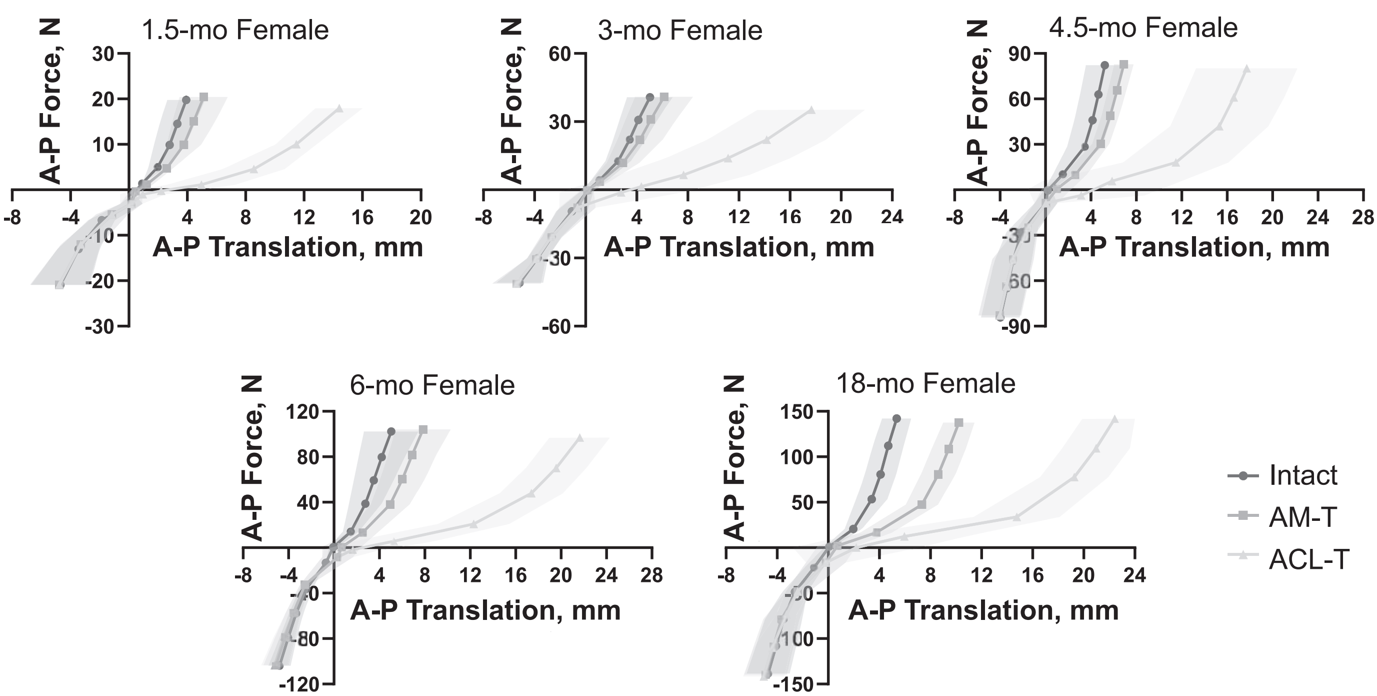

Figure 3. Mean anterior-posterior (A-P) force-translation curves for female joints in each age group under intact, anteromedial bundle-transected (AM-T), and anterior cruciate ligament-transected (ACL-T) states. Shaded regions represent the standard deviation in joint translation, interpolated between data points.

