Tohoku J. Exp. Med., 2016, **240**, 287-294

# Increased Postural Sway and Changes in the Neuromuscular Activities of the Ankle Stabilizing Muscles at Ovulation in Healthy Young Women

**Haneul Lee[1] and JongEun Yim[2]**

[1]Department of Physical Therapy, Gachon University, Incheon, Republic of Korea
[2]Department of Physical Therapy, Sahmyook University, Seoul, Republic of Korea

Lateral ankle sprains are one of the most common injuries among the physically active subjects. Ankle inversion-eversion laxity is greater and dynamic postural control is less in women compared with men. The purpose of this study was to examine differences in postural sway and its effects on the neuromuscular activities of the ankle stabilizing muscles during the menstrual cycle in young women. Fourteen young women with regular menstrual cycles participated in this experiment. Postural sway and electromyographic signals of the lateral gastrocnemius, peroneus longus (PL), and tibialis anterior (TA) were recorded while the participants performed eight different balance tasks at ovulation and early follicular phase during one full menstrual cycle. Significantly greater postural sway in the two most difficult balance tasks was observed at ovulation compared to that in the early follicular phase ($p < 0.001$). A similar pattern was also observed in terms of PL activity, while TA activity was significantly greater in the most difficult balance task at ovulation. In addition, TA-PL co-contraction (TA/PL ratio) was significantly higher at ovulation compared with that in the follicular phase in the two most difficult balance tasks ($p < 0.01$). Young women could benefit from increased understanding of the varying neuromuscular activation patterns throughout the menstrual cycle. The results of this study suggest that health professionals should be aware of the physiological effects and the shifts in neuromuscular strategies in each menstrual cycle phase in order to prevent increased risk of lower extremity injury.

**Keywords:** ankle; estrogen; menstrual cycle; neuromuscular activation; postural sway
Tohoku J. Exp. Med., 2016 December, **240** (4), 287-294.

## Introduction

An increasing number of women incur knee injuries during sports activities (Beynnon et al. 2005; Eiling et al. 2007; Kjaer and Hansen 2008; Hansen et al. 2009b; Lee et al. 2013a, 2015), and the incidence, especially of non-contact anterior cruciate ligament (ACL) injuries, is two to eight times higher in women than in men who are performing the same activities (Deie et al. 2002; Liu and Luo 2005; Eiling et al. 2007; Hansen et al. 2009b; Park et al. 2009a, 2009b; Boden et al. 2010; Lee et al. 2013a; Stijak et al. 2015).

The menstrual cycle occurs due to fluctuations in sexual hormone levels and is composed of follicular, ovulatory, and luteal phases, each of which have markedly different hormonal profiles (Shultz et al. 2004; Lee et al. 2013a; Khowailed et al. 2015). Estrogen, one of the primary female sex hormones, peaks at ovulation and has been specifically studied regarding its relationship to human connective tissues (Liu et al. 1997; Heitz et al. 1999; Deie et al. 2002; Shultz et al. 2004; Liu and Luo 2005).

Human connective tissues such as tendons, muscles, and ligaments are composed of collagen fibers closely packed together (Park et al. 2009a). The closer the collagen fibers are packed together, the greater the mechanical strength, which then influences the strength of the muscles and ligaments (Liu et al. 1997; Park et al. 2009b).

Much research has revealed that 17-*β* estradiol receptors are present in human connective tissues (Yu et al. 2001; Shultz et al. 2005; Kjaer and Hansen 2008; Hansen et al. 2009b; Silbernagel et al. 2015) and that these receptors change the mechanical properties of tendons and ligaments (Heitz et al. 1999; Shultz et al. 2005; Lee et al. 2014). An obvious sex-related difference was observed in the role of estrogen in regulating muscle mass and ligament laxity (Lee et al. 2013b), and its relationship to ligament laxity has been investigated (Shultz et al. 2004; Hansen et al. 2009a; Lee et al. 2013a).

Several studies have explained that estrogen has an inhibitory effect on collagen synthesis, which alters connec-

---

Received August 25, 2016; revised and accepted November 15, 2016. doi: 10.1620/tjem.240.287.

Correspondence: Jongeun Yim, DSc., Department of Physical Therapy, Sahmyook University, 815 Hwarangro, Nowon-gu, Seoul 139-742, Republic of Korea.
e-mail: jeyim@syu.ac.kr

tive tissue metabolism (Miller et al. 2007; Silbernagel et al. 2015). Decreased collagen synthesis causes ligament laxity and weak muscle strength because of increased serum levels of estrogen and decreases in collagen formation and fibroblast proliferation (Park et al. 2009b; Lee et al. 2013a, 2014). Since estrogen level peaks during the ovulation phase, significant increases in knee laxity have been detected in the ovulation and mid-luteal phases compared with the early follicular phases (menses), when estrogen levels are lower (Deie et al. 2002; Shultz et al. 2005). Estrogen has also been shown to control protein synthesis in human connective tissue (Liu et al. 1996; Hansen et al. 2011).

The ankle plays a major role in posture and locomotion (Baumhauer et al. 1995; Vieira et al. 2013; Cattagni et al. 2014; Duclos et al. 2014; Pozzi et al. 2015), and small changes in the center of gravity are corrected by the ankle by shifting the center of gravity position on the basal plane (Page et al. 2010). Combined with the tibialis muscles, the peroneus muscles support and stabilize the ankle joint (De Ridder et al. 2014, 2015). The peroneus longus (PL) is well known to provide lateral stability, while the tibialis anterior (TA) muscle is increasingly activated in order to control the lateral displacement of the tibia (Page et al. 2010; De Ridder et al. 2014; Duclos et al. 2014; Pozzi et al. 2015). The TA/PL muscle ratio has been used to assess ankle unitability in a previous study (De Ridder et al. 2015). Calf muscles also support the anterior/posterior stability of the ankle joint, especially when the body is challenged (Vieira et al. 2013).

