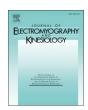
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Running-related muscle activation patterns and tibial acceleration across puberty



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ABSTRACT

This study examined whether differences exist in tibial acceleration transients and electromyography (EMG) variables during running across female pubertal development. Sixty-four girls classified as pre- (n=19), early/mid- (n=22) and late/post-pubertal development (n=23) ran in a laboratory whilst EMG data were recorded from quadriceps, hamstring and calf muscle groups, and acceleration transients from a triaxial accelerometer. The late/post-pubertal girls exhibited delayed vastus lateralis onset (mean difference (MD) = 0.02, 95% CI = 0.008, 0.34 ms)) compared to pre-pubertal girls, lower vastus lateralis pre-activation (MD = 7.02, 95% CI = 12.63, 1.42%) compared to early/mid-pubertal girls, and longer time to peak (TTP) anterior/posterior (A/P) tibial acceleration compared to pre-pubertal girls (MD = 0.02, 95% CI = 0.006, 0.03 s). By contrast, late/post-pubertal girls demonstrated earlier semitendinosus onset compared to both early/mid- (MD = 0.02, 95% CI = 0.03, 0.005 ms) and pre-pubertal girls (MD = 0.02, 95% CI = 0.04, 0.007 ms). No other between-group differences were found for peak A/P, vertical and TTP vertical tibial acceleration (p>0.05). Subsequently, regression analysis revealed that EMG variables accounted for approximately 34% $(R^2=0.34)$ of the variance in TTP A/P tibial acceleration. These findings highlight that neuromuscular recruitment patterns and kinetics differ across female pubertal development while running and should be further explored in the context of adolescent female musculoskeletal injuries.

1. Introduction

Growth of the musculoskeletal system during puberty leads to adaptations in long bone, cartilage and muscle that do not occur simultaneously, and may contribute to a higher risk of musculoskeletal injury during adolescence (Faust, 1977, Tanner, 1986, Michaud et al., 2001). In fact, pubertal musculoskeletal injuries increase by approximately 10% from pre-puberty to post-puberty (Michaud et al., 2001), with overuse injuries reported to affect females more than males (Stracciolini et al., 2014). Although a causal relationship between pubertal development and overuse musculoskeletal injuries is not well established, the aforementioned studies highlight that growth-related changes, particularly during female pubertal development, may be a contributing factor.

Recently, emerging evidence has highlighted that neuromuscular and biomechanical changes begin to occur during female pubertal development (Barber-Westin et al., 2006, Myer et al., 2010, Wild et al., 2012, 2013B). Despite the variation in tasks evaluated across studies (i.e., running versus landing), higher external hip and knee joint

moments (Wild et al., 2016, Sayer et al., 2018), decreased hamstring muscle strength (Wild et al., 2013A, 2013B) and delayed onset of vastus medialis activation (Wild et al., 2016) have been observed in girls at later compared to earlier stages of puberty. While these findings indicate that female pubertal development may be associated with potentially suboptimal changes in neuromuscular function, there remains a lack of substantive evidence to support these claims. Hence, further research investigating neuromuscular performance across pubertal development stages during dynamic tasks is required.

Tibial 'shock' and lower limb muscle activation patterns across female pubertal stages are two areas requiring further investigation. 'Shock' is a biomechanical term used to describe the magnitude and rate of lower extremity loading during the contact phase of dynamic tasks (Lafortune et al., 1996, Coventry et al., 2006). For example, during the early stance phase of running, the tibia undergoes a rapid 'shock' (typically measured by accelerometers mounted on the proximal tibia) that must be absorbed by both passive structures (i.e., soft tissues, cartilage, synovial fluid and bone) and active muscles (Mizrahi and Susak 1982, Lafortune et al., 1996, Coventry et al., 2006). Importantly,

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controlled muscular contraction is the most effective way of dissipating potentially harmful tibial acceleration transients moving up the kinetic chain (Mizrahi and Susak, 1982). Support for this notion is provided from novel neuromusculoskeletal modelling research by Saxby and colleagues (Saxby et al., 2016a,b), in which the surrounding lower limb musculature was shown to contribute to 80% of tibiofemoral joint loads while running, with the remaining 20% due to external forces. Hence, it stands to reason that muscle activation patterns measured via electromyography (EMG) may play an important role in regulating the magnitude and timing of tibial 'shock' during running.