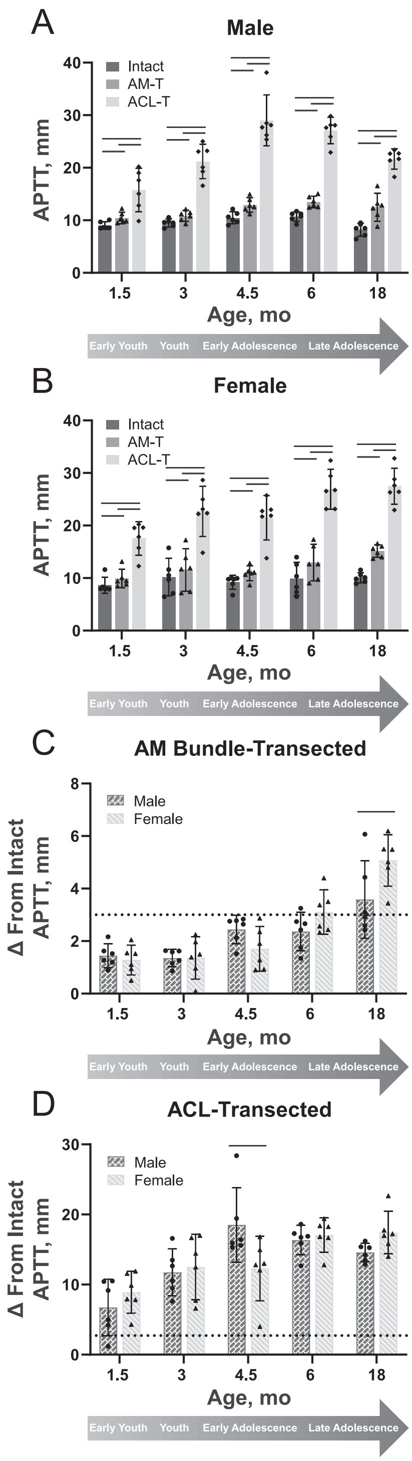

Figure 4. Anterior-posterior tibial translation (APTT) increased substantially after anterior cruciate ligament (ACL) transection in A, males and B, females at all ages but less so after anteromedial (AM) bundle transection alone. Data from intact joints has been previously reported.[22,24] C, The change in APTT after AM bundle transection (relative to the intact joint) increased throughout growth and was greater in females than in males during late adolescence (18 months). D, The change in APTT after ACL transection exceeded the clinical threshold for injury diagnosis at all ages. Dashed line represents clinical threshold (3-mm side-side difference) for ACL injury diagnosis. Bars represent $P < .05$ between groups and within ages. Individual data points presented along with mean ± 95% CI.

females at late adolescence ($P = .009$ between sexes at 18 months). The change in APTT after ACL transection was similar between males and females ($P = .5$–1.0 between sexes at 1.5, 3, 6, and 18 months) apart from early adolescence, when it was 20% greater in male specimens ($P = .01$ between sexes at 4.5 months).

Similarly, ACL transection led to substantial VV rotational destabilizations in males (10%–38%) and females (20%–72%; P values $< .05$ between intact and ACL-T within all age groups, apart from 4.5- and 18-month-old males; Figure 5A and B; see Supplemental Table S5). However, AM bundle transection had a much smaller effect on VV rotational stability in both males (3%–12%) and females (3%–21%; P values $< .05$ between intact and AM bundle transection for 1.5- and 4.5-month-old males and 3- to 18-month-old females). Additionally, VV rotation in all states decreased by 65% to 75% in males and females throughout skeletal growth ($P < .001$, main effect of age). The change in VV rotation after AM bundle transection was affected by sex only during early youth, when the effect of partial injury was 122% greater in male than female specimens ($P < .001$ between males and females at 1.5 months; Figure 5C; see Supplemental Table S6). Additionally, the change in VV rotation after ACL transection was not influenced by age ($P = .14$) or sex ($P = .93$; Figure 5D; see Supplemental Table S6).

In the intact knee, the AM and PL bundles of the ACL were the dominant restraints against applied anterior load under maximum anterior translation in both sexes throughout skeletal growth (Figure 6A and B; see Supplemental Table S7). With both bundles, the ACL restrained 94% to 110% of the applied anterior load in the joint in both sexes at all ages, while contributions from the MCL, LCL, PCL, and menisci were minimal (<5%). (Contributions can exceed 100% due to opposing forces in other tissues.) The force contribution of the AM bundle under anterior drawer testing was significant (relative to 0) in male and female specimens at all ages (P values $< .005$), while force contributions of the PL bundle were only significant during youth in males ($P = .005$ at 3 months) and females ($P = .001$ at 1.5 and 3 months). The relative force contributions of the bundles shifted throughout skeletal growth, as previously reported in greater detail.[22,24] The AM bundle restraint against anterior drawer testing increased from 57% in males and 44% in females during youth (3 months) to 87% in males and 91% in females during late adolescence (18 months; $P < .0001$ main effect of age; Figure 6C). Additionally, the AM bundle contribution was greater in males than in females by 14% across all ages ($P = .003$ for main effect of sex). The PL bundle contribution decreased correspondingly from youth to late adolescence ($P < .0001$ for main effect of age; Figure 6D) and was smaller in male than female specimens by 14% across all ages ($P = .009$ for main effect of sex). The interaction between age and sex was not significant for either bundle contribution ($P > .4$).

In the AM-T joint, the PL bundle restrained nearly all the applied anterior force under maximum anterior translation in all joints (Figure 7A and B; see Supplemental Table S8). The PL bundle contribution under applied anterior load (88%–109%) was significant in AM-T joints across all groups ($P < .0001$ versus 0 for all), while contributions from the MCL, LCL, PCL, and menisci were minimal (<5%). Additionally, the PL bundle restraint against