Center of pressure (CoP) displacement has been used to identify postural instability (Kalron and Achiron 2013), and it has a negative correlation with the maximal isometric torque of the ankle muscles (Cattagni et al. 2014), as weakness of the ankle-stabilizing muscles is a major cause of postural instability (Cattagni et al. 2014, 2016). Previous research has revealed that functional ankle instability is caused by a decreased ability to control postural sway when ankle invertor strength is decreased (Baumhauer et al. 1995). Similarly, Louwernes and colleagues showed that the PL plays a major role in maintaining balance, while the TA plays a supportive role (Louwerens et al. 1995). In both reports it was shown that TA activation increased in order to stabilize the ankle joint when the balance was improper (Baumhauer et al. 1995; Louwerens et al. 1995).

Many studies have investigated the effect of the menstrual cycle on ACL laxity and knee stabilizing muscles such as the quadriceps and hamstring (Heitz et al. 1999; Rozzi et al. 1999; Yu et al. 2001; Shultz et al. 2004, 2005; Pollard et al. 2006; Park et al. 2009b; Lee et al. 2013b, 2014; Khowailed et al. 2015; Stijak et al. 2015). However, only a few studies that provide evidence on ankle stability have been conducted (Hosea et al. 2000; Trimble et al. 2002; Petrofsky and Lee 2015; Silbernagel et al. 2015).

Therefore, we hypothesized that postural sway and the neuromuscular activities of the ankle stabilizing muscles would increase at ovulation in healthy young women. The aim of this study was to compare the postural sway and neuromuscular activities occurring at ovulation and in the early follicular phase.

## Methods

### *Participants*

Nineteen healthy young female volunteers with regular menstrual cycles (age, 20.8 ± 1.8 years; height, 161.3 ± 4.3 cm; weight, 57.2 ± 7.3 kg) participated in this study. All of the participants had to meet the following inclusion criteria: the presence of a regular menstrual cycle for more than one year at least; no history of pregnancy, cardiovascular disease, neuromuscular disorders, diabetes, vestibular diseases, and any lower extremity joint injuries; no use of any medication that would affect sex hormones; and nonsmoking behavior. The exclusion criteria were as follows: any lower limb injuries or surgeries, lower limb pain at the time of testing, and equilibrium disorders. All of the participants signed informed consent forms prior to beginning the study. This study was approved by the Gachon University Institutional Review Board (IRB).

### *Measurements*

*Self-reported menstrual cycle*: The participants were asked to report their menstrual cycles over the last three months. Based on their average menstrual cycle length (days), their expected ovulation date was calculated by the research coordinator. For the ovulation phase, the participants were instructed to start using a digital ovulation predictor kit (Clearblue, SPD GmbH, Geneva, Switzerland) (Ellis et al. 2011) with 99% accuracy from days 10 to 12, depending on their cycle length. When a positive result was detected, the participants were asked to immediately contact the research coordinator to schedule the data collection. For the follicular phase, the participants were instructed to contact the research coordinator once their period began, and were then scheduled for data collection between days 1 and 3 of their cycles.

*Electromyography*: Muscle activity was measured with a wireless surface Noraxon DTS system (Noraxon U.S.A. Inc., Arizona, USA) at a sampling rate of 1,000 Hz. Prior to EMG measurement, the skin was shaved and wiped with alcohol to reduce skin impedance. Disposable pre-gelled bipolar Ag/AgC1 surface electrodes of 11.4 mm diameter were then placed with an inter-electrode distance of 20 mm and positioned over the muscle belly parallel to the muscle fibers. The electrodes were placed to measure muscle activations in the lower extremities during the balance tasks, and were attached to the muscle valley of the lateral gastrocnemius (LG), PL, and TA on each participant's dominant leg (Fig. 1).

Surface EMG electrodes were secured with the bandwidth set between 20 and 350 Hz, and the notch filter set at 60 Hz. We normalized the signal of each muscle to the maximal voluntary contraction (MVC).

*Postural sway (Static postural control analysis)*: The displacement of the subject's COP was measured by using a valid and reliable force platform (Zebris FDM-S pressure platform, Zebris Medical GmbH, Germany) (Kalron and Achiron 2013). The force platform of 3.2 × 6.8 m in size consisted of 2,560 individually calibrated capacitive force sensors, each approximately 0.85 × 0.85 cm, underneath the platform at a sampling rate of 60 Hz. This enabled an analysis of the distribution of static forces beneath the feet during standing. The MR 3.8 software (Noraxon U.S.A.) was used to integrate the force signals and graphic representation of the CoP during balance tasks. Sway rate (mm/s), defined as the mean speed of the movement of the CoP during each balance task period, was used to express each individual's postural sway.

*Balance task*: Eight different static balance tasks on the force platform were used in the study. Three factors that are known to affect balance and the risk of falling, including vision, somatosensation (surface compliance), and the base of support, were altered both individually and in combination during the balance tasks. To alter the visual input, two levels of vision (eyes open and closed) were used. The Aeromat balance block (AGM Group, Aeromat Fitness Product, Fremont, CA, USA) was used to provide the participants with two different types of surface compliance (a firm and a foam surface). Two standing positions, either with feet apart (centers of the calcaneus in the same distance as the two anterior superior iliac spine), or in tandem (feet in a heel-toe position with the non-dominant foot in front), were used to test balance according to the base of support.