No previous studies have examined tibial 'shock' and associated muscle activation patterns during running across the stages of female pubertal development, However, Dufek and colleagues (2008) reported that running-related impact attenuation measured using a uniaxial accelerometer was approximately 40% lower in pubertal compared to pre-pubertal girls (Dufek et al., 2008). Whilst these findings provide some evidence that puberty may influence tibial acceleration while running, this study only measured a single, undefined plane and failed to implement appropriate pubertal classification methods (Tanner et al., 1976, Tanner, 1986). Other studies have investigated the influence of female development on external knee joint moments during running and landing, and have reported higher joint moments at latter stages of puberty (Wild et al., 2016, Sayer et al., 2018). For instance, we recently reported that girls at late/post-pubertal development run with approximately 12-18% higher external knee extension moments than their pubertal and pre-pubertal counterparts (Sayer et al., 2018). Moreover, other research has found that girls increase their landingrelated vertical ground reaction force (GRF) as they progress through puberty (Swartz et al., 2005, Hewett et al., 2006, Quatman et al., 2006). Given that vertical and sagittal plane GRFs are major contributors to joint moments that may contribute to knee injury (Pairot-de-Fontenay et al., 2019), and the magnitude and timing of tibial acceleration transients are closely associated with the peak GRF's (Elvin et al., 2007, Crowell et al., 2010), it is plausible that pubertal differences in runningrelated tibial 'shock' may also exist.

To date, only one study has investigated neuromuscular recruitment patterns across the stages of female pubertal development. Wild et al. (2016) reported delayed onset of vastus medialis, but no difference in rectus femoris, hamstrings, medial gastrocnemius and tibialis anterior muscles during single limb landing as girls progressed through pubertal development. Although these findings may suggest muscle activation patterns do not change across female pubertal development, this study did not investigate distinct pubertal groups (i.e., pre, mid and late/postpubertal stages), and used a landing task rather than running. Therefore, a more comprehensive analysis of running-related muscle recruitment strategies is needed to determine if differences exist between pubertal groups and, given the link between muscle recruitment and dissipation of tibial acceleration transients (Mizrahi and Susak 1982, Lafortune et al., 1996, Coventry et al., 2006), to establish if muscle activation patterns contribute to any differences in the timing and magnitude of peak tibial acceleration.

The primary aim of this study was to examine whether girls classified as pre-pubertal, early/mid-pubertal and late/post-pubertal exhibit differences in lower limb muscle activation patterns (i.e., onset and amplitude) and the timing and magnitude of peak anterior and vertical tibial acceleration transients during barefoot running. The secondary aim was to explore relationships between measures of muscle activation and tibial acceleration adjusted for pubertal development stage. Our primary hypothesis was that girls in the late/post-pubertal group would exhibit both increased anterior and vertical peak tibial acceleration, as well as delayed onset and lower amplitude of quadriceps, hamstrings and gastrocnemius muscles compared to early/mid-and pre-pubertal girls. Our secondary hypothesis was that an inverse relationship would exist between the onset and amplitude of lower limb muscle activation and the magnitude and time to peak (TTP) of anterior and vertical peak tibial acceleration as pubertal stage advances.

2. Methods

2.1. Study population

Sixty-four recreationally active females were recruited from the student body at the University of Melbourne, local schools, community centres and sporting facilities. Inclusion criteria were: (i) aged 7–25 years old; (ii) participating in regular physical activity (> 30 min of moderate and/or vigorous activities daily); and, (iii) healthy weight (body mass index < 30 kg/m²). Exclusion criteria were: (i) history of lower limb injury, knee pain or medical condition affecting walking, running and jumping (ii) previous anterior cruciate ligament or meniscal injury or (iii) current bi-or tri-phasic oral contraceptive pill (OCP) use. All participants, together with parents/guardians of those < 18 years of age, signed an informed consent form. This study received ethics approval from the University of Melbourne Human Research Ethics Committee (#1442604).