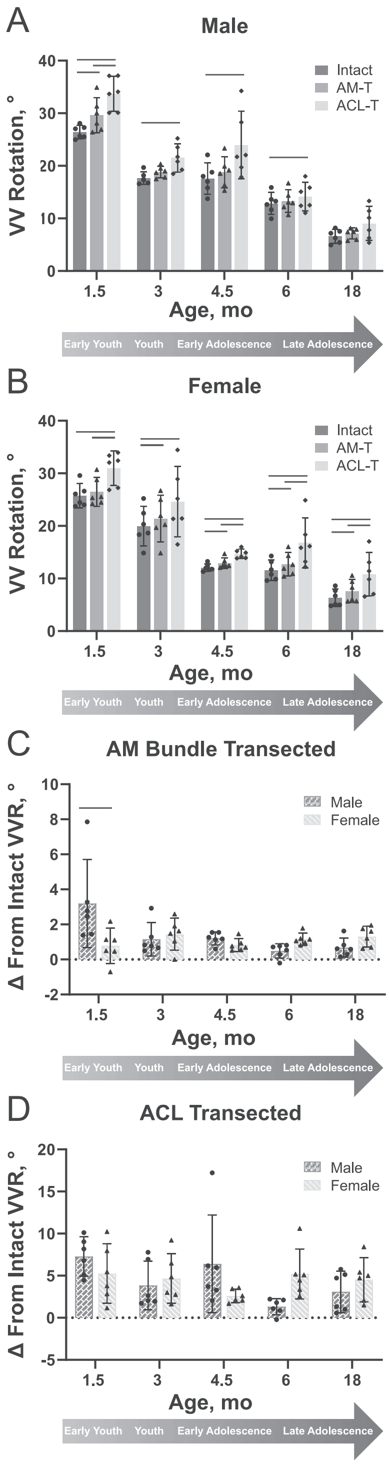

Figure 5. Anterior cruciate ligament (ACL) transection led to substantial increases in varus-valgus rotation (VVR) in A, males and B, females at all ages, whereas anteromedial (AM) bundle transection had a smaller effect on VVR. The change in VVR after C, AM bundle transection (relative to the intact joint) and D, ACL transection. Bars represent $P < .05$ between groups, within ages. Individual data points presented along with mean ± 95% CI.

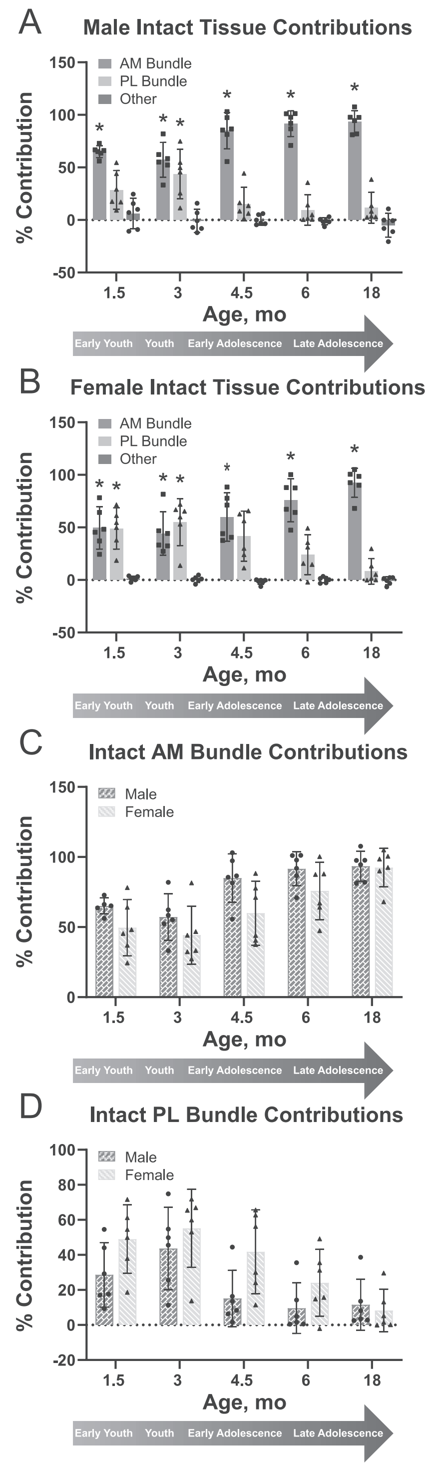

Figure 6. Soft tissue contributions, as a percentage of the applied force restrained by the joint under maximum anterior translation in the intact joint. The anteromedial (AM) and posterolateral (PL) bundles of the anterior cruciate ligament (ACL) were the primary restraints against anterior drawer throughout skeletal growth in A, males and B, females. * $P < .005$ versus 0 by 1-sample *t* test. Intact joint bundle contribution data has been previously reported as a percentage of ACL force.[22,24] C, The AM bundle contribution increased throughout growth ($P < .0001$) and was greater in males than in females ($P = .003$), whereas D, the PL bundle contribution decreased throughout growth ($P < .0001$) and was smaller in males than in females ($P = .009$). Individual data points presented as mean ± 95% CI.

anterior drawer testing in the AM-T joint did not vary with age ($P = .4$) or sex ($P = .8$; Figure 7C).