The eight different balance tasks were as follows: 1) feet apart, eyes opened, and firm surface (FAEO-FIRM); 2) feet apart, eyes closed, and firm surface (FAEC-FIRM); 3) feet apart, eyes opened, and foam surface (FAEO-FOAM); 4) tandem standing, eyes opened, and firm surface (TEO-FIRM); 5) feet apart, eyes closed, and foam surface (FAEC-FOAM); 6) tandem standing, eyes opened, and foam surface (TEO-FOAM); 7) tandem standing, eyes closed, and firm surface (TEC-FIRM); 8) tandem standing, eyes closed, and foam surface (TEC-FOAM). The participants were asked to perform the tasks in a random order for 10 seconds each, while two trained researchers stood nearby to prevent falls.

### *Procedure*

After obtaining informed consent, the investigator ascertained the basic characteristics of the participants, including age, height, weight, menstrual cycle length, self-reported cycle, and subjective dysmenorrhea.

All participants were asked to contact the investigator at two time points: on the day of ovulation and on the day menstruation was confirmed. Upon arrival at the laboratory, the participants rested comfortably in a temperature-regulated room for 20 minutes to stabilize their body conditions. Then, the investigator instructed the participants as to how to perform the balance tasks and gave them time to imagine how to perform them in order to become accustomed to the tasks. Next, electromyographic (EMG) signals and posture control variables were recorded while the subjects performed eight different balance tasks on a force platform.

### *Statistical analyses*

The GPower 3.1 software was used to calculate the sample size required so that a reasonable expectation would be likely to detect an expected effect size of 0.75 between the two different menstrual cycle phases, with an alpha error probability of 0.05 and a power of 0.80 (Khowailed et al. 2015). A sample size of 13 participants was required to provide a statistical power of 81%.

The SAS 3.9 software for Windows 10 was used to analyze the data. Data were summarized by using means and standard deviations (SD). The assumption of normality of the continuous variables was examined by using the Kolmogorov-Smirnov test. One-way repeated-measures analysis of variance was used to examine mean postural sway, EMG activity, and TA/PL ratio during eight different balance tasks in each phase of menstruation. The Bonferroni pairwise comparisons test for multiple comparisons was used to compare mean values of the variables between any two different balance tasks. A paired $t$ test was conducted to compare postural sway, EMG activity, and TA/PL ratio between the follicular phase and ovulation. The level of significance was set at $\alpha < 0.05$.

## Results

A total of 14 participants completed the study. One participant withdrew from the study because of a muscle spasm in the lower extremity from a car accident, and four other participants withdrew due to menstrual cycle conflicts. General participant characteristics are described in Table 1.

The differences in mean postural sway rate among the eight different balance tasks (F = 152.26, $p < 0.001$, $\eta^2 = 0.845$) and between the ovulation and follicular phase (F = 5.22, $p = 0.030$, $\eta^2 = 0.157$) during the menstrual cycle were significant. Concretely, no significant difference was found in the postural sway rate during the menstrual cycle for the six least difficult balance tasks ($p > 0.05$), but significant differences were found in TEC-FIRM and TEC-FOAM ($p = 0.044$ and 0.025, respectively; Fig. 1).

The LG showed significantly different muscle activity among the eight different balance tasks in both the follicular phase (F = 56.721, $p < 0.001$, $\eta^2 = 0.814$) and at ovulation (F = 31.575, $p < 0.001$, $\eta^2 = 0.708$). However, no significant difference in mean LG muscle activity was found between the follicular phase and ovulation for any of the balance tasks ($p > 0.05$, Table 2).

A significant difference in mean PL muscle activity was found among the different balance tasks in the follicular phase (F = 85.616, $p < 0.001$, $\eta^2 = 0.868$) and at ovulation (F = 64.338, $p < 0.001$, $\eta^2 = 0.832$). PL muscle activity was significantly increased in TEO-FIRM, TEO-FOAM, TEC-FIRM, and TEC-FOAM as compared to that in

Table 1. General Charaterisitcs in women with menstual cycle (N = 14).

| | Mean ± SD |
|---|---|
| Age (years) | 20.34 ± 1.39 |
| Height (cm) | 160.71 ± 4.86 |
| Weight (kg) | 56.57 ± 7.63 |
| Body mass index (kg/m$^2$) | 21.85 ± 2.33 |
| Cycle length (days) | 29.32 ± 3.19 |
| Subjective Dysmenorrhea | 3.2 ± 1.85 |

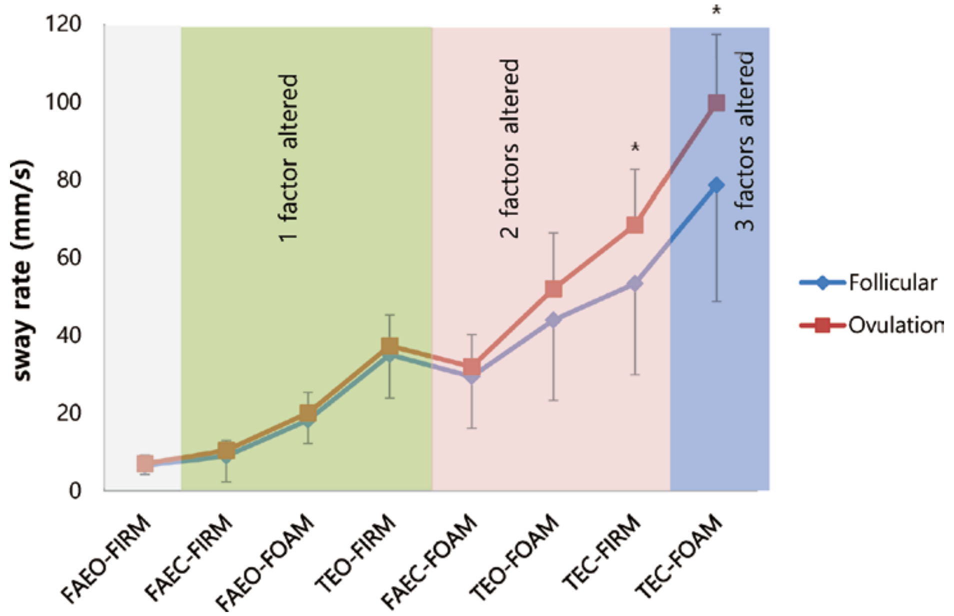

Fig. 1. Postural sway between follicular phase and ovulation.
The data are shown as mean ± SD of sway rate in different stations of balance platform between the follicular phase and ovulation (*significant difference).

