Walking patterns and hormonal cycles in females

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ATTESTATION OF AUTHORSHIP

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Chapters 2 and 3 of this thesis represent stand-alone papers that will be submitted to a peer-reviewed journal for consideration for publication. My contribution and the contribution by the various co-authors to each of these papers are outlined at the beginning of each chapter and in the "candidate contribution to co-authored papers" table. All co-authors have approved the inclusion of the joint work in this Masters' thesis.

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ABSTRACT

Concussion is a mild traumatic brain injury caused by direct or indirect biomechanical forces to the head or body, resulting in temporary disruption of brain function and a range of physical, cognitive, emotional, and sleep-related symptoms, often without structural abnormalities detectable on standard neuroimaging. Female athletes often experience prolonged symptoms and higher rates of secondary musculoskeletal (MSK) injuries following concussion compared to males, potentially due to hormonal fluctuations and physiological characteristics that influence neurological recovery and physical function. This thesis explores the relationship between menstrual cycle-related hormonal changes and gait alterations, and their potential implications for concussion outcomes, recurrent concussion, and secondary musculoskeletal injury susceptibility in females.

Despite growing evidence linking concussion-related hormonal disruption, menstrual cycle changes, and post-concussion gait alterations, limited research has examined how these factors interact. To address this gap, a scoping review was conducted to evaluate current evidence on the influence of menstrual cycle related hormonal changes and gait alterations on concussion outcomes their potential implications for secondary musculoskeletal injury and recurrent concussion in females. The literature review highlighted that fluctuations in oestrogen and progesterone may affect symptom severity and recovery, with injuries sustained during the luteal phase often associated with more pronounced symptoms. In addition, gait-related studies demonstrated persistent post-concussion impairments, particularly during dual-task conditions, including reduced walking speed, altered cadence, and impaired stability, which may increase vulnerability to secondary MSK injuries. However, no studies directly investigated the combined relationship between menstrual cycle-related hormonal changes and gait alterations.

To further explore this relationship, a prospective observational pilot case series study was conducted between 1 October 2025 and 15 March 2026. The study included 10 healthy female participants with no history of concussion or musculoskeletal injury, who attended multiple study visits (up to 12 sessions) over a three-month period. Participants completed questionnaires, performed single-task, dual-task, and head-turn walking assessments, and provided blood samples for hormonal analysis. Significant hormonal variations occurred across menstrual phases, particularly in oestradiol, progesterone, luteinising hormone, follicle-stimulating hormone, testosterone, thyroid-stimulating hormone, and dehydroepiandrosterone. However, no significant differences were identified in gait parameters across menstrual phases, with gait performance remaining stable despite hormonal fluctuations.

The findings contribute new insights into the relationship between menstrual cycle-related hormonal changes and gait alterations, suggesting that hormonal fluctuations alone may not substantially influence gait in healthy females. Although limited by the small sample size and absence of concussed participants, this study provides important feasibility data and methodological guidance for future research investigating the relationship between menstrual cycle-related hormonal changes and gait alterations, and their potential implications for secondary musculoskeletal injury and recurrent concussion in females recovering from concussion.

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LIST OF ABBREVIATIONS

ACL= Anterior Cruciate Ligament

BIST= Brain Injury Screening Tool

CNS=Central Nervous System

COM = Centre of Mass

CRQA = Cross-Recurrence Quantification Analysis

CT = Computed Tomography

DHEA-S = Dehydroepiandrosterone sulphate

DTC = Dual Task Cost

DTC-C = Cognitive Dual Task Cost

DTC-M = Motor Dual Task Cost

DT = Dual Task

EMG = Electromyography

EuroQol = European Quality of Life Questionnaire

EDTA= Ethylenediaminetetraacetic acid

FP = Follicular Phase

FSH = Follicle-Stimulating Hormone

FT4 = Free Thyroxine (T4)

GnRH = Gonadotropin Releasing Hormone

HR = Hazard Ratio

HT = Head Turn

HPO = Hypothalamic Pituitary Ovarian Axis

IGA = Instrumental Gait Analysis

ImPACT = Immediate Post-Concussion Assessment and Cognitive Testing

ID = Identification

LH = Luteinizing Hormone

MC = Menstrual Cycle

MP = Menstrual Phase

MRI = Magnetic Resonance Imaging

MMP = Matrix Metalloproteinases

MSK = Musculoskeletal

mTBI = Mild Traumatic Brain Injury

OP = Ovulatory Phase

OCPs = Oral Contraceptive Pills

PCSI = Post-Concussion Symptom Inventory

PIS = Participant Information Sheet

PSS = Perceived Stress Scale

rCBF = Regional Cerebral Blood Flow

SD = Standard Deviation

SE = Standard Error

SRBI = Sports-Related Brain Injury

SRC = Sports-Related Concussion

ST = Single Task

T4 = Thyroxine

TSH = Thyroid-Stimulating Hormone

WHO = World Health Organization

DEFINITIONS

Concussion

Concussion has been defined by World Health Organization (WHO) in 2004 as a temporary brain function disturbance from a head/body impact, causing physical, cognitive, emotional, or sleep problems, with no immediate brain bleeds or swelling, often without loss of consciousness. Concussion symptoms include headache, dizziness, difficulties with memory or concentration, irritability, nausea, fatigue, sensitivity to light or noise, and disturbances in sleep. Symptoms of mild traumatic brain injury can range from mild, transient symptoms to extended periods of altered consciousness. However, most individuals who sustain a concussion, particularly athletes, experience complete symptom resolution and a favourable recovery within 10 days following the injury (McCrory et al., 2017).

Menstrual cycle

Menstrual health is an important aspect of female biological functioning, playing a vital role from puberty through to menopause. The menstrual cycle is a monthly sequence of hormonal changes that prepares the body for pregnancy, beginning on the first day of menstruation and ending the day before the next menstrual period. This cycle is regulated by fluctuations in key hormones, including follicle-stimulating hormone (FSH), luteinizing hormone (LH), oestrogen, and progesterone, which influence changes in both the ovaries and the uterus. Although the average menstrual cycle lasts approximately 28 days, a normal cycle can range between 21 and 35 days. The cycle consists of four main phases: menstruation, the follicular phase, ovulation, and the luteal phase. If pregnancy does not occur, the thickened uterine lining, along with blood and tissue, is shed from the body during menstruation (Hennegan et al., 2021).

Gait

Walking is a specific form of gait and the most common mode of human locomotion. The gait cycle begins when one-foot contacts the ground and ends when the same foot contacts the ground again. It is divided into two primary phases: the stance phase, during which the foot remains in contact with the ground, and the swing phase, when the foot moves forward through the air(Webster & Darter, 2019). A broad spectrum of potential assessment strategies used to evaluate normal and abnormal gait, and it is important in the assessment of patients during the process of rehabilitation and recovery from the effects of neurological disease, musculoskeletal injury, disease progress or amputation of a lower limp. Gait assessments range from simple clinical observation to computer analysis of biomechanics and spatiotemporal parameters (stride length, step time, cadence (steps/minute), stance time, swing time, and double support time), termed Instrumental Gait Analysis (IGA) (Hulleck et al., 2022).

Secondary Musculoskeletal (MSK) injury

In this thesis, a secondary musculoskeletal injury is defined as a new injury to muscles, bones, joints, tendons, nerves, ligaments, and related soft tissue occurring after the index concussion. Secondary musculoskeletal injury risk refers to the likelihood of sustaining such an injury during the specified post-concussion follow-up

period (Howell et al., 2018). This concept is distinct from recurrent concussion, which refers to a subsequent concussive event. Persistent impairments in balance, gait, neuromuscular control or movement strategy following concussion may contribute to increased susceptibility to secondary musculoskeletal injury (Howell et al., 2018).

CHAPTER 1: INTRODUCTION

1.1 Background

Concussion is a mild traumatic brain injury (mTBI) caused by direct or indirect biomechanical forces to the head or body, resulting in temporary disruption of brain function and a range of physical, cognitive, emotional, and sleep-related symptoms, often without structural abnormalities detectable on standard neuroimaging (McCrory et al., 2017). Recovery following mTBI is generally described across acute, subacute, and chronic phases. The acute phase occurs immediately after injury and is marked by neurometabolic disturbances and symptom onset, while the subacute phase involves ongoing physiological recovery and gradual symptom resolution over days to weeks. The chronic phase refers to the persistence of symptoms beyond the expected recovery period, with some individuals experiencing prolonged post-concussion symptoms that may continue for months or longer (Mayer et al., 2017).

Female athletes frequently experience longer symptom duration and prolonged recovery compared to males (McCrory et al., 2017). These sex differences are thought to be influenced by hormonal fluctuations and physiological characteristics unique to females, which may affect both neurological recovery and physical function following injury (Kara et al., 2020). During concussion recovery, persistent alterations in balance, gait, neuromuscular control, and movement strategies may increase susceptibility to subsequent musculoskeletal (MSK) injury (Howell et al., 2018; Lynall et al., 2015). In this thesis, secondary MSK injury risk refers specifically to the risk of sustaining a new MSK injury following concussion. Individuals recovering from concussion may also be at increased risk of recurrent head trauma; however, the mechanisms underlying this increased injury susceptibility in females remain poorly understood.

Emerging evidence suggests that concussion may disrupt normal neuroendocrine function, particularly affecting the hypothalamic pituitary gonadal axis, which regulates reproductive hormone production. In females, this disruption may alter menstrual cycle patterns and hormone concentrations, potentially influencing both recovery and long-term health outcomes. For example, 90% of females reported menstrual irregularities, including missed periods, within the year following brain injury compared to only 23% prior to injury (Ripley et al., 2008), indicating significant neuroendocrine dysfunction. Furthermore, greater concussion severity has been associated with longer durations of amenorrhea, while shorter durations of menstrual disruption have been linked to better recovery and return to normal functioning.