All participants were categorised into one of three modified phases of puberty: (i) pre-pubertal (Tanner stage I), (ii) early/mid-pubertal (Tanner stage II-III and either growth spurt or menarche) and, (iii) late/post-pubertal (Tanner stage IV-V, both menarche and growth spurt essential) stages. Tanner staging was based upon self- or parental-rated breast development via an online, de-identified questionnaire containing pictures and modified diagrams (Tanner et al., 1976, Tanner 1986). Specific information about the pubertal classification process can be found in our previously published study (Sayer et al., 2018).

2.2. Physical activity

Physical activity data were collected using a modified version of the Children's Leisure Activity Study Survey (CLASS) (Telford et al., 2004, Huang et al., 2009). This was to ensure that i) participants met the inclusion criteria of being physically active, and ii) pubertal groups were participating in comparable levels of physical activity The CLASS classifies intensity levels of sport and physical activity participation throughout a typical week as "moderate" and "vigorous" using the metabolic equivalent table (Telford et al., 2004). The reliability (ICC = 0.71) and validity (r = 0.48) of the CLASS have been established (Telford et al., 2004, Huang et al., 2009).

2.3. Hormonal consideration

As higher estrogen levels have the capacity to influence lower limb biomechanics (Wild et al., 2012), all girls were tested when estradiol levels were low. The present study included participants from a larger cohort study we previously conducted and provides detailed information on this process for eumenorrheic and OCP users (Sayer et al., 2018). Confirmation that all participants were tested when estradiol levels were low (including the pre-pubertal group) was obtained via a 5 mL saliva estradiol sample taken at the time of testing and later analysed at the manufacturer's laboratory (Nutripath Integrative Pathology, Melbourne, Australia). For analysis, saliva samples were stored at $-20\,^{\circ}\mathrm{C}$ and analysed via enzyme immunoassay according to the manufacturer's instructions. All participants were required to have $<18\,\mathrm{pmol/L}$ according to the reference ranges provided by the manufacturer.

2.4. Procedure

Following saliva collection, the dominant limb was selected for analysis via the footedness subscale of the Lateral Preference Inventory (LPI) (Coren, 1993). Participants then had their skin prepared for the placement of bipolar Ag-Al surface electrodes (Norotrode 20, Myotronics, USA) by cleaning the skin with alcohol-based wipes. Eight electrodes were placed in line with the muscle fibres of the bellies of the rectus femoris (RF), vastus medialis (VM), vastus lateralis (VL), biceps



Fig. 1. Tibial acceleration position. A triaxial Noraxon DTS 3D wireless accelerometer (24 G) was attached to a steel bracket and fixed approximately 1 cm distal to the tibial tuberosity of the test limb prior to the running task.

femoris (BF), semitendinosus (ST), medial (MG) and lateral gastrocnemius (LG) muscles, with Noraxon DTS 2400 wireless telemetry sensors attached to each (Cram et al., 1998). Circular surface electrodes of 10 mm sensor diameter were placed at an inter-electrode distance of 20 mm on the aforementioned muscles. These were attached to the corresponding Noraxon wireless recording devices that were taped to the skin over the closest region of bony prominence to create grounding conditions for each device. Before fixing the accelerometer to the participant's leg, the primary investigator calibrated the device for each participant and axis using an average rest signal of 1 s. Following this, a Noraxon DTS 3D wireless accelerometer (24 G) attached to a steel bracket with an adjustable belt was fixed approximately 1 cm distal to the tibial tuberosity (Fig. 1). The running task was then explained to participants using a standardized set of instructions that emphasized the importance of completing each trial at a comfortable pace using their natural running style. Before data collection, as all girls were habitually shod runners, participants were allowed approximately five minutes to familiarize themselves with running barefoot in the laboratory. We chose to test participants barefoot as our primary aim was to investigate differences in muscle activation and acceleration patterns between pubertal stages, and different footwear worn by participants within and across the pubertal stages would have potentially confounded differences. In order to ensure any differences we observed between groups could be attributed to pubertal maturation, and not simply due to differences in footwear worn (Hall et al., 2013, Sayer et al., 2019), we standardised test conditions across all participants by ensuring everyone was tested barefoot.