Table 2. Neuromuscular activity on ankle stabilizing muscles in different stations of balance platform between the follicular phase and ovulation (N = 14).

| | Muscles | Follicular phase | Ovulation | *p*-value[a] |
|---|---|---|---|---|
| FAEO-FIRM | LGCM | 6.91 ± 2.42 | 6.56 ± 2.56 | 0.757 |
| | PL | 4.87 ± 2.04 | 4.64 ± 1.41 | 0.720 |
| | TA | 1.92 ± 0.98 | 1.85 ± 0.62 | 0.838 |
| | TA/PL ratio | 0.44 ± 0.21 | 0.44 ± 0.20 | 0.959 |
| FAEC-FIRM | LGCM | 7.47 ± 2.77 | 7.00 ± 3.66 | 0.613 |
| | PL | 5.66 ± 1.92 | 5.23 ± 1.51 | 0.529 |
| | TA | 2.48 ± 0.87 | 2.28 ± 0.67 | 0.442 |
| | TA/PL ratio | 0.47 ± 0.19 | 0.46 ± 0.15 | 0.842 |
| FAEO-FOAM | LGCM | 6.79 ± 3.01 | 6.54 ± 3.50 | 0.438 |
| | PL | 6.49 ± 2.90 | 4.95 ± 2.26 | 0.112 |
| | TA | 3.00 ± 1.15 | 2.66 ± 0.86 | 0.241 |
| | TA/PL ratio | 0.50 ± 0.17 | 0.57 ± 0.13 | 0.354 |
| TEO-FIRM | LGCM | 12.61 ± 3.05 | 13.44 ± 6.17 | 0.671 |
| | PL | 16.95 ± 4.94 | 17.26 ± 5.15 | 0.874 |
| | TA | 6.52 ± 3.58 | 9.15 ± 3.41 | 0.032 |
| | TA/PL ratio | 0.39 ± 0.18 | 0.52 ± 0.09 | 0.066 |
| FACE-FOAM | LGCM | 7.55 ± 2.89 | 7.03 ± 3.90 | 0.654 |
| | PL | 6.99 ± 2.58 | 6.47 ± 2.53 | 0.273 |
| | TA | 3.12 ± 1.17 | 3.30 ± 1.26 | 0.653 |
| | TA/PL ratio | 0.43 ± 0.16 | 0.51 ± 0.13 | 0.046 |
| TEO-FOAM | LGCM | 14.89 ± 3.42 | 14.04 ± 7.30 | 0.733 |
| | PL | 22.25 ± 8.04 | 21.74 ± 5.39 | 0.843 |
| | TA | 11.19 ± 5.74 | 14.58 ± 5.35 | 0.152 |
| | TA/PL ratio | 0.53 ± 0.24 | 0.68 ± 0.22 | 0.117 |
| TEC-FIRM | LGCM | 19.54 ± 7.01 | 16.90 ± 8.18 | 0.362 |
| | PL | 31.17 ± 10.66 | 23.48 ± 5.04 | 0.016 |
| | TA | 16.61 ± 9.70 | 19.56 ± 5.04 | 0.348 |
| | TA/PL ratio | 0.54 ± 0.21 | 0.86 ± 0.28 | 0.003 |
| TEC-FOAM | LGCM | 26.39 ± 6.45 | 22.64 ± 9.55 | 0.187 |
| | PL | 39.09 ± 10.51 | 30.35 ± 9.04 | 0.008 |
| | TA | 23.34 ± 7.51 | 30.15 ± 7.34 | 0.018 |
| | TA/PL ratio | 0.61 ± 0.17 | 1.02 ± 0.17 | < 0.001 |

LGCM, lateral gastrocnemius; PL, peroneus longus; TA, tibialis anterior.
[a]*p* value from paired t-test.

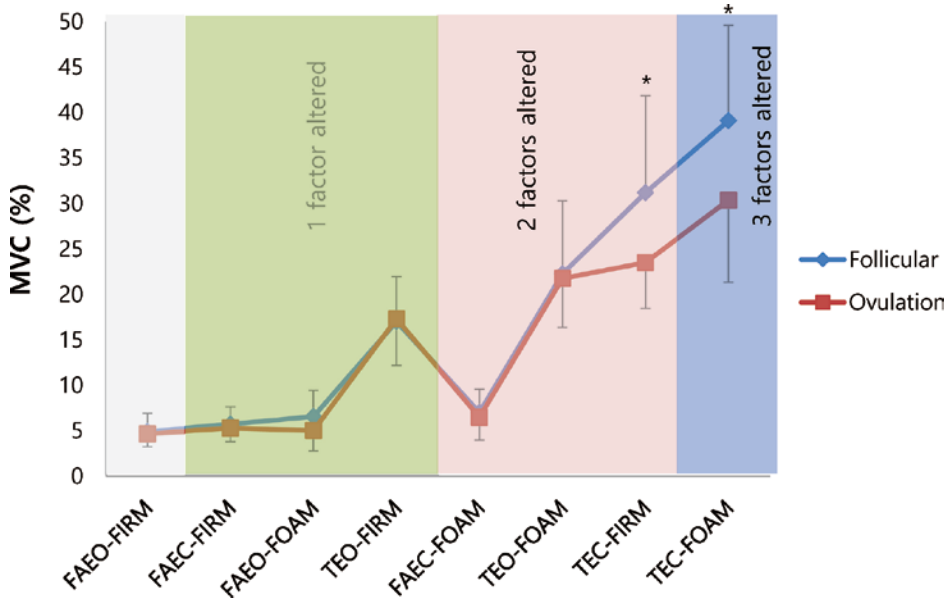

Fig. 2. Peroneus longus muscle activity between follicular phase and ovulation.
The data are shown as mean ± SD of EMG activity on peroneus longus muscle in different stations of balance platform between the follicular phase and ovulation (*significant difference).