Progesterone, a gonadal hormone primarily produced by the ovaries, plays an important neuroprotective role in the central nervous system through progesterone receptors widely distributed throughout the brain (Chen et al., 2021). During the menstrual cycle, progesterone remains low throughout the follicular and ovulatory phases before rising and peaking during the mid-luteal phase. Sports-related brain injury (SRBI) sustained during this phase may trigger an acute decline in progesterone levels, which has been associated with poorer recovery outcomes (Bazarian et al., 2010; Moreno, 2017; Snook et al., 2017). Conversely, elevated progesterone levels have been associated with lower Perceived Stress Scale (PSS) scores, suggesting reduced perceived stress and more effective physiological regulation. Similarly, oestrogen is another key gonadal hormone that fluctuates across the menstrual cycle and may influence neurological response to injury (Moreno, 2017). Oestrogen levels peak prior to ovulation, and if head trauma occurs when oestrogen levels are at their

peak, a sudden decrease in circulating oestrogen may occur, potentially worsening concussion symptoms and negatively affecting recovery outcomes (Moreno, 2017; Snook et al., 2017). Together, these findings suggest that hormonal fluctuations and concussion-related neuroendocrine disruption may play a significant role in female-specific recovery trajectories.

While oestrogen and progesterone are important ovarian hormones involved in menstrual-cycle regulation, FSH, LH, and thyroid-stimulating hormone (TSH) also contribute to the regulation of reproductive function (Orlowski & Sarao, 2026). FSH and LH are released from the anterior pituitary gland in response to pulsatile gonadotropin-releasing hormone (GnRH) from the hypothalamus. FSH promotes the growth and maturation of ovarian follicles and supports oestrogen production (Orlowski & Sarao, 2026), while LH contributes to follicular maturation and triggers ovulation through the mid-cycle LH surge (Reed & Carr, 2018). Following ovulation, LH also supports corpus luteum function and progesterone production (Reed & Carr, 2018). TSH regulates thyroid hormone production, which is closely linked with reproductive function and helps maintain normal ovarian activity and menstrual-cycle regularity (Jacobson et al., 2018). Consequently, changes in FSH, LH, or TSH may influence follicular development, ovulation, ovarian hormone production, and menstrual-cycle patterns.

Following concussion, disruption of hypothalamic or pituitary pathways may affect GnRH signalling and subsequently alter FSH and LH secretion (Snook et al., 2017). Such alterations may interfere with follicular development, ovulation, and the normal timing or regularity of the menstrual cycle (Snook et al., 2017). In addition, activation of the hypothalamic-pituitary-adrenal axis following concussion may further influence reproductive hormone signalling (Carr & Fleddermann, 2025). Menstrual disturbances have been reported following concussion, with one study finding significantly higher odds of multiple abnormal menstrual patterns following concussion (OR 5.85, 95% CI 1.61-21.22) (Snook et al., 2017). Concussion-related hypothalamic-pituitary dysfunction may also affect the hypothalamic-pituitary-thyroid axis, potentially contributing to altered TSH and thyroid hormone regulation. Changes in thyroid hormone levels may further influence reproductive function through effects on sex hormone-binding globulin, ovarian function, and the metabolism and clearance of oestrogen. Consequently, thyroid dysfunction may contribute to menstrual disturbances, including altered bleeding patterns, irregular cycles, or amenorrhoea (Kapper et al., 2024).

These endocrine alterations may also be relevant to concussion recovery because reproductive and thyroid hormones are involved in physiological processes that influence neuromuscular function, energy metabolism, and physical performance (Rettberg et al., 2014). Consequently, disruption of the hypothalamic-pituitary-gonadal or hypothalamic-pituitary-thyroid axes following concussion may potentially affect movement control and physical function during recovery (Ripley et al., 2008). In particular, altered hormonal profiles could contribute to changes in neuromuscular function or movement strategies, which may be relevant to gait and balance during physical activity (Chen et al., 2021). Such changes may potentially influence mechanical loading and movement control, thereby contributing to susceptibility to subsequent injury; however, the relationship between post-concussion endocrine changes, gait alterations, recovery, and secondary injury risk remains unclear (Messerschmidt et al., 2021). Therefore, examining FSH, LH, and TSH alongside oestrogen and progesterone may provide a more comprehensive understanding of hormonal changes

associated with menstrual-cycle regulation and their potential relevance to physiological recovery, gait, and injury susceptibility following concussion.

Beyond their effects on brain recovery, hormonal fluctuations across the menstrual cycle may also influence anatomical and physiological factors related to injury risk. Relaxin, a peptide hormone produced by the corpus luteum following ovulation, is biologically active in both pregnant and non-pregnant females and typically peaks during the luteal phase of the menstrual cycle (Goldsmith et al., 1995). Relaxin has endocrine and immunological functions and works synergistically with oestrogen to stimulate matrix metalloproteinases (MMPs), which degrade type I collagen and suppress new collagen synthesis (Galey et al., 2003). As collagen is a major structural component of ligaments, these effects may reduce collagen density and strength, compromise ligament integrity and increasing susceptibility to injury. This may be particularly relevant for the anterior cruciate ligament (ACL), as female ACL tissue has been shown to bind significantly greater amounts of relaxin compared to males (Dragoo et al., 2003). Peak serum relaxin concentrations typically occur between menstrual cycle days 21–24, a time associated with increased ACL injury risk (Parker et al., 2024). Interestingly, oral contraceptive use has been shown to reduce serum relaxin concentrations, suggesting a potential protective effect against ACL injury (DeFroda et al., 2019).

Menstrual-cycle characteristics can vary both between individuals and across cycles, meaning that menstrual phase alone may not consistently represent an individual's underlying hormonal or physiological state. Conditions such as polycystic ovary syndrome (PCOS), menstrual irregularity, and hormonal contraceptive use can alter menstrual patterns and endogenous hormonal profiles, contributing to variability in physiological and neuromuscular responses. Fluctuation in oestradiol, progesterone, LH, and FSH may influence neuromuscular function, including muscle activation, joint stability, postural control, and coordination (Colenso-Semple et al., 2023; Elliott-Sale et al., 2021; McNulty et al., 2020).

Menstrual irregularities, including oligomenorrhoea, polymenorrhoea, amenorrhoea, anovulatory cycles, and luteal-phase deficiency, are relatively common among female athletes and may also be associated with injury risk (Cheng et al., 2021; Nédélec et al., 2021). Suzuki-Yamanaka et al. (2025) reported differences in injury risk across menstrual phases in eumenorrheic athletes, while menstrual dysfunction has been associated with more severe injuries and an increased susceptibility to bone stress injuries (Cheng et al., 2021; Thein-Nissenbaum et al., 2012). Hormonal contraceptive use and PCOS may further contribute to differences in hormonal and neuromuscular responses. Although oral contraceptive pill (OCP) use has been associated with greater knee stability and reduced ACL injury risk in some athletic populations (Herzberg et al., 2017; Hewett et al., 2007), hormonal contraceptives are not consistently protective against musculoskeletal injuries (White et al., 2023). PCOS is also associated with altered androgen and metabolic hormone profiles, which may influence muscle strength, power, aerobic capacity, and endurance (Caliskan Guzelce et al., 2019; Kamal et al., 2025; Sacinti et al., 2026). Therefore, menstrual regularity, contraceptive use, and conditions such as PCOS should be considered when interpreting associations between menstrual phase, hormonal concentrations, neuromuscular function, and gait outcomes (Colenso-Semple et al., 2023; Elliott-Sale et al., 2021; McNulty et al., 2020).

These hormonal and physiological variations may also influence postural control and gait, which are particularly relevant during concussion recovery. Studies have demonstrated greater sway and increased sway velocity of the whole-body centre of mass during walking in concussed individuals, particularly when performing a simultaneous cognitive task, with these impairments persisting for up to 28 days post-injury (Howell et al., 2020; Parker et al., 2006). Significant alterations have also been observed in spatiotemporal gait parameters, including step length, swing phase, double-support phase, gait speed, and cadence, particularly among individuals reporting ongoing concussion symptoms (Messerschmidt et al., 2021). Female athletes may experience greater functional impairment than males, showing larger reductions in gait speed, cadence, and swing phase, along with increased double-support time. These deficits may reflect persistent neuromotor dysfunction, potentially increasing vulnerability to secondary MSK injuries and repeated concussions.

Hormonal fluctuations during the menstrual cycle may further influence postural stability and gait mechanics. During ovulation, when oestrogen levels are highest, increased ligament elasticity and structural changes in the foot, including increased foot length and plantar fascia flexibility, have been associated with reduced balance performance and increased tremor during challenging balance tasks (Chen et al., 2021; Petrofsky & Lee, 2015). These physiological changes may contribute to greater postural instability and elevate the risk of falls or injury. Although previous studies have independently examined concussion-related hormonal disruption, menstrual cycle hormone fluctuations, and post-concussion gait impairments (Bazarian et al., 2010; DeFroda et al., 2019; Ripley et al., 2008), limited research has integrated these factors to understand their combined impact on secondary MSK injury risk in female athletes. Specifically, it remains unclear how menstrual-cycle-related hormonal changes may influence gait mechanics and balance following concussion and whether these changes contribute to future injury susceptibility. This represents a significant gap in current literature, particularly in relation to integrated models of post-concussion recovery in females.