Three successful running trials were recorded, in which a trial was considered successful if participants: (i) completed a clean strike of a concealed force place (AMTI, Inc., Watertown, MA, USA), and (ii) ran at 2.8–3.2 m/s as measured via photoelectric timing gates on either side of the force plate. Running speed was controlled because variations between participants can affect joint biomechanics and impact forces (Winter, 2009). In the event the participant either ran too quickly or

slowly, they were advised to modify their speed accordingly until the desired speed was attained.

2.5. Data analysis

The EMG signals were pre-amplified 500 times and low-pass filtered (common mode rejection ratio > 100 dB; input impedance > 100 Mohm). Signals were then wirelessly transmitted (fixed manufacturer delay of 312 ms) then output via a Noraxon DTS analog module and digitized with 16-bit resolution at 2400 Hz using a Vicon analog-to-digital converter. The GRF > 20 N (Vicon, Nexus) was used to detect initial contact and the wireless delay, accelerometer and EMG signals were corrected to align with this time point. Data analysis was performed in Matlab R2015b using custom programs (Mathworks).

For EMG onset, the Teager-Kaiser Conditioning method was used on the band-passed signal (Solnik et al., 2010). The EMG data were rectified and low pass filtered with a 50 Hz zero-lag Butterworth filter. Muscle activation onset was defined as the point when the EMG signal exceeded a threshold of 15 standard deviations above baseline signal for 25 ms (Solnik et al., 2010). For the amplitude analysis, the EMG signal was rectified and low-pass filtered with a 20 Hz Butterworth filter (Ford et al., 2011). The amplitude before initial contact was defined as the mean amplitude from -100 ms to initial contact and the during early stance phase as the mean amplitude from initial contact to +100 ms (Fagenbaum and Darling, 2003). All amplitude variables were normalized to peak EMG during the stance phase of each trial and represented as a percentage (Ford et al., 2011).

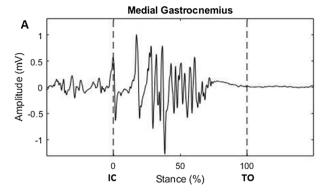
The acceleration data were processed as follows. Signals were filtered with a 60 Hz low pass zero-lag 4th order Butterworth filter. Similar to a previous study (Lafortune and Hennig, 1991), we conducted a power spectrum analysis of the tibial acceleration signal to determine the filter cut-off frequency. Consistent with Lafortune and Hennig (1991), our analysis revealed the majority of the signal was below 60 Hz, hence this was chosen as the filter cut-off frequency. From the filtered acceleration-time curves, the following acceleration variables were calculated: (1) magnitude (gravities; g) of peak anterior and vertical tibial acceleration, and (2) TTP (seconds) from initial contact to peak anterior and vertical tibial acceleration. An example of the EMG and acceleration signals is illustrated in Fig. 2.

2.6. Statistical analysis

Means and standard deviations (SD) were calculated for all participant characteristics, EMG onset, amplitudes and acceleration variables. A one-way analysis of variance (ANOVA) was used to test for between-group differences. In the event of a main effect, *post-hoc* analyses were performed using Fisher's Least Significant Difference tests, with the mean difference (MD) and 95% confidence intervals (CI) reported. In the event of between-group differences for EMG (onset and/or amplitude) and acceleration variables (peak and TTP), regression analysis (enter method) was used to explore the relationship between independent predictors (e.g. EMG variables) and the dependant variable of tibial acceleration adjusting for pubertal development group. All data were analysed using the Statistical Packages for Social Science (SPSS, version 23, IBM) and significance was set at 0.05.