In the ACL-T joint, tissue contributions to anterior drawer restraint were more variable across specimens, but the MCL was the largest contributor, followed by the

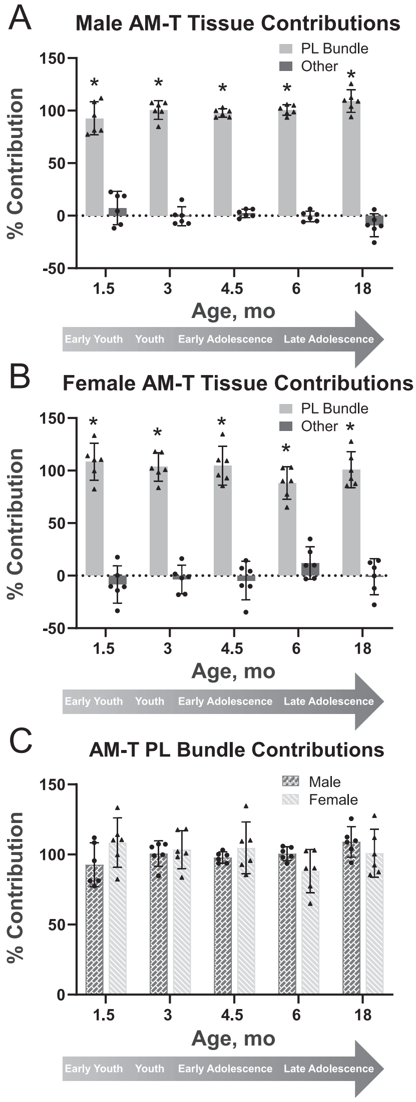

Figure 7. Soft tissue contributions, as a percentage of the applied force restrained by the joint under maximum anterior translation in the anteromedial bundle-transected (AM-T) joint. The posterolateral (PL) bundle was the primary restraint against anterior drawer testing in A, males and B, females. * *P* < .005 versus 0 by 1-sample *t* test. C, The PL bundle contribution in the AM-T joint did not vary with age or sex. Individual data points presented as mean ± 95% CI.

medial meniscus in most cases (Figure 8A and B; see Supplemental Table S9). In males, the MCL restrained 62% to 94% of the applied anterior load across all ages ($P <$ .005 versus 0 for all). Similarly, in females, the MCL contribution restrained 55% to 71% of the applied anterior load (P values < .005 for 1.5, 3, 6, and 18 months; $P = .007$ for 4.5 months). The medial meniscus was the next largest contributor to anterior drawer restraint for both sexes. The medial meniscus contribution ranged from 6% to 18% at individual age groups for males ($P = .02$–.8 versus 0 for all ages) and 15% to 35% for females ($P = .008$–.09 versus 0 for all ages). Between sexes, the MCL contribution was larger in males than in females by 18% ($P = .005$ main effect of sex) and did not vary throughout skeletal growth ($P = .4$ for main effect of age; Figure 8C). The medial meniscus contribution was smaller in male than in female specimens by 16% ($P = .004$ for main effect of sex) and did