1.2 Aim of thesis

This thesis aimed to investigate the relationship between menstrual cycle-related hormonal changes and gait alterations following concussion, and to explore their potential implications for secondary musculoskeletal injury risk by integrating hormonal testing with gait parameter analysis. Sport related concussion injuries often occur, female athletes are more susceptible to concussions, with prolonged symptoms and higher rates of secondary MSK injuries compared to males. So far, the only way to diagnose if someone has a mild or moderate traumatic brain injury (often called a concussion) is by scanning their brain with a CT or MRI scanner and performing several medical exams. There can be quite a delay between someone being injured and when they can be seen by medical specialist and given a CT or MRI scan.

Scientists have been trying to develop blood tests and gait analysis methods that would help to provide a fast and easy way to diagnose the recovery stage from concussion and prevent further injuries than how we currently do it. This research is trying to test if menstrual hormone's levels and gait parameters are useful to monitor risk of secondary MSK injuries and recurrent concussion.

Additionally, females seem to suffer from longer and worse symptoms after concussion. To identify if the menstrual cycle (period) plays a role in the severity of concussion symptoms, blood tests will be performed to look at the levels of female sex hormones. The levels of these hormones after injury may help to determine why females have worse symptoms compared with males and will help to design cycle specific gait and balance training to prevent further injuries and improve their performance.

1.3 Research questions

The following research questions were proposed and addressed in this thesis:

1. How do menstrual cycle-related hormonal changes influence post-concussion outcomes in females following concussion?
2. What gait alterations occur in females following concussion?
3. What evidence exists regarding the relationship between menstrual cycle-related hormonal changes and gait alterations and their potential implications for secondary musculoskeletal injury and recurrent concussion in females following concussion?

1.4 Thesis structure

This thesis consists of four chapters. Chapter 1 provides background information on the role of menstrual-cycle related hormonal changes in concussion, including their influence on symptom severity, gait alterations, and their potential implications for secondary musculoskeletal injury and recurrent concussion. It also reviews current research in this area and identifies existing gaps in knowledge that underpin the rationale for this thesis.

Chapter 2 presents a scoping review of recent studies examining the relationship between menstrual cycle-related hormonal changes and gait alterations, and their potential implications for secondary musculoskeletal injury and recurrent concussion, as well as concussion symptom severity, in females following concussion.

Chapter 3 presents the results of a prospective observational pilot study. Data were collected from October 2025 to March 2026, involving ten healthy females with regular menstrual cycles. The analysis provides insight into hormonal changes across the menstrual cycle in healthy participants, along with corresponding changes in gait across different phases. Unfortunately, due to time constraints and limited availability of naturally menstruating females with concussion, no concussed female participants could be recruited for this study. Collectively, these findings provide a preliminary understanding of the relationship between menstrual cycle-related hormonal changes and gait alterations in healthy females, with potential implications for future research investigating secondary musculoskeletal injury and recurrent concussion following concussion.

Chapter 4 presents a discussion of the key findings and the limitations of this thesis, along with recommendations for future research and concluding remarks. It discusses how concussion-related gait alterations may have potential implications for secondary musculoskeletal injury and recurrent concussion. These insights contribute to addressing the main research questions by exploring the relationships between

menstrual cycle-related hormonal changes, gait alterations, and their potential implications for injury-related outcomes. The thesis concludes with a reference section that includes all sources cited throughout the study.

PRELUDE TO CHAPTER 2

This thesis chapter investigates current literature on the relationship between the menstrual cycle-related hormonal changes and alterations in gait, and their potential implications for secondary musculoskeletal injury and recurrent concussion following concussion in females. With the increasing participation of women in sports, the incidence of concussion injuries is also expected to rise. Therefore, it is essential for researchers and clinicians to understand concussion mechanisms specific to females, as well as the prevalence and nature of associated injuries, to develop effective injury prevention strategies.

The aim of this chapter was to systematically review the global literature examining the influence of menstrual cycle-related hormonal changes and gait alterations, and concussion outcomes in females. Studies published in English since 2014 were included to address three key research questions. First, studies examining menstrual cycle-related hormonal changes in relation to post-concussion symptom severity were analysed to identify the hormones most associated with symptom burden and recovery outcomes. Second, studies investigating gait following concussion were reviewed to identify key gait alterations and their potential relevance to secondary musculoskeletal injury. Third, studies examining the relationship between hormonal changes and gait alterations were reviewed to determine whether evidence exists linking these factors with secondary musculoskeletal injury and recurrent concussion. However, no published studies were identified that directly examined this relationship, highlighting an important gap in the current evidence base.

This review provides a synthesis of current evidence on menstrual cycle phases, hormonal changes, gait alterations, and concussion outcomes in females. The findings highlight the limited evidence connecting these factors with secondary musculoskeletal injury and recurrent concussion and identify important priorities for future research and injury prevention.

CHAPTER 2: WALKING PATTERNS AND HORMONAL CYCLES IN FEMALES POST-CONCUSSION: A SCOPING REVIEW

Overview

Objective: This scoping review aimed to critically review the existing literature on menstrual cycle-related hormonal changes, gait alterations, and concussion outcomes in females following concussion, including evidence regarding their potential implications for secondary musculoskeletal injury and recurrent concussion.

Methods: A systematic search was conducted using CINAHL, SPORTDiscus, MEDLINE, Academic Search Complete (via EBSCO), and Scopus, databases for all studies published since 2014 that reported on menstrual cycle-related hormonal changes, gait alterations, and concussion outcomes in females following concussion. Studies examining the potential implications of these factors for secondary musculoskeletal injury and recurrent concussion were also considered for inclusion. Only studies published in English with full-text availability were included. No restrictions were placed on the country of publication or participant demographics.

Findings: Eleven studies met the inclusion criteria. The reviewed literature suggests that menstrual cycle-related hormonal fluctuations (oestrogen and progesterone) may influence concussion outcomes and symptom severity in females, with injuries sustained during the luteal phase associated with more pronounced symptom presentations compared to other phases. Included gait related studies show that females experience persistent post-concussion gait alterations, especially during dual-task conditions, such as reduced walking speed, altered cadence, and impaired stability, which may increase the risk of secondary musculoskeletal injury. However, no studies have directly examined the combined relationship between menstrual cycle phases, hormones and gait alterations, highlighting a significant gap in the literature.

Conclusions: Overall, current evidence suggests that menstrual cycle (MC) related hormonal fluctuations and post-concussion gait alteration may independently influence concussion-related outcomes in females. However, evidence regarding their potential implications for secondary musculoskeletal injury and recurrent concussion remains limited, and their combined relationship remains largely unexplored. This limits a comprehensive understanding of how hormonal status and physiological recovery following concussion may interact. Future research should address this gap to support more targeted, sex-specific assessment and rehabilitation strategies following concussion.

Highlights:

- Menstrual cycle related hormonal changes may influence concussion symptom severity and recovery outcomes.
- The luteal phase has been associated with poorer post-concussion outcomes, possibly due to progesterone withdrawal effects.
- Evidence regarding hormonal influence on recovery remains inconsistent across studies.
- Concussed females demonstrate significant gait alterations, particularly during dual-task conditions.

- Gait impairments (e.g., reduced speed, instability, altered cadence) are linked to increased secondary musculoskeletal injury risk.
- Females tend to exhibit greater dual-task cost and more cautious gait strategies compared to males.
- No existing studies have directly examined the combined effect of menstrual cycle related hormonal changes and gait alterations on secondary MSK injury risk.
- There is a critical need for integrated, sex-specific research in this area.

2.1 Introduction

Concussion is a brain injury that occurs when a direct or indirect impact on the head or body causes rapid movement of the brain within the skull. Although it often does not produce visible structural damage, it can disrupt normal brain function through neurometabolic and physiological changes (Abdelmalik et al., 2019; Essex et al., 2025; Woolf, 1992). Globally, brain injuries affect approximately 69 million individuals annually, with around 3.8 million cases linked to sports and recreational activities (Dick, 2009; Zuckerman et al., 2015). In sports involving both sexes, concussion incidence in female athletes is comparable to males; however, symptom presentation and recovery trajectories differ, with females often reporting greater symptom severity and more pronounced cognitive and emotional impairments following injury (Adams et al., 2018; Covassin et al., 2013; Merritt et al., 2019; Mez et al., 2020).

These sex-related differences in symptom burden and recovery suggest that biological factors may influence concussion outcomes. Female athletes appear to experience longer recovery periods and a higher incidence of secondary musculoskeletal injuries compared with males following concussion (Kara et al., 2020; McGroarty et al., 2020). This raises the possibility that endocrine and neuromuscular mechanisms may contribute to these differences.

One key biological system that may underpin these differences is the hypothalamic pituitary ovarian (HPO) axis, which regulates the secretion of gonadotropin-releasing hormone (GnRH), luteinizing hormone (LH), follicle-stimulating hormone (FSH), oestrogen, and progesterone. These hormones fluctuate across the menstrual cycle (typically 21–35 days; see Figures 1 and 2). While primarily reproductive in function, oestrogen and progesterone also play important roles in neurological regulation, including energy metabolism, fluid balance, thermoregulation, and neural excitability (Rettberg et al., 2014). This establishes a potential link between hormonal state and brain function. Given this relationship, concussion may disrupt the HPO axis and alter hormonal regulation. Emerging evidence suggests that concussion can lead to menstrual irregularities and disturbances in cognitive, emotional, and physiological functioning, including sleep and headache patterns (Carr & Fleddermann, 2025; Snook et al., 2017). This indicates a bidirectional interaction: concussion may affect endocrine function, while hormonal status at the time of injury may also influence symptom expression and recovery outcomes.