3. Results

Demographic characteristics for the cohort, according to stage of puberty, are presented in Table 1. As expected, differences were found for age, mass and height (p < 0.05), whereby the late/post-pubertal girls were taller, heavier and older than their early/mid- and pre-pubertal counterparts (p < 0.05, Table 1). There were no differences in estradiol concentration between the three groups. (p > 0.05).



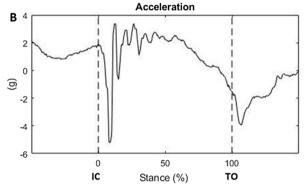


Fig. 2. Typical EMG and acceleration signal. (A) Represents a sample of the typical EMG signal from the medial gastrocnemius. The x-axis represents the percentage (%) stance phase with initial contact (IC) at 0 and toe off (TO) at 100% of stance. The y-axis represents the amplitude of the EMG signal in millivolts (mV). (B) Represents the typical vertical acceleration curve (positive values) which took place in early stance with the y-axis representing acceleration in gravitational constants (g) and the x-axis the percentage of stance.

Table 1 Participant characteristics according to pubertal development. All variables are reported as mean $\,\pm\,$ SD.

Variable	Pre-pubertal	Early/mid-pubertal	Late/post-pubertal
n A co (weeks)	19 9.4 + 1.2	22 11.1 + 1.4 [†]	23 19.8 ± 4.0 ^{†,‡}
Age (years) Weight (kg)	9.4 ± 1.2 31.2 ± 5.2	$37.2 \pm 5.5^{\dagger}$	$60.4 \pm 9.9^{\dagger,*}$
Height (m)	1.4 ± 0.1	$1.5 \pm 0.1^{\dagger}$	$1.7 \pm 0.1^{\dagger,*}$
Estradiol (pmol/L)	8 ± 5.9	7.5 ± 4.3	10.8 ± 5
Moderate physical activity (mins)	524.5 ± 327.9	380.0 ± 193.3	537.2 ± 294.4
Vigorous physical activity (mins)	245.5 ± 174.0	302.3 ± 317.6	291.3 ± 317.6
Total physical activity (mins)	770.0 ± 373.5	682.3 ± 552.1	828.5 ± 552.1

 $^{^{\}dagger}$ Denotes significantly different to pre-pubertal group (p < 0.05).

3.1. Muscle onset timing

A main group effect was found for VL, VM and ST (p < 0.05, Table 2). Post-hoc analysis for VL revealed that the late/post-pubertal girls had delayed onset (i.e., closer to initial contact) compared to prepubertal girls (MD = 0.02, 95% CI = 0.008, 0.34 ms, p = 0.002). Likewise, post-hoc results for VM also showed that the late/post-pubertal girls had delayed onset (MD = 0.02, 95% CI = 0.36, 0.005 ms, p = 0.012) compared to pre-pubertal girls (Table 2). In contrast, there was evidence that ST onset was earlier (i.e. further before initial contact) in the late/post-pubertal group compared to both early/mid-(MD = 0.02, 95% CI = 0.03, 0.005 ms, p = 0.009) and pre-pubertal girls (MD = 0.02, 95% CI = 0.04, 0.007 ms, p = 0.005).

Table 2 EMG onset according to pubertal development. All variables are reported as mean \pm SD in milliseconds measured before initial contact.

Variable	Pre-pubertal	Early/mid pubertal	Late/post-pubertal
Rectus femoris (ms) Vastus lateralis (ms)	0.03 ± 0.01 0.07 + 0.02	0.03 ± 0.02 0.06 + 0.03	0.03 ± 0.02 $0.05 + 0.02^{\dagger}$
Vastus medialis (ms)	0.07 ± 0.02 0.08 ± 0.02	0.06 ± 0.03 0.06 ± 0.02	0.06 ± 0.02
Lateral gastrocnemius (ms)	0.05 ± 0.03	$0.08 \pm 0.04^{\dagger}$	0.07 ± 0.03
Medial gastrocnemius (ms)	0.10 ± 0.04	0.12 ± 0.04	0.09 ± 0.05
Biceps femoris (ms) Semitendinosus (ms)	0.16 ± 0.01 0.16 + 0.03	0.17 ± 0.03 0.17 ± 0.02	0.18 ± 0.03 $0.19 \pm 0.02^{\uparrow,\ddagger}$

ms-milliseconds.