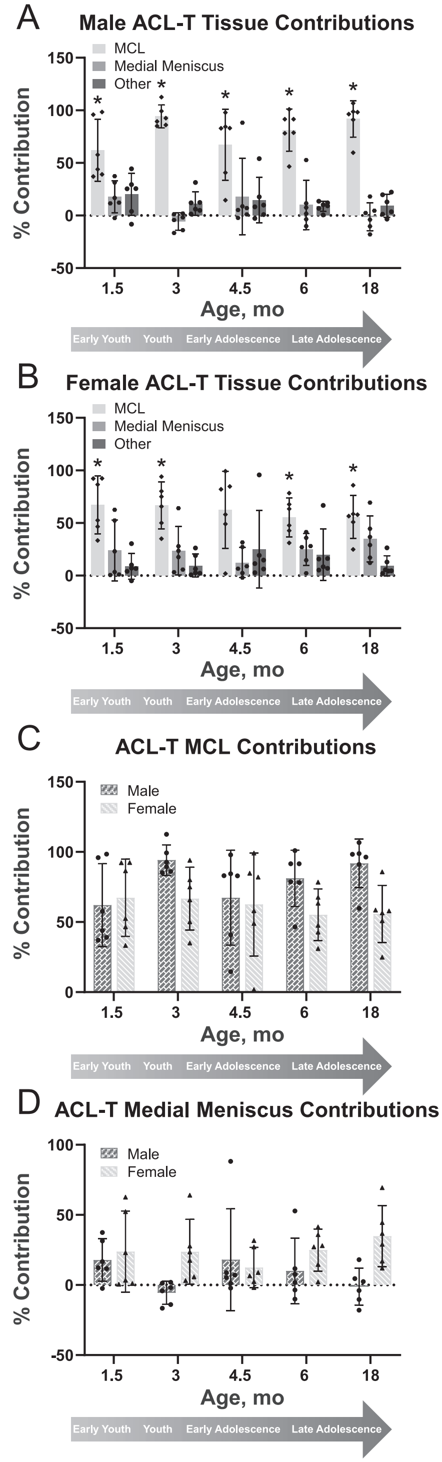

Figure 8. Soft tissue contributions, as a percentage of the applied force restrained by the joint under maximum anterior translation in the anterior cruciate ligament-transected (ACL-T) joint. The medial collateral ligament (MCL) was the largest restraint against anterior drawer testing for most joints, with lesser contributions from the medial meniscus in A, males and B, females. * *P* < .005 versus 0 by 1-sample *t* test. C, MCL contribution in the ACL-T joint was greater in males than in females (*P* = .005), while D, medial meniscus contribution was smaller in males than in females (*P* = .004). Individual data points presented as mean ± 95% CI.

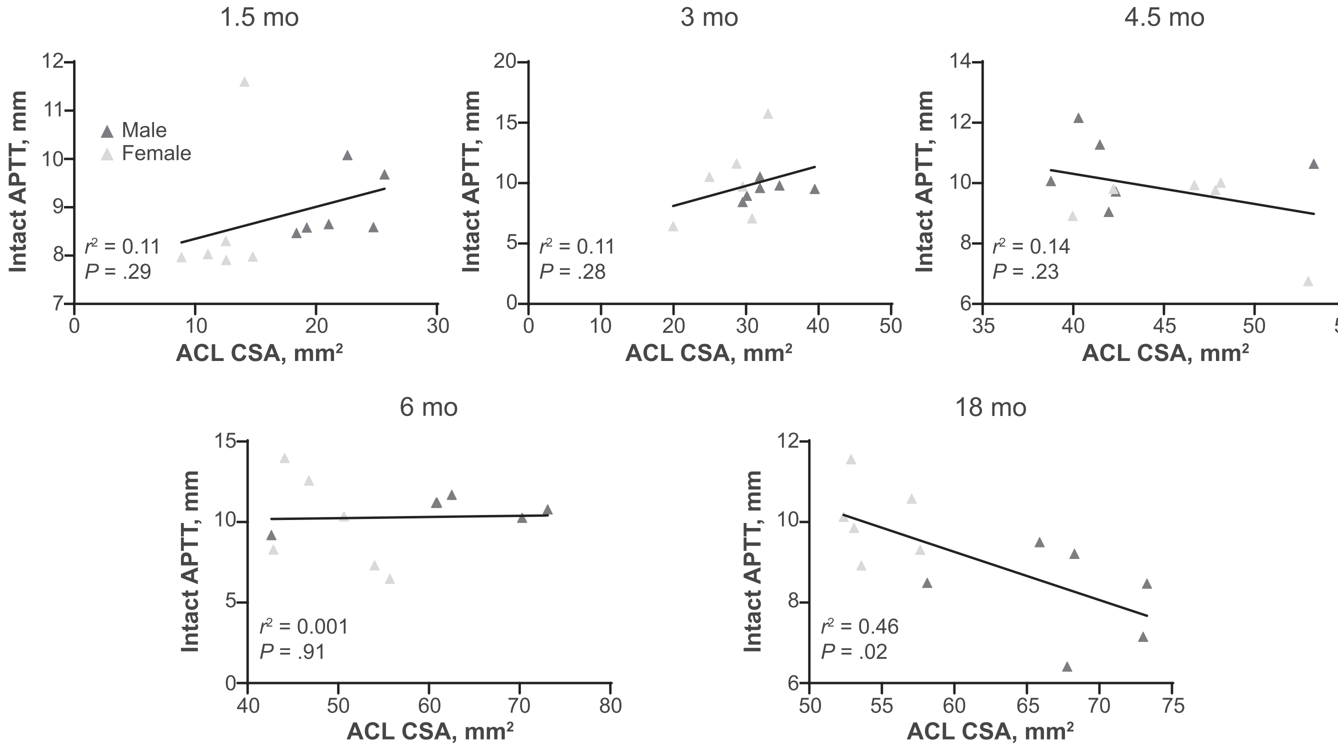

Figure 9. Intact joint anterior-posterior tibial translation (APTT) versus anterior cruciate ligament (ACL) cross-sectional area (CSA), plotted by age group. Statistical results from linear regression shown in graph corner.

not vary throughout skeletal growth ($P = .7$ for main effect of age; Figure 8D). Contributions of the LCL, PCL, and lateral meniscus remained low (<7% in males and <16% in females).