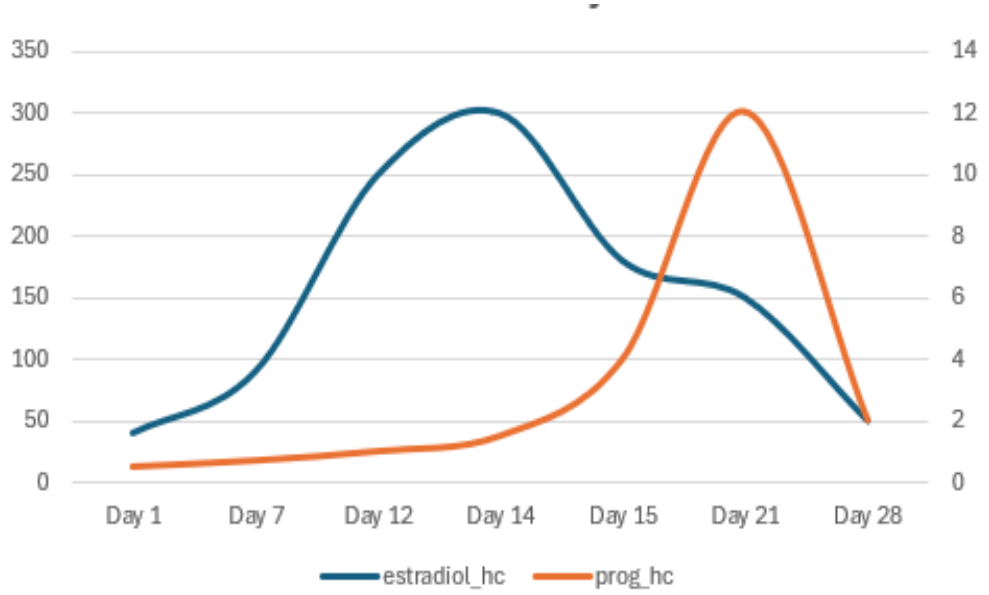

Figure 1 Hormonal fluctuations across the menstrual cycle phases- oestrogen & progesterone.

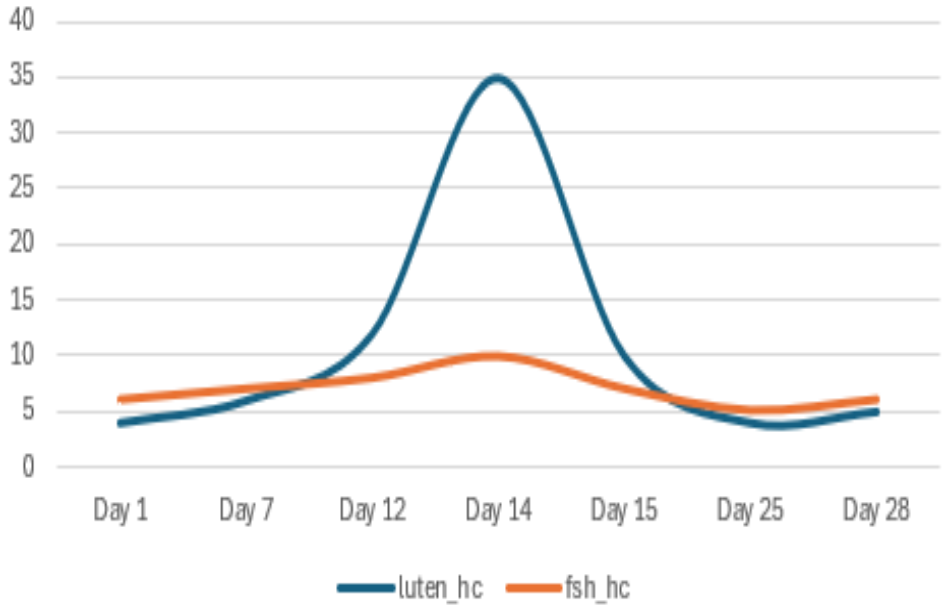

Figure 2 Hormonal fluctuations across the menstrual cycle phases- follicle-stimulating hormone (FSH) & luteinizing hormone (LH).

If hormonal state influences neurological and physiological responses to injury, it is also plausible that it affects musculoskeletal vulnerability. During the menstrual cycle, elevated oestrogen in the pre-ovulatory phase has been associated with reduced connective tissue stiffness, which may increase susceptibility to ligament injuries such as anterior cruciate ligament (ACL) tears (Hewett et al., 2007). In addition, relaxin peaks during the mid-luteal phase and has been linked to increased ligament laxity and injury risk (Parker et al., 2024). These hormonal fluctuations may also influence neuromuscular control, fatigue, and perceived exertion, thereby affecting injury risk (Chidi-Ogbolu & Baar, 2019). Oral contraceptive use has been associated with improved knee stability and reduced impact forces, further supporting a hormonal contribution to injury risk modulation (Herzberg et al., 2017).

In parallel, concussion is known to cause neuromotor and postural control deficits. Impairments in dynamic balance, such as reduced performance on the Y-Balance Test, have been associated with increased risk of concussion and reinjury (Seniloli et al., 2022). Even after clinical recovery, persistent gait alterations, including reduced walking speed and increased double support time have been observed (Martini et al., 2011). These deficits become more pronounced during dual-task conditions, further increasing injury susceptibility (Chou et al., 2023). Acute concussion is also associated with subtle neurocognitive impairments affecting attention, working memory, and visual processing, alongside vestibular–visual integration deficits contributing to balance dysfunction (Guskiewicz et al., 2001).

Importantly, these neuromotor deficits may not occur in isolation. Over longer recovery periods, concussion may also disrupt the HPO axis, leading to altered menstrual cycle regulation and hormonal instability (Lynall et al., 2020; Snook et al., 2017). This suggests that hormonal dysregulation and neuromotor impairment may co-exist, potentially compounding the risk of recurrent injury. However, the interaction between these two systems has not been well established.

Despite increased participation of female athletes in sport, existing research remains predominantly focused on male populations (Roberts & Forsyth, 2018). This gap is partly due to the added complexity introduced by hormonal variability across the menstrual cycle and sex-specific physiological differences (Costello et al., 2014; Oosthuyse & Bosch, 2010). Consequently, there is a lack of integrated evidence examining how menstrual hormone fluctuations and gait changes jointly influence concussion outcomes in females. Understanding this interaction is important because menstrual cycle phases may influence both symptom severity and neuromotor stability following concussion, potentially affecting recovery trajectories and secondary MSK injury risk. Identifying these relationships may support more targeted monitoring and rehabilitation strategies that account for hormonal status alongside functional recovery.

This scoping review therefore aimed to address the following questions:

1. How do menstrual cycle-related hormonal changes influence post-concussion symptom severity in females following concussion?
2. What gait alterations occur in females following concussion?
3. What evidence exists regarding the relationship between menstrual cycle-related hormonal changes and gait alterations in relation to secondary musculoskeletal injury and recurrent concussion in females following concussion?

2.2 Methods

This scoping review was conducted in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses extension for Scoping Review guidelines ("PRISMA Extension for Scoping Reviews (PRISMA-ScR): Checklist and Explanation,") (2018).

2.2.1 Eligibility

Types of studies

The review included a range of study designs, including prospective cohort studies, nested cohort studies, preliminary clinical studies, and cross-sectional studies, with follow-up periods ranging from a single post-injury assessment to several months post-injury. The eligibility criteria were limited to original human studies published between 2014 and 2025. The 2014 start date was selected to capture the relatively recent development of research examining menstrual-cycle-related factors in the context of concussion and gait.

Types of participants

The target population comprised naturally menstruating females with regular menstrual cycles, including both athletes and non-athletes, as well as concussed and non-concussed adults. Studies involving mixed populations were eligible only when data for naturally menstruating females with regular menstrual cycles were reported separately in the results.

Types of intervention/exposure

This review examined the effects of different menstrual cycle phases, hormonal levels, quality of life, and gait parameter alterations on concussion-related outcomes, including neurocognitive performance and symptom presentation, in female athletes and healthy individuals. Evidence regarding the potential implications of these factors for secondary musculoskeletal injury and recurrent concussion was also considered. All menstrual cycle phases were considered, including the menstruation phase (MP), follicular phase (FP), ovulation phase (OP), and luteal phase (LP). The review focused primarily on the influence of menstrual cycle phases and gait alterations on concussion-related outcomes, while also considering their potential relevance to secondary injury and recurrent concussion.

Types of outcome measures

The outcomes of interest for this review included concussion symptom severity assessed using validated questionnaire-based scales, menstrual cycle phase assessment using self-reported last menstrual period and menstrual history, circulating hormonal levels (e.g., progesterone and oestradiol) measured through serum blood sampling, and gait parameters evaluated using walkway systems and wearable sensors.

2.2.2 Information sources

The following electronic databases were searched from June to August 2025: CINAHL, SPORTDiscus, MEDLINE, Academic Search Complete (via EBSCOhost), and Scopus. The search strategy was developed based on the key concepts derived from the research questions, including menstrual cycle/hormonal changes, female population, gait, secondary injury risk, concussion risk, concussion symptoms, and post-concussion

outcomes. Grey literature was also considered during the search process; however, no grey-literature sources met the predefined eligibility criteria and therefore none were included in the final review.

2.2.3 Search strategies

Multiple search combinations were used to address the study objectives, with keywords and their synonyms combined for each database. Boolean operators (AND/OR) were used to combine search terms and ensure a broad yet targeted retrieval of studies.

For question one, the search included terms related to menstrual cycle phases and hormonal fluctuations (e.g., menstrual cycle, menstrual phase, hormonal fluctuation, oestrogen, progesterone, follicular phase, luteal phase, ovulatory), secondary injury outcomes (e.g., reinjury, recurrence, subsequent injury), lower-limb injuries (e.g., knee, ankle, ACL), female populations, and post-concussion conditions. For question two, the search strategy incorporated terms related to gait and locomotion (e.g., spatiotemporal parameters, kinematic, kinetic, walking, stride length, cadence, gait speed, balance, postural control), secondary injury outcomes, female populations, and post-concussion conditions. For question three, all relevant concepts from questions one and two were combined to examine the relationship between menstrual cycle-related hormonal changes and gait alterations, and their potential implications for secondary injury outcomes following concussion in females.