- † Denotes significantly different to pre-pubertal group (p < 0.05).
- * Denotes significantly different to early/mid-pubertal group (p < 0.05).

3.2. Muscle amplitude before initial contact

A main group effect was found only for amplitude of VL before initial contact (p=0.04) and LG (p=0.008, Table 3). Specifically, the *post-hoc* results showed the late/post-pubertal group had lower amplitude of VL compared to early/mid-pubertal girls (MD = 7.02, 95% CI = 12.63, 1.42%, p=0.015). In contrast, higher activation of LG was evident in the late/post- (MD = 7.35, 95% CI = 2.42, 12.28%, p=0.004) and early/mid pubertal groups (MD = 6.48, 95% CI = 1.55, 11.42%, p=0.01) compared to pre-pubertal girls.

3.3. Muscle amplitude during early stance phase

Main group effects were found for BF (p < 0.001) and VM (p = 0.03, Table 4). The late/post-pubertal girls had a lower BF amplitude during early stance compared to early/mid- (MD = 11.57, 95% CI = 17.33, 5.81%, p < 0.001) and pre-pubertal groups (MD = 11.57, 95% CI = 17.56, 5.57%, p > 0.001). For VM, the early/mid-pubertal group had a higher amplitude compared to pre-pubertal group (MD = 7.84, 95% CI = 2.01, 13.67%, p = 0.009).

3.4. Tibial acceleration during stance phase

There were no between-group differences found for peak A/P, peak vertical and TTP vertical acceleration (p>0.05, Table 5). However, a main group effect was found for TTP A/P acceleration, whereby the TTP was longer for late/post-pubertal compared to pre-pubertal girls (MD = 0.02, 95% CI = 0.006, 0.03 s, p=0.003).

Multiple linear regression analysis revealed a significant association ($F_{(8,41)}=2.66,\ p=0.02$) between TTP A/P acceleration, EMG variables (VL, VM and ST onset; BF and VM early stance amplitude; VL and LG amplitude prior to initial contact) and pubertal stage. Approximately 34% of the variance in TTP A/P acceleration was explained by the independent variables ($R^2=0.34$). However, the only significant predictors were pubertal stage (B = 0.01, 95% CI = 0.01, 0.02, p=0.04) and ST onset (B = 0.18, 95% CI = 0.11, 0.35, p=0.04), in which latter stages of development and earlier activation of ST were associated with a longer TTP A/P acceleration.

4. Discussion

Female pubertal development is associated with neuromuscular and biomechanical changes (Barber-Westin et al., 2006, Myer et al., 2010, Wild et al., 2012, 2013B) that are postulated to contribute to the higher incidence of musculoskeletal injury amongst pubescent girls and young women (Michaud et al., 2001). Given the lack of studies reporting the association between pubertal development, muscle activation and tibial acceleration during running, the present study provides further

[‡] Denotes significantly different to early/mid-pubertal group (p < 0.05).

Table 3

EMG amplitude prior to initial contact according to pubertal development. All variables are reported as mean ± SD as a percentage of the peak signal during the stance phase.

Variable	Pre-pubertal	Early/mid pubertal	Late/post-pubertal
Rectus femoris (%)	14.30 ± 5.47	17.44 ± 10.65	12.28 ± 4.05
Vastus lateralis (%)	14.07 ± 6.22	16.81 ± 13.83	$9.78 \pm 5.16^{\circ}$
Vastus medialis (%)	15.09 ± 6.71	15.68 ± 6.24	12.07 ± 7.28
Lateral gastrocnemius (%)	10.33 ± 4.98	$16.82 \pm 7.44^{\dagger}$	17.69 ± 10.01
Medial gastrocnemius (%)	21.15 ± 11.97	27.56 ± 15.29	23.99 ± 11.06
Biceps femoris (%)	39.69 ± 13.97	42.03 ± 19.44	42.12 ± 27.77
Semitendinosus (%)	52.73 ± 31.41	58.97 ± 45.51	62.32 ± 58.72