Linear regressions between intact-joint APTT and ACL CSA within age groups were not significant (P values $>$ .05) for early youth through adolescence (1.5–6 months; Figure 9). However, at late adolescence (18 months), the APTT and ACL CSA regression was significant ($P = .015$) with a negative slope. Additionally, across all ages, normalized joint laxity (APTT normalized to AP tibial width) was inversely related to ACL CSA for males and females ($P < .05$; see Supplemental Figure S1). Linear regressions between the change in joint laxity after ACL transection versus MCL CSA were not significant within age groups (Figure 10) but approached significance, with a negative slope, at late adolescence ($P = .054$ for the 18-month-old group). Across all ages, MCL CSA showed little relation to normalized ACL-T APTT (see Supplemental Figure S2) and the change in APTT after ACL transection (see Supplemental Figure S3).

DISCUSSION

We found age- and sex-dependent increases in joint laxity in response to AM bundle transection but not complete ACL transection in a skeletally immature porcine model, partially confirming our hypothesis of age- and sex-dependent responses to injury. Specifically, the change in APTT after AM bundle transection increased with age and was larger in female than in male pigs during late adolescence. In addition, ACL transection led to several-fold increases in APTT, which remained similar between sexes and across ages. The VV rotation increased 0.5° to 3° after AM transection and 1.3° to 7° after ACL transection. The changes in VV rotation due to injury were similar across ages and sexes. Contrary to our hypothesis, the forces experienced by tissues after AM bundle transection and ACL transection did not significantly differ with age or sex. The restraint against applied anterior loads in healthy knee joints was primarily split between the AM and PL bundles of the ACL; the AM bundle contribution increased throughout skeletal growth and was greater in male than in female specimens. After AM bundle transection, the PL bundle accounted for the majority of the applied anterior force in all cases. However, after ACL transection, the MCL became the largest restraint against anterior drawer testing, followed by the medial meniscus. Furthermore, the MCL contribution was greater while the medial meniscus contribution was smaller in males compared with females.

We expected that joint stability would be inversely related to the CSA of the primary stabilizing tissue (ie, the ACL in intact joints and the MCL in ACL-deficient joints). However, although ACL CSA increased and intact joint laxity decreased throughout skeletal growth, linear regressions within age groups indicated that these factors were related only during late adolescence. We[22] have reported, similarly, that ACL in situ stiffness was positively correlated with ACL CSA across skeletal growth but only during late adolescence when analyzed by specific age groups. Additionally, despite being just above the significance threshold, MCL CSA appeared inversely related to the joint instability caused by ACL injury only during late adolescence. This relationship was not observed when comparing all age groups. Together, these results indicated that the expected structure-function relationship held true only once animals reached late adolescence. However,