2.2.4 Data extraction

Data collection process

Only full-text, peer-reviewed articles published in the English language were included in this review. The English-language restriction was applied to ensure that the included studies could be accurately assessed and interpreted by the researcher. The study selection process involved manual screening of titles and abstracts, followed by full-text review. All screening and full-text assessments were conducted by the researcher (SM), with the supervisors (PH, NS, AZ) consulted where clarification was required to determine final inclusion.

Prior to full-text screening, duplicate records were identified and removed using EndNote reference management software. The remaining studies were then assessed by SM against the pre-established inclusion and exclusion criteria to ensure relevance to the research questions. As female-only studies examining gait alterations following concussion were limited, studies in which females comprised more than 50% of the study population were also considered for inclusion, provided that gait related data for the female participants were reported separately, in accordance with the predefined inclusion criteria. Where eligibility was uncertain, this was discussed with the supervisory team and resolved by consensus. Only studies meeting all eligibility criteria were included in the final review.

Data from the included studies were extracted into Microsoft Excel spreadsheets. The extraction process was conducted by the researcher (SM), and the extracted data were subsequently reviewed by the supervisors (PH, NS, AZ) for accuracy and consistency. Where discrepancies or uncertainties were identified during the review of the extracted data, these were discussed and resolved through consensus. Relevant information was recorded, including study title, author(s), year of publication, population characteristics, age, menstrual cycle phase in relation to concussion and symptom severity, characteristics of concussion symptoms, gait/walking

parameters, type of injuries, outcome measures, and key findings. In addition, study limitations and research gaps were systematically documented during the extraction process.

Data synthesis

A narrative synthesis approach was considered the most appropriate method for this scoping review due to the heterogeneity in study designs, inconsistencies in data reporting, and variation in study populations. This method enables the integration and interpretation of findings from studies with diverse methodologies and characteristics. The synthesis was conducted in accordance with the framework proposed by Popay et al. (2006), with studies grouped based on the available evidence to address the research questions. Where possible, findings from studies examining different menstrual-cycle phases, hormonal variations in relation to post-concussion outcomes and factors potentially relevant to secondary injury risk, and walking parameters, including gait alterations with potential relevant to secondary injury, were compared.

2.3 Findings

2.3.1 Study design and characteristics

A total of 754 studies were initially identified through the database searches (Figure 3). Studies were screened using a checklist developed based on the inclusion and exclusion criteria. After removing duplicates and irrelevant records, 405 studies remained for title and abstract screening. Following this stage, 16 studies were selected for full-text review. Of these, five studies were excluded as they did not specifically focus on menstrual cycle phases and/or examined primary injury rather than secondary injury outcomes. Ultimately, 11 studies met the inclusion criteria and were included in the final review. Study characteristics relating to menstrual cycle phases, related hormonal changes, and post-concussion outcomes in females following concussion are summarised in Table 1. Study characteristics relating to gait alterations and their potential implications for secondary injury in females following concussion are summarised in Table 2.

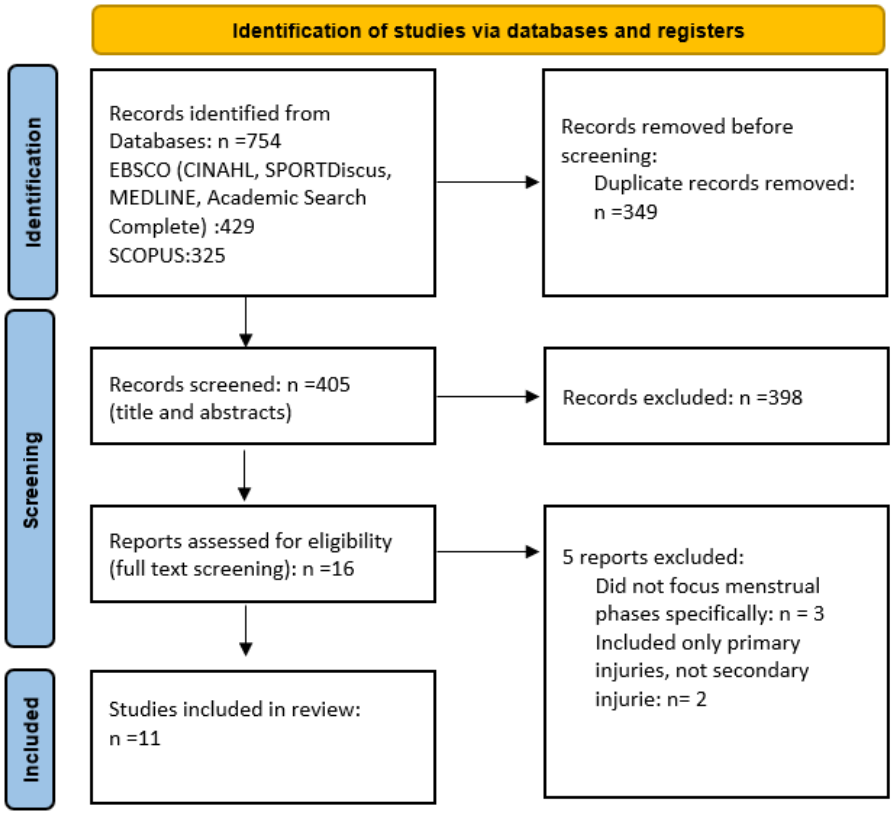

Figure 3 PRISMA-ScR flow diagram of study selection.

This scoping review aimed to explore the relationship between menstrual cycle-related hormonal changes, gait alterations, and post-concussion outcomes in females following concussion, including evidence regarding their potential implications for secondary musculoskeletal injury and recurrent concussion. This review aimed to address three key objectives while identifying gaps in the current literature related to menstrual cycle dynamics and gait-related injury outcomes in females following concussion.

First, it sought to examine how menstrual cycle-related hormonal changes influence post-concussion symptom severity in females following concussion. Second, it aimed to identify the gait alterations that occur in females following concussion. Third, it sought to determine what evidence exists regarding the relationship between menstrual cycle-related hormonal changes and gait alterations in relation to secondary musculoskeletal injury and recurrent concussion in females following concussion.

Regarding menstrual cycle assessment, some studies focused on the menstrual phase at the time of injury (Roby et al., 2023; Wunderle et al., 2014) while others examined circulating hormone levels such as progesterone and oestradiol (Chen et al., 2021; Goeckner et al., 2025; Ott et al., 2024). Menstrual cycle phases were determined using self-reported last menstrual period and/or serum hormonal measurements.

A total of 1,930 participants were included across 11 independent datasets, with more than 1,650 identified as female. Studies exclusively focusing on female populations in relation to gait alterations and secondary injury risk following concussion remain limited. Therefore, studies were included if females comprised more than 50% of the study population, due to the scarcity of female-specific research in this area over the past decade.

Three studies did not report the exact number of female participants, although they indicated the proportion of females within the total sample (David et al., 2020; Howell et al., 2020; Shumski et al., 2024). Across all studies, participant ages ranged from 13 to 60 years. All included studies focused on naturally menstruating individuals with regular menstrual cycles.

Follow-up durations varied considerably across studies, ranging from immediate post-injury assessment (within 24 hours) to return-to-play assessments within 7 days, to one-month follow-up evaluations and extending to 3–4 months post-injury (Goeckner et al., 2025; Ott et al., 2024; Roby et al., 2023). However, direct evidence relating menstrual cycle-related hormonal changes to secondary musculoskeletal injury risk was limited and no studies meeting the inclusion criteria directly addressed this relationship.

Most menstrual cycle studies primarily examined post-concussion symptom presentation, symptom severity, and quality of life outcomes (Ott et al., 2024; Roby et al., 2023). Commonly used outcome measures included the Post-Concussion Symptom Inventory (PCSI), Brief Symptom Inventory–Global Severity Index, Perceived Stress Scale (PSS), Immediate Post-Concussion Assessment and Cognitive Testing (ImPACT), and EuroQol, which were used to assess symptom severity and quality of life.

In contrast, gait-related studies focused on differences between single-task (only focusing on walking) and dual-task walking (walking while doing another task at the same time) conditions in concussed populations (Howell et al., 2020; Jain et al., 2024; Lowe et al., 2023; Messerschmidt et al., 2021). These studies evaluated dual-task cost (motor and cognitive) alongside spatiotemporal gait parameters such as gait speed, cadence, step length, step width, swing phase, and double support time. Additionally, some studies applied cross-recurrence quantification analysis to examine gait dynamics, including percent determinism, average diagonal line length, laminarity, and trapping time under both single- and dual-task conditions (David et al., 2020). Additionally, one study specifically investigated the association between cognitive function and tandem gait performance (Shumski et al., 2024).

Gait assessment methods varied across studies and included tools such as the GAITRite® walkway system (14′ × 2′; CIR Systems), tandem gait timing, and wearable inertial sensors (Opal sensors; APDM Inc., Portland, OR). Additional measures included self-reported concussion history and the Immediate Post-Concussion Assessment and Cognitive Testing (ImPACT) to evaluate symptom severity and cognitive function. However direct evidence linking gait alterations and secondary musculoskeletal injury risk and recurrent concussion remains limited, and no studies meeting the inclusion criteria directly address the relationships.

Table 1 Study characteristics of menstrual cycle phases, hormonal changes, and post-concussion outcomes in females following concussion.