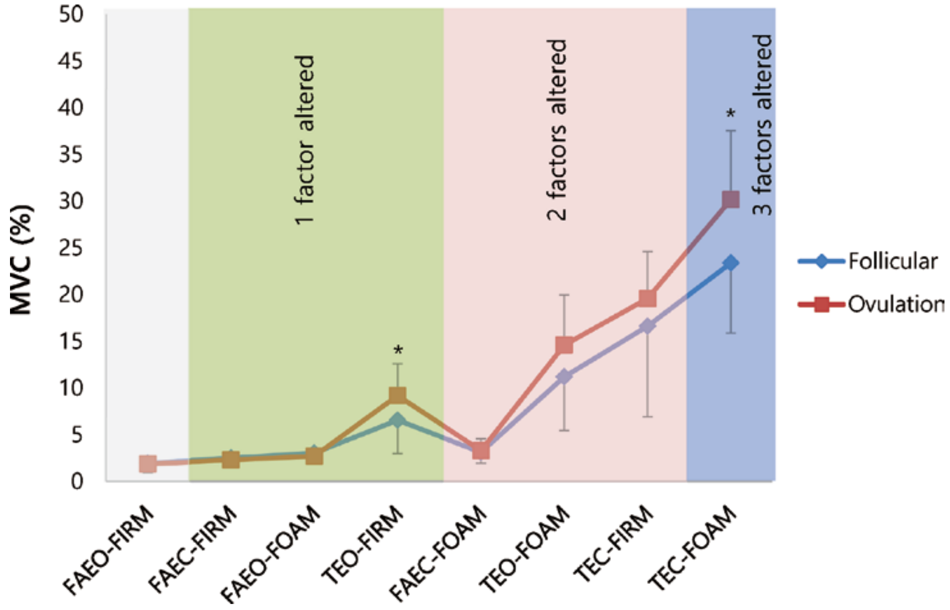

Fig. 3. Tibialis anterior muscle activity between follicular phase and ovulation.
The data are shown as mean ± SD of EMG activity on tibialis anterior muscle in different stations of balance platform between the follicular phase and ovulation (*significant difference).

FAEO-FIRM, FAEC-FIRM, FAEO-FOAM, and FAEC-FOAM ($p < 0.001$). PL muscle activation was significantly higher in the follicular phase than at ovulation during the TEC-FIRM and TEC-FOAM tasks ($p = 0.016$ and 0.008, respectively; Table 2, Fig. 2).

Similarly, a significant difference in TA muscle activity among the different balance tasks in both the follicular phase ($F = 44.107$, $p < 0.001$, $\eta^2 = 0.772$) and ovulation ($F = 96.519$, $p < 0.001$, $\eta^2 = 0.881$) was observed. TA muscle activity also significantly increased in TEO-FIRM, TEO-FOAM, TEC-FIRM, and TEC-FOAM as compared to that in FAEO-FIRM, FAEC-FRIM, FAEO-FOAM, and FAEC-FOAM ($p < 0.001$). However, TA muscle activation showed a different pattern between the two phases. TA muscle activity was significantly higher in ovulation than in the follicular phase in the TEO-FIRM and TEC-FOAM tasks ($p = 0.032$ and 0.018, respectively; Table 2, Fig. 3). Moreover, the TA-PL co-contraction (TA/PL ratio) was significantly higher at ovulation than in the follicular phase for the TEC-FIRM and TEC-FOAM balance tasks ($p = 0.003$

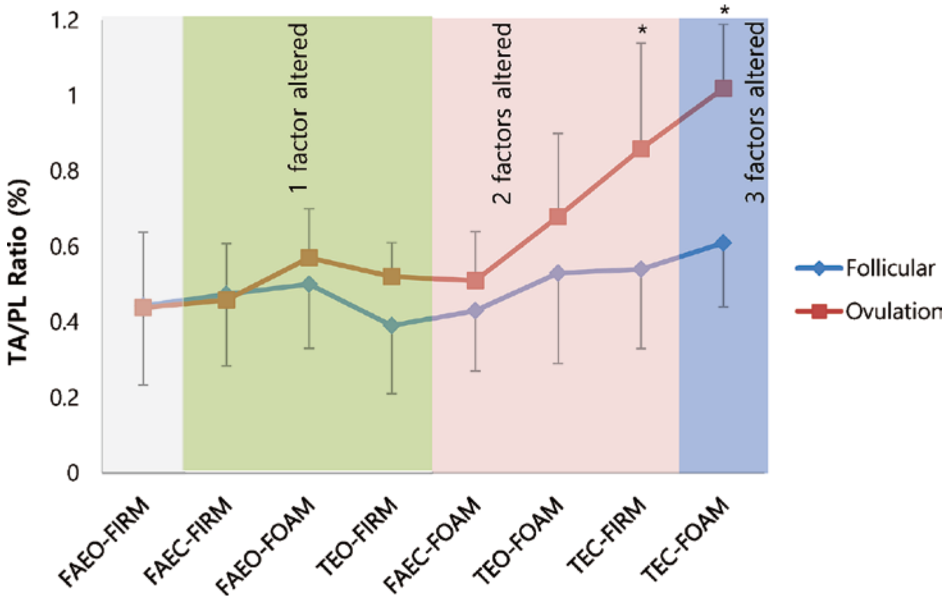

Fig. 4. TA/PL ration between follicular phase and ovulation.
The data are shown as mean ± SD of TA/PL ratio in different stations of balance platform between the follicular phase and ovulation (*significant difference).

and < 0.001, respectively; Table 2, Fig. 4).