 $^{^{\}dagger}$ Denotes significantly different to pre-pubertal group (p < 0.05).

evidence of neuromuscular-biomechanical changes across stages of pubertal development. Specifically, girls at latter stages of puberty activated their VL and VM closer to initial contact, while ST onset was further from initial contact (i.e., ST activated earlier than the aforementioned muscles). For muscle amplitude, latter stages of puberty evoked smaller amplitudes of VL activation but higher LG amplitudes before initial contact and a smaller amplitude of BF following initial contact, partly supporting our primary hypothesis. Regarding tibial acceleration, a longer TTP A/P tibial acceleration was found at latter stages of puberty which was not consistent with our primary hypothesis and an earlier ST onset before initial contact at latter stages of puberty was associated with a longer TTP A/P acceleration from the regression model.

Our findings of delayed VM and VL onset, in combination with lower VL pre-activation amplitude, extend previous research and support the contention that altered neuromuscular strategies emerge during puberty (Wild et al., 2016). Whilst the cross-sectional design of this study limits the capacity to infer causation and the nature of barefoot running may have contributed to these novel findings (Hall et al., 2013), it is tempting to hypothesise that delayed onset and lower amplitude prior to initial contact may contribute to higher joint loads previously linked with adolescent knee joint injuries (Brown et al., 2014, Myer et al., 2014). The relationship between muscle activation patterns and joint moments in females during single-limb landing is supported by Brown et al. (2014) who found that greater RF pre-activation significantly predicted lower knee flexion moments - a relevant biomechanical variable in the context of adolescent patellofemoral pain (Lankhorst et al., 2013). Moreover, MacLean and colleagues (McLean et al., 2010) reported a strong relationship (r = 0.61-0.80) between higher external knee abduction moments and delayed onset of VL, VM, gastrocnemius and medial hamstrings in healthy adolescent females during a single-limb choice reaction task (i.e., participants jump either left or right in the direction of a random light stimulus). While the present study extends our understanding of neuromuscular recruitment patterns across female pubertal development, the aforementioned studies are both landing-based with differences in muscle onset definition and study design which make it difficult to draw conclusions. Hence,

our findings pertaining to delayed onset of VL and VM and a lower VL amplitude prior to initial contact may be clinically relevant in the context of patellofemoral pain (Lankhorst et al., 2013), but more importantly should be further evaluated using more sophisticated measures of joint and muscle forces via neuro-musculoskeletal modelling (Saxby et al., 2016a,b).

We found no between group differences for peak A/P or vertical acceleration, instead finding a longer TTP A/P acceleration across puberty. This is surprising given that we previously reported higher running-related external knee joint moments (i.e., knee flexion moment) at latter stages of puberty (Sayer et al., 2018), which we hypothesised would similarly result in higher tibial shock given the association between acceleration transients and GRFs (Elvin et al., 2007). While other studies in healthy individuals have investigated tibial acceleration during gait (Tirosh et al., 2017) and a single-limb landing task (Coventry et al., 2006), their findings are conflicting and unfortunately do not provide any further insight related to acceleration across female pubertal development. Hence, future prospective studies are needed to determine if pubertal differences remain unchanged or potentially emerge across time.

Finally, the regression analysis did not support our secondary hypothesis, as an opposite direction of change was found with earlier ST onset associated with a longer TTP A/P acceleration as pubertal stage advanced. These findings may be due to the fact that the girls were healthy and adequately adapted their neuromuscular system to increase knee stiffness and reduce 'shock' while running (Tam et al., 2017), thereby slowing the rate of tibial A/P acceleration. However, it should be acknowledged that the regression model only accounted for 34% of the variation in TTP A/P acceleration, which highlights that other unmeasured factors are contributing to a longer TTP at later stages of puberty. Unfortunately, we did not investigate muscle strength or rate of force development which are two factors that may also be associated with tibial A/P acceleration and need to be investigated in future prospective studies. Given that higher impact loading rates are associated with musculoskeletal injury (Davis et al., 2016), future pubertal studies characterising the relationship between neuromuscular activation, tibial shock and risk of injury are required.