Author (Year)	Study design; Focus area	Population; Sample size; Age	Menstrual cycle / hormones	Menstrual Phase calculation method	Key outcomes; Outcome measure tools	Assessment time	Main findings
Wunderle et al. (2014)	Nested cohort; Concussion outcomes	Concussed females; n=144; 16-60	Follicular vs luteal vs synthetic progesterone	Serum progesterone concentration	QoL, symptoms; EuroQoL, Rivermead Post Concussion Questionnaire	One month after enrolment	Luteal phase → poorer outcomes; supports progesterone withdrawal hypothesis
Chen et al. (2021)	Preliminary study; Hormones and brain function, post-injury symptoms presentation	Female athletes; n=32; 18-22	Progesterone levels	Serum hormone level	rCBF, stress; PSS	3-10 d post injury	Higher progesterone → improved stress outcomes and cerebral blood flow
Roby et al. (2023)	Prospective cohort; Concussion & menstrual cycle pattern changes and symptom recovery	Concussed females; n=512; 13-18	Cycle phase at injury	calculated using self reported last menstrual date	Symptom burden & type of menstrual cycle; PCSI, self reported MC changes	<28 days post injury & 3-4 month post injury	No significant MC changes associated with MP at injury; trend toward higher symptoms in menstruation/early luteal
Ott et al. (2024)	Observational; Hormones and symptoms	Adolescent female athletes; N=38; 14-18	Luteal vs follicular	serum hormone level	Symptom severity; PCSS, ImPACT	one week, one-month post-injury	Luteal phase → worse symptoms & neurocognitive scores; no effect on recovery timeline
Goeckner et al. (2025)	Prospective cohort; Hormones and recovery	Female athletes/ cadets; n=749; 19	Progesterone, Oestradiol	serum hormone concentration	Psychological distress; Brief Symptom Inventory Global Severity Index scores	>24hrs immediate admission, 7d of RTP	No recovery link; higher oestradiol → increased depressive symptoms

Note: MP = menstrual phase; QoL = quality of life; rCBF = regional cerebral blood flow; PSS = Perceived Stress Scale; PCSI = Post-Concussion Symptom Inventory; PCSS = Post-Concussion Symptom Scale; ImPACT = Immediate Post-Concussion Assessment and Cognitive Testing; RTP = return to play; n = sample size; MC = menstrual cycle; LH = luteinizing hormone; FSH = follicle-stimulating hormone.

Table 2 Study characteristics of gait alterations and their potential implications for secondary injury in females following concussion.

Author (Year)	Study design; Focus area	Population; Sample size (F/M); Age	Key outcomes; Outcome measures tools	Main findings
David et al. (2020)	Cohort (CRQA); Gait & injury risk	SRC; n=8 (63%/37%); 12-18, Non-SRC; n=24 (46%/54%), control; n=15 (53%/47%)	DT stability (diagonal line length, trapping time, percent determinism, and laminarity); Opal Sensor	Altered dual-task gait (slower speed, increased diagonal line length, and higher trapping time) is associated with increased risk of subsequent musculoskeletal injury.
Howell et al. (2020)	Cohort; Gait stability	concussed; n=43 (56%/44%); 12-17, healthy; n=38 (55%/45%)	diagonal line length, trapping time, percent determinism, and laminarity in ST & DT; Opal Sensor	Concussion athletes showed higher gait determinism, laminarity, and trapping time, with no additional dual-task effect.
Messerschmidt et al. (2021)	Cross-sectional; Sex differences in gait	concussed athletes; n=98 (44/54); 18-19	Gait parameters; GAITRite + ImPACT	Females demonstrated higher DTC of gait speed, cadence, % of swing phase & %of double support phase
Lowe et al. (2023)	Cohort; Single & dual-task gait	concussed athletes; n=29 (12/17); 14-18	DTC & spatiotemporal parameters; GAITRite walkway	Females showed higher dual-task cost in acute & chronic stages than males
Jain et al. (2024)	Prospective cohort; Neuromotor control deficits during dual & single walking	concussed adolescents; n=15 (13/2); 16-18, uninjured; n=17 (10/7); 17-19	cadence, gait speed, step duration & muscle activation; Opal sensors+ EMG	Both groups displayed worse performance on gait metrics during DT & Altered neuromotor patterns in concussed group
Shumski et al. (2024)	Cross-sectional; Cognition & tandem gait	concussed athletes; n=126 (56% /44);18<, healthy athletes; n=42 (40.5%/59.5%); 18<	Dual-task cost, gait time, motor speed, reaction time; Stopwatch	Concussion history is associated with altered dual-task gait–cognition relationships and improved dual-task cost response rate, with no other significant group differences in cognitive or tandem gait outcomes.

Note. SRC = sport-related concussion; F = female; M = male; CRQA = cross-recurrence quantification analysis; DT = dual-task; ST = single-task; DTC = dual-task cost; GAITRite = GAITRite® walkway system; ImPACT = Immediate Post-Concussion Assessment and Cognitive Testing; EMG = electromyography.

2.3.2 Hormonal influences on post-concussion outcomes in females

Influence of menstrual cycle phase and hormones on post-concussion symptoms and recovery trajectories

Across the included literature, five out of eleven studies examined menstrual cycle–related factors, with only two directly investigating the role of menstrual cycle phase on concussion symptom severity following mTBI (Roby et al., 2023; Wunderle et al., 2014). Findings from the included studies suggests that the menstrual cycle phase at the time of injury may influence post-concussion outcomes; however, findings are inconsistent.

Wunderle et al. (2014) reported that individuals injured during the luteal phase experienced significantly poorer quality-of-life outcomes compared to those injured during the follicular phase, supporting the notion of hormonally mediated vulnerability. In contrast, Roby et al. (2023) did not identify a clear association between menstrual cycle phase and post-injury menstrual changes, although they did observe a trend toward increased symptom burden in individuals injured during menstruation or the early luteal phase. These mixed findings suggest that while the menstrual phase may influence symptom severity, the relationship is not well established across studies.

One potential explanation for these findings is the progesterone withdrawal hypothesis proposed by Wunderle et al. (2014), which suggests that a rapid decline in progesterone levels following concussion may exacerbate post-concussion symptoms. The authors found that individuals using synthetic progesterone (oral contraceptives) reported fewer symptoms overall than those not using hormonal contraceptives. However, the strength of this conclusion is limited by potential confounding factors, as the hormonal contraceptive group was not intended to represent a specific intervention for head injury and the observational nature of the study. Nevertheless, the findings provide preliminary support for the progesterone withdrawal hypothesis and highlight potential differences in symptom reporting associated with hormonal status.

Methodological differences across studies further limit the comparability of findings. While Wunderle et al. (2014) employed objective hormonal measurements taken within hours of injury, Roby et al. (2023) relied on self-reported menstrual cycle data, which may be subject to recall bias and misclassification. Additionally, small sample sizes and variability in participant characteristics, such as age and activity level, reduce the generalisability of results. The lack of statistically significant findings after adjustment for multiple comparisons in Roby et al. (2023) further weakens the evidence base.

Inconsistencies are also evident in the reported timing of concussion across menstrual cycle phases. Wunderle et al. (2014) reported a higher incidence of concussion during the late luteal phase, whereas Roby et al. (2023) observed a greater proportion of injuries occurring during the menstruation or early luteal phase . These conflicting findings may reflect differences in study design, sample characteristics, and methods used to determine menstrual cycle phase, rather than true physiological variation. Overall, while there is emerging evidence to suggest an association between menstrual cycle phase and concussion symptom severity, the current literature remains inconclusive due to methodological limitations and inconsistent findings. This represents a critical gap in the literature, particularly in understanding secondary MSK injury risk beyond concussion recurrence.

Neuroprotective effects of sex hormones following concussion

A small but growing body of literature has explored hormonal responses following concussion, with three key studies investigating the role of sex hormones in influencing symptom severity and recovery trajectories (Chen et al., 2021; Goeckner et al., 2025; Ott et al., 2024). Collectively, these studies suggest that fluctuations in ovarian hormones across the menstrual cycle may affect concussion outcomes. However, the evidence remains inconsistent and methodologically varied, limiting the strength of conclusions that can be drawn.

Progesterone has been proposed to play a neuroprotective role due to its influence on connective tissue properties, including fibroblast activity and collagen synthesis, as well as its widespread effects within the central nervous system. Chen et al. (2021) reported that higher progesterone levels were associated with more favourable stress responses and increased regional cerebral blood flow, suggesting a potential protective mechanism mediated through cerebral perfusion. While these findings support a beneficial role of progesterone, they are based on preliminary data and therefore require cautious interpretation.

In contrast, Ott et al. (2024) reported that higher progesterone levels during the luteal phase were associated with increased symptom severity and poorer neurocognitive performance in adolescent female athletes following sport-related concussion. Specifically, deficits were observed across multiple domains, including verbal memory, visual memory, reaction time, and symptom clusters measured by the Post-Concussion Symptom Scale. These findings challenge the notion of a uniformly protective effect of progesterone and instead suggest that its impact may vary depending on the timing within the menstrual cycle or the acute phase of injury. Despite these differences, Ott et al. (2024) also found no significant association between menstrual cycle phase and overall recovery timelines, with similar neurocognitive outcomes observed between luteal and follicular phases at follow-up. This indicates that hormonal influences may be more relevant in the acute phase of concussion rather than in long-term recovery. Similarly, Goeckner et al. (2025), in a large prospective cohort study, reported no significant association between hormone levels and recovery outcomes, further weakening the evidence for a direct hormonal effect on recovery trajectories. This inconsistency highlights the complexity of hormonal responses following concussion and the need for more precise hormonal monitoring (Goeckner et al., 2025).