## Discussion

In the present study, the investigator compared differences in the postural sway and neuromuscular activities of the ankle-stabilizing muscles during the menstrual cycle in healthy young women. We observed significantly greater postural sway during ovulation, when the estradiol concentration peaks, during the two most difficult balance tests, compared to the follicular phase, when estradiol levels are comparably low. This would support the previous evidence pointing to a greater reduction in balance ability and tremor during ovulation than during the early follicular phase (menstruation) (Petrofsky and Lee 2015). Likewise, other studies reported a significant increase in postural sway in the mid-luteal phase, when estrogen levels are relatively higher, in women with premenstrual symptoms (Friden et al. 2003, 2005). A previous study also showed greater ankle laxity at ovulation than in other phases of the menstrual cycle (Shultz et al. 2012). This would support the finding that the connective tissues in the foot and ankle complex are affected by estradiol levels. However, the exact variability of ankle laxity throughout the menstrual cycle remains controversial. In fact, Ericksen and colleagues showed no differences in ankle laxity or dynamic postural control according to hormonal fluctuations during the menstrual cycle (Ericksen and Gribble 2012).

Postural sway rate increased during the difficult balance tasks, which altered two or more factors that are known to affect balance throughout the menstrual cycle (Petrofsky and Lee 2015). However, the neuromuscular activation of the ankle was not assessed at the same times throughout the menstrual cycle.

The results of our study demonstrate the differences in ankle stabilizing muscle activation in the two most difficult balance tasks (TEC-FIRM and TEC-FOAM) between the time of ovulation, when serum estradiol concentrations peak, and in the early follicular (menstruation) phase, when serum estradiol concentrations are relatively low. In the same manner, our results revealed that when women are faced with a reduced capacity to control postural sway, as in the two most difficult balance tasks, neuromuscular activation relied more on the TA during ovulation than during the follicular phase. A previous study reported that at ovulation, decreased quadriceps activity, which was associated with increased hamstring activity in the modulation of knee joint stiffness, might be affected by elevated estradiol concentrations (Khowailed et al. 2015). Although the findings did not show a negative relationship between the PL and TA, such as that of the quadriceps and hamstring, in terms of neuromuscular activation, these muscles showed different neuromuscular activation patterns throughout the menstrual cycle.

Increased activation of the TA in the two most difficult balance tasks may compensate for the increased postural sway due to more extreme ankle laxity at ovulation than in the early follicular phase. As TA activation increased, PL activation decreased when the two-altered (TEC-FIRM) and three-altered (TEC-FOAM) sensory factors acted together. Thus, the TA/PL ratio was significantly greater at ovulation than in the early follicular phase. De Ridder and colleagues suggested that a higher ratio indicated invertor/evertor co-contraction in favor of the invertors, which revealed that the TA muscle is more targeted than the PL when the TA/PL ratio is high (De Ridder et al. 2015).

Our hypothesis was that at times of high estradiol concentrations, such as ovulation, postural sway increases, which might subsequently alter the neuromuscular activation of the ankle-stabilizing muscles. Previous studies investigating neuromuscular activation of the knee during the menstrual cycle in women found significant differences in muscular strategies corresponding to the changes in estradiol serum concentrations (Khowailed et al. 2015). The participants in the present study also demonstrated significant TA and PL neuromuscular co-contraction strategies of the ankle in the most difficult balance tasks at ovulation versus during the early follicular phase.

Although this study achieved its aims, it has several limitations. Estradiol serum levels were not actually measured, but were assumed based on self-reported detection of the ovulation and menstruation phases. In addition, we only examined one full menstrual cycle in each participant. Lastly, shifts in the neuromuscular activation of the ankle stabilizing muscles in different situations such as dynamic balance, walking, or running should be considered in future studies.

Based on the present study, it would be of benefit to healthy young women to recognize the variation in neuromuscular activation patterns throughout their menstrual cycles. Furthermore, health professionals working with this population should also be aware of these physiological effects and the accompanying neuromuscular strategies in order to prevent increased risk of lower extremity injury.

## Conflict of Interest

The authors declare no conflict of interest.

## Acknowledgments

This research was supported by the Gachon University research fund of 2015 (GCU-2015-0059). Moreover, we thank Kyungtae Kim and all the personnel at the women's health and physical activity laboratory at Gachon University.

## References

Baumhauer, J.F., Alosa, D.M., Renstrom, A.F., Trevino, S. & Beynnon, B. (1995) A prospective study of ankle injury risk factors. *Am. J. Sports Med.*, **23**, 564-570.

Beynnon, B.D., Bernstein, I.M., Belisle, A., Brattbakk, B., Devanny, P., Risinger, R. & Durant, D. (2005) The effect of estradiol and progesterone on knee and ankle joint laxity. *Am. J. Sports Med.*, **33**, 1298-1304.

Boden, B.P., Sheehan, F.T., Torg, J.S. & Hewett, T.E. (2010) Noncontact anterior cruciate ligament injuries: mechanisms and risk factors. *J. Am. Acad. Orthop. Surg.*, **18**, 520-527.

Cattagni, T., Scaglioni, G., Laroche, D., Gremeaux, V. & Martin, A. (2016) The involvement of ankle muscles in maintaining balance in the upright posture is higher in elderly fallers. *Exp. Gerontol.*, **77**, 38-45.