Table 4

EMG amplitude during early stance according to pubertal development. All variables are reported as mean ± SD as a percentage of the peak signal during the stance phase.

Variable	Pre-pubertal	Early/mid pubertal	Late/post-pubertal
Rectus femoris (%)	45.92 ± 9.53	47.29 ± 7.81	49.47 ± 8.0
Vastus lateralis (%)	43.71 ± 8.07	46.21 ± 8.13	46.2 ± 7.95
Vastus medialis (%)	40.83 ± 7.01	$48.67 \pm 10.77^{\dagger}$	46.36 ± 8.31
Lateral gastrocnemius (%)	44.47 ± 11.40	49.02 ± 10.79	48.79 ± 9.74
Medial gastrocnemius (%)	50.08 ± 15.65	51.96 ± 13.73	41.91 ± 9.95
Biceps femoris (%)	45.92 ± 8.36	45.92 ± 9.20	$34.35 \pm 11.00^{\dagger, *}$
Semitendinosus (%)	42.86 ± 6.09	45.65 ± 9.74	41.91 ± 12.30

 $^{^{\}dagger}$ Denotes significantly different to pre-pubertal group (p < 0.05).

^{*} Denotes significantly different to early/mid-pubertal group (p < 0.05).

^{*} Denotes significantly different to early/mid-pubertal group (p < 0.05).

Table 5
Tibial acceleration according to pubertal development. All variables are reported as mean \pm SD.

Variable	Pre-pubertal	Early/mid pubertal	Late/post-pubertal
Peak A/P tibial acceleration (g) TTP A/P tibial acceleration (s) Peak vertical tibial acceleration (g) TTP vertical tibial acceleration (s)	5.02 ± 1.48 0.03 ± 0.01 8.60 ± 2.75 0.03 ± 0.01	4.41 ± 1.54 0.04 ± 0.02 9.36 ± 4.53 0.03 ± 0.01	$4.02 \pm 0.82 0.05 \pm 0.02^{\circ} 7.00 \pm 2.82 0.03 \pm 0.01$

A/P: anterior-posterior.

TTP: time to peak.

*Denotes significantly different to early/mid-pubertal group (p < 0.05).

While this study is the first to report pubertal tibial acceleration and EMG temporal variables during running, there are several limitations. Firstly, the cross-sectional study design does not allow conclusions regarding causation between our measured variables and pubertal development. Furthermore, the sample size for the regression analysis may be insufficient as previous research suggests that models with the number of predictors in our model (i.e., eight) should have a minimum of 110 participants (VanVoorhis C.R. 2007). In addition, the lack of peak isokinetic muscle force and rate of force development data in the model did not allow us to explore the association between muscular performance parameters and tibial acceleration transients. Finally, girls were habitually shod runners and tested barefoot, which may have influenced the resulting EMG and acceleration variables and limits the external validity of these findings as footwear would typically be worn for running. Moreover, barefoot running can induce changes in spatiotemporal variables, kinetics and kinematics through varied foot strike patterns which we did not measure and may explain some of the differences reported in this study. Hence, we suggest that future studies perform regression analysis with a larger sample size (i.e., > 110 participants) and include these additional measures, as this may provide valuable insight to mechanisms underpinning elevated TTP A/P tibial acceleration transients and muscle activation patterns at latter stages of puberty.

5. Conclusion

This study reports altered muscle activation patterns and a longer TTP A/P acceleration in girls at the latter stages of female pubertal development. Latter stages of pubertal development and earlier ST onset were predictors of a longer TTP A/P acceleration. It is not known if these differences in muscle activation patterns and TTP A/P acceleration have implications for musculoskeletal running injuries which future prospective studies should examine.

Declaration of Competing Interest

All authors declare no conflict of interest and at no stage did any of funding organizations influence study design, data collection or analysis of results.

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Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jelekin.2019.102381.

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