Oestrogen has also been widely recognised for its neuroprotective properties, including reducing inflammation, limiting oxidative stress, and promoting neuronal repair through receptor-mediated mechanisms. However, much of this evidence is derived from preclinical studies, and similar benefits have not been consistently demonstrated in human populations. Supporting this, Goeckner et al. (2025) found that increased oestradiol levels were associated with greater psychological distress, particularly depressive symptoms, rather than improved recovery outcomes. This suggests that hormonal influences may be more closely linked to psychosocial responses than to physiological recovery.

Overall, while there is some evidence to suggest that hormonal fluctuations across the menstrual cycle may influence concussion symptom severity, the current literature is characterised by conflicting findings, methodological limitations, and variability in study design. Importantly, there is a lack of consistent evidence demonstrating a clear relationship between hormone levels and recovery outcomes. This represents a

significant gap in the literature and highlights the need for future research to better understand the interaction between hormonal fluctuations, neuromuscular control, and injury risk in female populations.

2.3.3 Dual-task gait deficits as indicators of secondary injury risk in post-concussion females

Six of the eleven included studies investigated gait alterations in females following concussion, with findings providing evidence relevant to the potential implications of gait alterations for secondary musculoskeletal injury and recurrent concussion (David et al., 2020; Jain et al., 2024; Lowe et al., 2023; Messerschmidt et al., 2021; Shumski et al., 2024). Although walking is generally considered a low-impact and safe form of physical activity with a relatively low injury risk (Reiner et al., 2013), concussion can significantly disrupt normal gait function. In the acute phase, individuals commonly report symptoms such as headache, dizziness, light sensitivity, impaired cognition, slower reaction time, and deficits in both static and dynamic balance (Buckley et al., 2016). Even when these symptoms appear to resolve clinically, impairments in postural control and gait may persist for several weeks or even years after injury (Howell et al., 2014). These persistent neuromotor deficits are clinically important because they may increase vulnerability to both recurrent concussion and secondary musculoskeletal injuries.

Research specifically examining gait-related injury risk in female populations remains limited; however, studies with female-majority samples provide important insight into sex-specific recovery patterns. In the included studies, females represented more than 50% of participants across both concussed and control groups, including 56.3% of athletes (Shumski et al., 2024), 56% (Howell et al., 2020), 63% (David et al., 2020), and 71.9% of participants (Jain et al., 2024). In addition, two studies reported findings specifically focused on female participants despite having a female representation of less than 50% in the overall sample, including 44.9% of varsity athletes (Messerschmidt et al., 2021), and 41.4% of athletes (Lowe et al., 2023). Across these studies, females consistently demonstrated a greater number of gait alterations during dual-task walking compared to single-task walking, particularly when compared with non-concussed controls. Female participants also appeared to show greater variability in symptom presentation than males, suggesting that recovery patterns may be more complex and prolonged.

A key strength of this body of literature is the use of dual-task gait assessment, which is reported to be more sensitive than single-task walking in identifying subtle post-concussion deficits (Howell et al., 2020; Jain et al., 2024). Studies used different walking conditions, including normal walking under single- and dual-task conditions and tandem gait task, to evaluate gait performance (Howell et al., 2020; Jain et al., 2024; Shumski et al., 2024). These assessments were supported by objective biomechanical tools such as Opal sensors placed at the lumbosacral junction and on the dorsum of each foot to assess gait dynamics (Howell et al., 2020), wireless electromyography (EMG) sensors placed bilaterally on six lower limb muscles to examine muscle activation patterns (Jain et al., 2024), and the GAITRite® portable walkway to assess spatiotemporal gait parameters during self-selected walking speed (Howell et al., 2020).

Several studies reported that dual-task gait instability may be an important predictor of future injury risk. Howell et al. (2020) found that concussed individuals walked significantly slower ($p = 0.02$) and demonstrated higher diagonal line length ($p = 0.006$) and trapping time ($p = 0.006$) compared with uninjured

controls, indicating reduced gait adaptability and impaired motor control. Similarly, David et al. (2020) reported significantly higher percent determinism (F = 4.70, *P* = .033, ηp2 = 0.058), laminarity (F = 9.08, *P* = .004, ηp2 = 0.105), and trapping time (F = 7.04, *P* = .01, ηp2 = 0.084) during both single- and dual-task walking conditions in the concussed group, highlighting altered gait stability even in the acute stage of concussion, approximately seven days post-injury. Importantly, Howell et al. (2020) further showed that dual-task gait variables such as diagonal line length (HR = 1.95, 95% CI: 1.05–3.60), trapping time (HR = 1.66, 95% CI: 1.09–2.52), and walking speed (HR = 0.01, 95% CI: 0.00–0.51) were significantly associated with increased risk of subsequent musculoskeletal injury. These findings suggest that persistent gait instability may remain even after clinical symptoms improve and may serve as an important marker for return-to-play decision making.

Sex differences were particularly evident in studies examining dual-task cost (DTC). Lowe et al. (2023) found that females demonstrated increased DTC across multiple spatiotemporal gait parameters, including normalized velocity, step length, and single- and double-limb support phases. Approximately 83% of females in the acute stage and 29% in the chronic stage showed elevated DTC, whereas male participants showed more limited changes, mainly in double-limb support. Similarly, Messerschmidt et al. (2021) reported that females showed greater reductions in gait speed, cadence, and swing phase, along with greater increases in double-support time when progressing from single-task to dual-task walking. Females demonstrated significantly higher DTC in gait speed (17.15% vs 11.85%; p = .005), cadence (9.72% vs 5.77%; p = .002), percentage of swing phase (3.39% vs 2.17%; p = .019), and double-support phase (−11.63% vs −7.51%; p = .018) compared with males. These findings indicate that females may experience prolonged neuromotor inefficiency and reduced postural stability, which may partly explain their increased susceptibility to secondary musculoskeletal injury after concussion.

Despite the overall trend toward impaired dual-task gait performance following concussion, findings across the literature are not entirely consistent. While most studies reported clear differences between concussed and uninjured individuals during dual-task gait assessment, Jain et al. (2024) found that both concussed and uninjured groups demonstrated similar reductions in gait performance under dual-task conditions compared to single-task walking (p < .001), with no significant differences between groups. This suggests that although neuromotor inefficiencies may persist beyond two weeks post-injury, they may not always be reflected in measurable gait performance. Similarly, Shumski et al. (2024) found that although the concussed group showed a greater number of significant correlations between cognitive performance and dual-task gait time, athletes with a history of concussion demonstrated a better dual-task cost response rate (p = 0.01), which may reflect compensatory movement strategies rather than complete recovery. This suggests that individuals may consciously or unconsciously adapt their movement patterns, such as by prioritising gait stability or allocating greater cognitive resources to the walking task, enabling them to maintain performance despite the persistence of underlying neuromotor deficits. This highlights the complexity of interpreting gait outcomes and suggests that improved performance does not always indicate full neuromotor restoration.

The variation in findings across studies should also be considered in the context of methodological differences. Studies that reported significant dual-task gait impairments and associations with subsequent

injury risk generally employed prospective designs and objective outcome measures, which may increase confidence in the reliability of their findings (David et al., 2020; Howell et al., 2020). In contrast, studies reporting less pronounced group differences used different assessment protocols, recovery timeframes, and participant characteristics, which may have influenced the detection of subtle gait deficits (Jain et al., 2024; Shumski et al., 2024). Consequently, although the evidence is not entirely consistent, findings from studies with more rigorous methodological approaches may provide a more reliable basis for interpreting the presence of persistent neuromotor alterations following concussion and their potential contribution to secondary injury risk.

Overall, the evidence suggests that concussed females demonstrate more pronounced alterations in spatiotemporal gait parameters, including reduced walking speed, altered cadence, decreased stability, and changes in swing and double-support phases, particularly during dual-task walking conditions. Dual-task gait assessments appear to be more sensitive than single-task walking for identifying persistent post-concussion deficits and may help explain the increased risk of secondary musculoskeletal injury and repeated concussion observed in female. These findings support the importance of incorporating objective gait assessment into post-concussion evaluation and rehabilitation, particularly when making return-to-sport decisions in female populations.

2.3.4 Influence of menstrual cycle-related hormonal changes and gait alterations on secondary injury risk and recurrent concussion in females following concussion

Despite growing interest in sex-specific factors affecting concussion outcomes, no studies have directly examined the combined influence of menstrual cycle-related hormonal changes, gait alterations on the risk of secondary musculoskeletal (MSK) injury and recurrent concussion in females following concussion. A broader review of the literature identified studies investigating the effects of menstrual cycle phases on various aspects of physical performance in healthy females and athletes. Menstrual cycle related differences have been reported in isometric and isotonic muscle strength (Pournasiri et al., 2023) aerobic capacity and VO_2 responses (Ekberg et al., 2024), fatigue and performance outcomes (Domínguez-Muñoz et al., 2023), and physiological recovery measures such as heart rate recovery and blood lactate clearance (Benito et al., 2023). Collectively, these findings indicate that hormonal fluctuations across the menstrual cycle can influence physiological and performance-related outcomes. However, the applicability of these findings to concussion populations remains uncertain, as existing studies have been conducted primarily in healthy or athletic females.

Hormonal changes across the menstrual cycle have been linked to alterations in ligament laxity, neuromuscular control, balance, and postural stability, all of which may affect gait mechanics and movement efficiency (Messerschmidt et al., 2021). Furthermore, fluctuations in ovarian hormones may influence the severity and duration of post-concussion symptoms, potentially affecting functional performance during recovery (Chen et al., 2021). These physiological and psychological responses suggest a plausible pathway through which menstrual cycle phase may influence recovery outcomes and susceptibility to subsequent injury following concussion.

This relationship may be particularly important given that gait impairments are commonly observed following concussion and have been identified as indicators of persistent neuromotor dysfunction and increased injury susceptibility (Howell et al., 2020). However, it remains unclear whether hormonal fluctuations across the menstrual cycle contribute to post-concussion gait alterations or increase the risk of secondary MSK injury. Consequently, the interaction between menstrual cycle phase, hormones, gait performance, and secondary injury risk following concussion remains poorly understood.