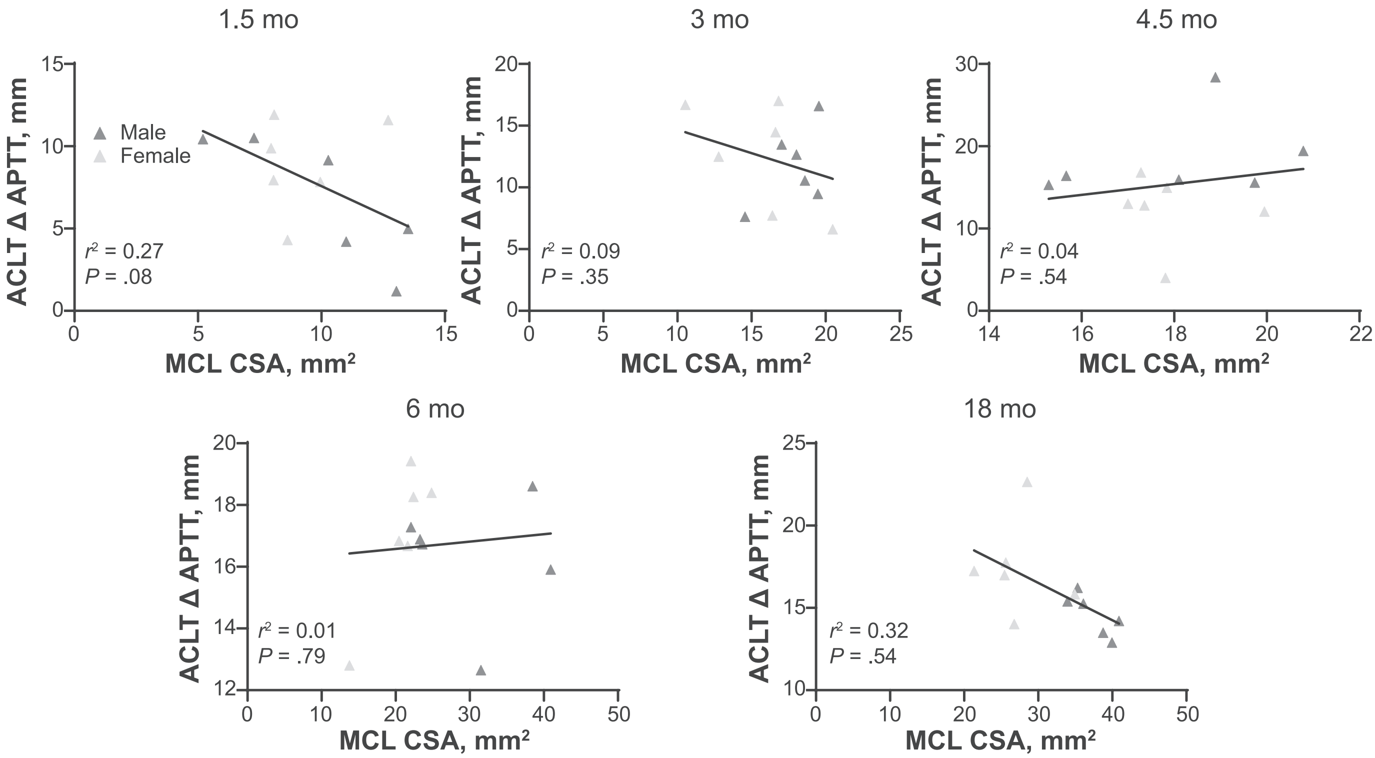

Figure 10. Change in anterior-posterior tibial translation (APTT) after anterior cruciate ligament transection (ACLT) versus medial collateral ligament (MCL) cross-sectional area (CSA), plotted by age group. Statistical results from linear regression shown in graph corner.

during skeletal growth, joint laxity seemed to be driven primarily by factors other than tissue size, such as tissue material properties.

Even though limited skeletally immature human data were available, findings from the older age groups in this research were generally consistent with prior results[4,17] of increased joint laxity after ACL and AM bundle injury in skeletally mature humans. Both an in vivo study[4] and an ex vivo cadaveric study[17] in skeletally mature humans demonstrated large (6-mm) differences in anterior tibial translation in ACL-injured knee joints versus healthy joints under a 134-N applied anterior load. Consistent with our research, AM bundle rupture resulted in much smaller (2.5-mm) differences in APTT.[4] The magnitude of the increases in APTT after ACL (14–18 mm) and AM bundle (3–5 mm) transection was somewhat greater in the late-adolescent age group in the present investigation. This was consistent with another study[26] describing 3-fold increases in APTT after ACL injury in skeletally mature porcine knees, indicating that porcine ACL injury led to greater joint destabilization than ACL injury in human knees. Furthermore, the ex vivo human cadaveric research revealed increases of approximately 150% in the force contributions of both the MCL and LCL under anterior drawer testing after ACL transection.[17] This varied from our work, in which the forces were increased only on the medial side of ACL-T joints. This difference was possibly due to varied testing protocols because we locked internal rotation to avoid excessive kinematic rotation inherent to porcine stifle-joint testing, as opposed to the human cadaveric study, which displayed a mean of 7° of internal tibial rotation under applied anterior drawer testing in ACL-deficient joints.[17] This internal rotation may have allowed the forces to be distributed more evenly between the medial and lateral compartments of the human knee.

Specific data comparing long-term degenerative changes, such as rates of osteoarthritis or subsequent injuries to secondary tissues, after partial ACL injury in humans are limited, especially in the pediatric population. Despite relatively small increases in joint laxity due to partial injury, the authors[16] of an in vivo study in a skeletally mature ovine model found significant gross and histological damage to cartilage 40 weeks after AM bundle injury compared with control specimens. However, given that our changes in laxity were smaller for younger animals, it is possible that skeletally immature patients with partial ACL injuries may experience lower rates of osteoarthritis and cartilage damage than skeletally mature patients. Moreover, the PL bundle supported the force typically carried by the intact ACL in AM-T joints, which may mean that secondary tissues are at lower risk of subsequent injury in patients with partial ACL tears compared with complete tears. In order to determine when nonoperative treatment of partial ACL tears is appropriate, more long-term in vivo studies in skeletally immature participants are necessary to evaluate if the smaller instabilities due to partial ACL injury lead to long-term degenerative changes.

Our findings have implications for the choice of operative versus nonoperative treatment of partial ACL injuries in young patients. Currently, few guidelines address when surgical intervention is needed for partial ACL injuries in skeletally immature patients. Surgical reconstruction is generally advised for pediatric patients with side-side differences >4 mm or an asymmetric pivot shift test.[3] Researchers[12] of skeletally immature patients additionally recommended reconstruction in older (>14 years) athletes or if more than 50% of the CSA was torn due to higher rates of progression to complete tear. The higher injury rates observed in mid- to late-adolescent patients seem consistent with this investigation, which showed larger instabilities after AM bundle transection in older participants. In a separate study[14] of adolescents and adults, age <20 years was a risk factor for partial injury to progress to a complete tear, perhaps indicating that surgical reconstruction is desirable in active adolescents and young adults with partial ACL tears. We found the greatest instabilities due to partial ACL injury in late adolescent female pigs. Although earlier authors did not find sex to be a significant factor for progression to a complete ACL tear,[12,14] these investigators compared sexes across all ages rather than within small age groups. Based on the porcine model, our results suggest that late adolescent females, in particular, may be more likely to require surgical intervention after partial ACL injury; however, these findings should be confirmed in humans.