Cattagni, T., Scaglioni, G., Laroche, D., Van Hoecke, J., Gremeaux, V. & Martin, A. (2014) Ankle muscle strength discriminates fallers from non-fallers. *Front. Aging Neurosci.*, **6**, 336.

De Ridder, R., Willems, T., De Mits, S., Vanrenterghem, J. & Roosen, P. (2014) Foot orientation affects muscle activation levels of ankle stabilizers in a single-legged balance board protocol. *Hum. Mov. Sci.*, **33**, 419-431.

De Ridder, R., Willems, T., Vanrenterghem, J. & Roosen, P. (2015) Influence of balance surface on ankle stabilizing muscle activity in subjects with chronic ankle instability. *J. Rehabil. Med.*, **47**, 632-638.

Deie, M., Sakamaki, Y., Sumen, Y., Urabe, Y. & Ikuta, Y. (2002) Anterior knee laxity in young women varies with their menstrual cycle. *Int. Orthop.*, **26**, 154-156.

Duclos, N.C., Maynard, L., Barthelemy, J. & Mesure, S. (2014) Postural stabilization during bilateral and unilateral vibration of ankle muscles in the sagittal and frontal planes. *J. Neuroeng. Rehabil.*, **11**, 130.

Eiling, E., Bryant, A.L., Petersen, W., Murphy, A. & Hohmann, E. (2007) Effects of menstrual-cycle hormone fluctuations on musculotendinous stiffness and knee joint laxity. *Knee Surg. Sports Traumatol. Arthrosc.*, **15**, 126-132.

Ellis, J.E., Johnson, S.J., Shaw, R. & Godbert, S. (2011) Superiority of Clearblue home ovulation tests in detecting the peak fertile days of the menstrual cycle compared to a simple calender method. *Hum. Reprod.*, **26**, I76-I77.

Ericksen, H. & Gribble, P.A. (2012) Sex differences, hormone fluctuations, ankle stability, and dynamic postural control. *J. Athl. Train*, **47**, 143-148.

Friden, C., Hirschberg, A.L., Saartok, T., Backstrom, T., Leanderson, J. & Renstrom, P. (2003) The influence of premenstrual symptoms on postural balance and kinesthesia during the menstrual cycle. *Gynecol. Endocrinol.*, **17**, 433-439.

Friden, C., Ramsey, D.K., Backstrom, T., Benoit, D.L., Saartok, T. & Linden Hirschberg, A. (2005) Altered postural control during the luteal phase in women with premenstrual symptoms. *Neuroendocrinology*, **81**, 150-157.

Hansen, M., Kongsgaard, M., Holm, L., Skovgaard, D., Magnusson, S.P., Qvortrup, K., Larsen, J.O., Aagaard, P., Dahl, M., Serup, A., Frystyk, J., Flyvbjerg, A., Langberg, H. & Kjaer, M. (2009a) Effect of estrogen on tendon collagen synthesis, tendon structural characteristics, and biomechanical properties in postmenopausal women. *J. Appl. Physiol.*, **106**, 1385-1393.

Hansen, M., Langberg, H., Holm, L., Miller, B.F., Petersen, S.G., Doessing, S., Skovgaard, D., Trappe, T. & Kjaer, M. (2011) Effect of administration of oral contraceptives on the synthesis and breakdown of myofibrillar proteins in young women. *Scand. J. Med. Sci. Sports*, **21**, 62-72.

Hansen, M., Miller, B.F., Holm, L., Doessing, S., Petersen, S.G., Skovgaard, D., Frystyk, J., Flyvbjerg, A., Koskinen, S., Pingel, J., Kjaer, M. & Langberg, H. (2009b) Effect of administration of oral contraceptives in vivo on collagen synthesis in tendon and muscle connective tissue in young women. *J. Appl. Physiol.*, **106**, 1435-1443.

Heitz, N.A., Eisenman, P.A., Beck, C.L. & Walker, J.A. (1999) Hormonal changes throughout the menstrual cycle and increased anterior cruciate ligament laxity in females. *J. Athl. Train*, **34**, 144-149.

Hosea, T.M., Carey, C.C. & Harrer, M.F. (2000) The gender issue: epidemiology of ankle injuries in athletes who participate in basketball. *Clin. Orthop. Relat. Res.*, 45-49.

Kalron, A. & Achiron, A. (2013) Postural control, falls and fear of falling in people with multiple sclerosis without mobility aids. *J. Neurol. Sci.*, **335**, 186-190.

Khowailed, I.A., Petrofsky, J., Lohman, E., Daher, N. & Mohamed, O. (2015) 17beta-Estradiol Induced Effects on Anterior Cruciate Ligament Laxness and Neuromuscular Activation Patterns in Female Runners. *J. Womens Health* (*Larchmt*), **24**, 670-680.

Kjaer, M. & Hansen, M. (2008) The mystery of female connective tissue. *J. Appl. Physiol.*, **105**, 1026-1027.

Lee, H., Petrofsky, J.S., Daher, N., Berk, L. & Laymon, M. (2014) Differences in anterior cruciate ligament elasticity and force for knee flexion in women: oral contraceptive users versus non-oral contraceptive users. *Eur. J. Appl. Physiol.*, **114**, 285-294.

Lee, H., Petrofsky, J.S., Daher, N., Berk, L., Laymon, M. & Khowailed, I.A. (2013a) Anterior cruciate ligament elasticity and force for flexion during the menstrual cycle. *Med. Sci. Monit.*, **19**, 1080-1088.