Rehabilitation after ACL injury is perhaps as important as the decision regarding surgical intervention, but current protocols for pediatric patients are primarily extrapolated from research in adults and no consensus in protocol guidelines has been achieved.[27,28] Rehabilitation after both operative and nonoperative management of ACL injuries typically involves bracing and restricted (with gradually increasing) range of motion and weight bearing, followed by some form of strength training and proprioceptive exercises.[28] Yet specific guidelines regarding progression and return to sport vary among studies. As summarized by Yellin et al,[28] some protocols are based on timing from injury (with variations in suggested timing), whereas others are based on physical therapy milestones. Furthermore, specific protocols for the conservative treatment of partial ACL tears are sparse. Few authors have compared outcomes between rehabilitation protocols in pediatric patients, but a recent study[29] determined that more conservative rehabilitation resulted in lower reinjury rates in pediatric athletes after ACL reconstruction. Additionally, injury-prevention programs involving neuromuscular training have shown success in lowering the incidence of knee injury in pediatric athletes.[30] Similar programs would likely protect partial ACL tears from progressing to complete tears considering that the remaining bundle of the ACL functioned similarly to the intact ACL immediately after injury in our work. We also noted that more force was placed on the MCL and medial meniscus in ACL-deficient joints, suggesting that rehabilitation strategies for complete ACL-T knees may need to focus on protecting the medial compartment of the knee. To establish clear guidelines and improve patient outcomes, more evaluation of rehabilitation protocols after ACL injury in pediatric athletes is needed.

Limitations

This study had several limitations. Primarily, because this research used an animal model to study a human condition, further work is needed to confirm the current findings in humans. In addition, the joint-loading conditions represented clinical examinations rather than in vivo conditions. Although anterior drawer examinations are clinically relevant for measuring joint laxity, they may not capture the most common in vivo loads experienced by tissues in the joint. For example, conditions involving compression of the knee joint, such as any weight-bearing activities, result in more engagement of the menisci.[31] Also, in vivo loading of the knee joint involves muscle contractions to stabilize or move the joint; we only measured forces of passive structures in the knee. On a related note, we did not assess contributions of the joint capsule to anterior drawer restraint. Even though the joint capsule contributed minimally to forces in the intact knee (see Supplemental Table S2), it has been found to resist 30% to 35% of the anterior load in ACL-deficient knee joints.[32] However, it was necessary to disrupt the joint capsule to access and transect the ACL in the present study. Loads applied to the knee joint in this research were also scaled to bone size as a surrogate for body weight. Yet loads are not typically scaled when performing joint laxity tests in humans. Consequently, this may have affected our findings of greater change in laxity due to partial injury throughout skeletal growth. Both peak knee loads during athletic tasks[33,34] and ACL material properties such as elastic modulus and ultimate strength[35] increase throughout skeletal growth. Therefore, scaled loads may more closely reflect the strains placed on the ACL in vivo. In addition, partial ACL-injury findings in this investigation were limited to AM bundle injury. Another limitation involved the limited power for sex comparisons within age groups, particularly for tissue force contributions where age $\times$ sex interaction terms were not statistically significant. The effect sizes of these factors were not known before the study, which would have allowed us to perform power calculations. Still, reasonably large effect sizes between sexes were observed in some cases (eg, effect sizes were 1.9–2 for MCL and medial meniscus contributions during late adolescence), and it seems likely that these differences within age groups would be significant with additional specimens. Finally, this was a cross-sectional study and did not capture longitudinal changes after ACL or partial ACL injury, such as tissue remodeling, which may influence long-term joint function after injury.

CONCLUSIONS

Function of the ACL, specifically the relative force distribution between its AM and PL bundles, shifted naturally during adolescence in both male and female pigs. Changes in laxity due to AM bundle injury were greater in late adolescent than in early adolescent or juvenile specimens. Furthermore, these changes in laxity were greater in female compared with male pigs during late adolescence. In the case of partial ACL injury, the remaining bundle appeared to support the primary role of the intact ACL. However, with a complete ACL injury, loads placed on the MCL and medial meniscus increased. Also, the loads placed on the MCL were higher in males, whereas loads placed on the medial meniscus were higher in females. More studies are needed to confirm that these findings translate to humans.

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SUPPLEMENTAL MATERIAL

Supplemental Tables S1–S9 and Figures S1–S3.

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