Lee, H., Petrofsky, J.S., Laymon, M. & Yim, J. (2013b) A greater reduction of anterior cruciate ligament elasticity in women compared to men as a result of delayed onset muscle soreness. *Tohoku J. Exp. Med.*, **231**, 111-115.

Lee, H., Petrofsky, J.S. & Yim, J. (2015) Do Oral Contraceptives Alter Knee Ligament Damage with Heavy Exercise? *Tohoku J. Exp. Med.*, **237**, 51-56.

Liu, S.H., Al-Shaikh, R., Panossian, V., Yang, R.S., Nelson, S.D., Soleiman, N., Finerman, G.A. & Lane, J.M. (1996) Primary immunolocalization of estrogen and progesterone target cells in the human anterior cruciate ligament. *J. Orthop. Res.*, **14**, 526-533.

Liu, S.H., Al-Shaikh, R.A., Panossian, V., Finerman, G.A. & Lane, J.M. (1997) Estrogen affects the cellular metabolism of the anterior cruciate ligament. A potential explanation for female athletic injury. *Am. J. Sports Med.*, **25**, 704-709.

Liu, X. & Luo, Z.P. (2005) Combined effects of estrogen and mechanical loading on anterior cruciate ligament fibroblast biosynthesis. *Scientific World Journal*, **5**, 5-8.

Louwerens, J.W., van Linge, B., de Klerk, L.W., Mulder, P.G. & Snijders, C.J. (1995) Peroneus longus and tibialis anterior muscle activity in the stance phase. A quantified electromyographic study of 10 controls and 25 patients with chronic ankle instability. *Acta. Orthop. Scand.*, **66**, 517-523.

Miller, B.F., Hansen, M., Olesen, J.L., Schwarz, P., Babraj, J.A., Smith, K., Rennie, M.J. & Kjaer, M. (2007) Tendon collagen synthesis at rest and after exercise in women. *J. Appl. Physiol.*, **102**, 541-546.

Page, P., Frank, C.C. & Lardner, R. (2010) *Assessment and treatment of muscle imbalance: the Janda approach*, Human Kinetics, Champaign, IL.

Park, S.K., Stefanyshyn, D.J., Loitz-Ramage, B., Hart, D.A. & Ronsky, J.L. (2009a) Changing hormone levels during the menstrual cycle affect knee laxity and stiffness in healthy female subjects. *Am. J. Sports Med.*, **37**, 588-598.

Park, S.K., Stefanyshyn, D.J., Ramage, B., Hart, D.A. & Ronsky, J.L. (2009b) Alterations in knee joint laxity during the menstrual cycle in healthy women leads to increases in joint loads during selected athletic movements. *Am. J. Sports Med.*, **37**, 1169-1177.

Petrofsky, J. & Lee, H. (2015) Greater Reduction of Balance as a Result of Increased Plantar Fascia Elasticity at Ovulation during the Menstrual Cycle. *Tohoku J. Exp. Med.*, **237**, 219-226.

Pollard, C.D., Braun, B. & Hamill, J. (2006) Influence of gender, estrogen and exercise on anterior knee laxity. *Clin. Biomech.* (*Bristol, Avon*), **21**, 1060-1066.

Pozzi, F., Moffat, M. & Gutierrez, G. (2015) Neuromuscular Control during Performance of a Dynamic Balance Task in Subjects with and without Ankle Instability. *Int. J. Sports Phys. Ther.*, **10**, 520-529.

Rozzi, S.L., Lephart, S.M., Gear, W.S. & Fu, F.H. (1999) Knee joint laxity and neuromuscular characteristics of male and female soccer and basketball players. *Am. J. Sports Med.*, **27**, 312-319.

Shultz, S.J., Kirk, S.E., Johnson, M.L., Sander, T.C. & Perrin, D.H. (2004) Relationship between sex hormones and anterior knee laxity across the menstrual cycle. *Med. Sci. Sports Exerc.*, **36**, 1165-1174.

Shultz, S.J., Sander, T.C., Kirk, S.E. & Perrin, D.H. (2005) Sex differences in knee joint laxity change across the female menstrual cycle. *J. Sports Med. Phys. Fitness*, **45**, 594-603.

Shultz, S.P., Sitler, M.R., Tierney, R.T., Hillstrom, H.J. & Song, J. (2012) Consequences of pediatric obesity on the foot and ankle complex. *J. Am. Podiatr. Med. Assoc.*, **102**, 5-12.

Silbernagel, K.G., Brorsson, A., Olsson, N., Eriksson, B.I., Karlsson, J. & Nilsson-Helander, K. (2015) Sex Differences in Outcome After an Acute Achilles Tendon Rupture. *Orthop. J. Sports Med.*, **3**, 2325967115586768.

Stijak, L., Kadija, M., Djulejic, V., Aksic, M., Petronijevic, N., Markovic, B., Radonjic, V., Bumbasirevic, M. & Filipovic, B. (2015) The influence of sex hormones on anterior cruciate ligament rupture: female study. *Knee Surg. Sports Traumatol. Arthrosc.*, **23**, 2742-2749.

Trimble, M.H., Bishop, M.D., Buckley, B.D., Fields, L.C. & Rozea, G.D. (2002) The relationship between clinical measurements of lower extremity posture and tibial translation. *Clin. Biomech.* (*Bristol, Avon*), **17**, 286-290.

Vieira, T.M., Minetto, M.A., Hodson-Tole, E.F. & Botter, A. (2013) How much does the human medial gastrocnemius muscle contribute to ankle torques outside the sagittal plane? *Hum. Mov. Sci.*, **32**, 753-767.

Yu, W.D., Panossian, V., Hatch, J.D., Liu, S.H. & Finerman, G.A. (2001) Combined effects of estrogen and progesterone on the anterior cruciate ligament. *Clin. Orthop. Relat. Res.*, 268-281.