This represents an important gap in the literature. Investigating how menstrual cycle-related hormonal changes across different menstrual phases influence gait alterations, neuromotor control, and injury susceptibility following concussion may provide valuable insights into sex-specific recovery processes. Such knowledge could contribute to more individualized rehabilitation approaches, improve return-to-sport decision-making, and ultimately reduce the risk of secondary MSK injury in female athletes recovering from concussion.

2.3.5 Critical appraisal

The findings of this review highlighted the potential influence of menstrual cycle phases, hormones and gait alterations on concussion outcomes with limited evidence regarding their potential implications for secondary musculoskeletal injury and recurrent concussion. Only eleven studies met the inclusion criteria, highlighting the need for further high-quality research to better understand these relationships.

A major limitation across the included studies was the difficulty in accurately identifying menstrual cycle phase at the time of mTBI. Although some evidence suggests that elevated progesterone during the luteal phase may be associated with increased symptom severity following sport-related concussion, hormonal measurements were often collected after injury or within an imprecise timeframe, reducing confidence in determining true hormonal status at the point of injury (Ott et al., 2024). This limitation was further compounded by the heavy reliance on self-reported menstrual data. Self-reported cycle length is frequently overestimated, and adolescents are particularly vulnerable to inaccuracies due to irregular menstrual cycles following menarche (Ott et al., 2024; Roby et al., 2023). The absence of objective verification methods such as ovulation testing or hormonal assays increases the risk of menstrual phase misclassification and weakens the validity of findings.

Participant classification also introduced potential bias. Most studies relied on binary self-identification of sex, which may have resulted in participant misclassification or exclusion. In addition, contraceptive use was commonly recorded only at baseline and was not updated over time, even though contraceptive status may change significantly between baseline assessment and injury occurrence (Goeckner et al., 2025). This is particularly important because hormonal contraceptives may influence concussion outcomes and symptom presentation (Wunderle et al., 2014). While progesterone has been proposed to have neuroprotective effects, some studies suggest that females injured during the follicular phase may experience better recovery, whereas those injured during the luteal phase may experience worse outcomes due to post-injury hormonal withdrawal. However, these findings remain inconsistent because of methodological limitations in hormone assessment and timing (Chen et al., 2021; Goeckner et al., 2025).

Another important limitation was the inconsistency of outcome measures across studies. Some studies reported no significant differences in concussion outcomes across menstrual phases, while others suggested that oral contraceptive users experienced fewer symptoms overall (Wunderle et al., 2014). However, the use of broad and non-specific measures such as the Perceived Stress Scale (PSS) limits interpretation, as this tool measures general stress rather than concussion-specific symptom burden. Since stress levels naturally fluctuate even in healthy individuals, this introduces substantial confounding. Future studies would benefit from incorporating more objective and specific markers such as cortisol levels, neurocognitive testing, and emotion regulation tasks to better capture physiological and functional changes following concussion (Chen et al., 2021).

The role of ovarian hormones, particularly oestradiol, also remains underexplored. Most studies focused primarily on progesterone, despite oestradiol and progesterone interacting dynamically rather than functioning independently. This hormonal interplay may have significant implications for neuromotor control, symptom severity, and recovery following concussion. More frequent hormonal sampling across the menstrual cycle, along with improved timing of measurements, is necessary to better characterise these interactions and their clinical significance (Goeckner et al., 2025).

Small sample sizes were another major limitation. Several studies were underpowered or restricted their analyses to a single menstrual phase, most commonly the follicular phase, which limits interpretation across the full menstrual cycle and reduces the ability to examine phase-dependent differences. Larger cohorts are needed to evaluate combined hormonal effects and strengthen statistical validity (Chen et al., 2021). Confounding variables were also inconsistently controlled across studies. Factors such as injury mechanism, sport type, training load, sleep, nutrition, competition level, and contraceptive use may independently influence concussion outcomes and gait performance (Ott et al., 2024). Failure to adequately control for these variables increases uncertainty when interpreting associations between menstrual cycle phase, gait alterations, and injury risk.

Methodological inconsistencies were evident in studies examining gait alterations and secondary injury risk. Unequal group sizes based on concussion history, symptom severity, and sport participation reduced comparability between studies. Variability in dual-task cost (DTC) measures also limited consistency and reduced confidence in clinical interpretation (Messerschmidt et al., 2021). Furthermore, many studies relied on laboratory-based or high-cost equipment, such as advanced gait analysis systems and wearable sensors, which may not be practical for routine clinical use and therefore limit translation into real-world rehabilitation settings (Lowe et al., 2023).

Ecological validity represents another important limitation. Most gait assessments were conducted in highly controlled laboratory environments that may not accurately reflect the complex cognitive-motor demands experienced during sport participation or daily activities. Although deficits such as reduced cadence and altered gait stability were observed during dual-task walking, it remains unclear how these findings translate to real-world situations such as competition, navigating crowded environments, or rapid decision-making during sport (Jain et al., 2024).

Prior concussion history was insufficiently addressed in several studies. Since many participants reported a history of previous concussions, cumulative neurological effects may have influenced neuromuscular outcomes and gait performance. Future studies should incorporate objective neuromuscular assessment tools such as surface electromyography (sEMG) and inertial sensors to better understand the long-term effects of repeated injury exposure (Jain et al., 2024). Similarly, reliance on self-reported concussion and injury history introduces recall bias, while the absence of musculoskeletal injury history limits interpretation because previous injuries may independently affect gait mechanics and motor control (Howell et al., 2020; Shumski et al., 2024).

Variability in recovery stage and symptom persistence contributed to heterogeneity across findings. Some studies included participants with prolonged recovery times compared to typical concussion trajectories, which may reduce generalisability to broader athletic populations (Howell et al., 2020). Although clinically feasible measures such as tandem gait and dual-task assessments were used in some studies, more sensitive gait assessment tools may be required to detect subtle or persistent deficits following concussion (Shumski et al., 2024). The absence of additional balance measures such as the Balance Error Scoring System (BESS) further limits clinical interpretation, and findings may not be generalisable beyond adolescent populations.

Despite these limitations, the current literature demonstrated several notable strengths. Many studies employed prospective or cohort study designs, allowing researchers to examine concussion outcomes, recovery trajectories, and injury risk over time (Howell et al., 2020; Lowe et al., 2023; Shumski et al., 2024). Several studies also incorporated objective assessment methods, including hormonal assays, gait analysis systems, wearable sensors, and dual-task paradigms, which provided valuable insights into the physiological and neuromotor consequences of concussion (Lowe et al., 2023; Messerschmidt et al., 2021; Wunderle et al., 2014). Furthermore, the inclusion of both hormonal and neuromotor factors has contributed to a more comprehensive understanding of the multifactorial mechanisms that may influence concussion outcomes and secondary injury risk in females. Collectively, these methodological strengths provide an important foundation for future research in this area.

2.3.6 Future directions

Based on these limitations, several recommendations can be made for future research in this area. Large multi-centre cohort studies should be prioritised to improve statistical power and enhance generalisability. Objective methods of menstrual cycle tracking, including ovulation testing, basal body temperature monitoring, digital cycle-tracking tools such as Fitr Woman, and salivary or serum hormonal assays, should replace reliance on self-reported menstrual history to improve phase classification accuracy (Chen et al., 2021; Ott et al., 2024). There is also a clear need to address the underrepresentation of females in concussion research by recruiting larger samples of naturally menstruating females and conducting longitudinal assessments across multiple menstrual phases. This would allow better understanding of within-cycle variability and support the development of sex-specific return-to-play (RTP) guidelines that account for hormonal influences rather than relying on standardised protocols designed primarily from male-based research.

Future studies should also rigorously control for confounding variables such as sport type, training load, sleep, nutrition, contraceptive use, and previous injury history while extending follow-up periods to examine both acute and long-term recovery trajectories (Ott et al., 2024). Additionally, research should directly investigate the relationship between menstrual cycle phases, hormones, concussion outcomes, and secondary musculoskeletal injuries, including whether injury patterns vary across menstrual phases, as current evidence remains limited.

Overall, addressing these methodological and conceptual limitations will strengthen the evidence base, improve clinical application, and support the development of more individualised, sex-specific approaches to concussion management, rehabilitation, and injury prevention in female athletes.

2.4 Conclusions

In conclusion, this scoping review highlighted that menstrual cycle phases and associated hormonal changes may influence concussion outcomes in females, including symptom severity, neurocognitive function, psychosocial responses, recovery trajectories, and gait performance. Variations in oestrogen and progesterone, particularly across the follicular and luteal phases, may contribute to differences in post-concussion presentation, with some studies suggesting worse outcomes during the luteal phase and supporting the progesterone withdrawal hypothesis. Concussed females demonstrated greater alterations in spatiotemporal gait parameters, particularly during dual-task conditions, including reduced walking speed, altered cadence, decreased stability, and changes in swing and double-support phases, which may increase the risk of secondary musculoskeletal injury and recurrent concussion. However, the interaction between menstrual cycle phase, associated hormonal fluctuations, gait alterations, and concussion outcomes remains unclear. It is currently uncertain whether and how menstrual cycle-related hormonal changes influence gait alterations following concussion and whether this interaction contributes to secondary musculoskeletal injury or recurrent concussion risk in females. Methodological limitations such as small sample sizes, inconsistent outcome measures, reliance on self-reported menstrual data, and limited objective hormonal assessment further reduced the strength of current evidence. Future research should prioritise large longitudinal studies using accurate menstrual cycle tracking, objective hormonal profiling, and comprehensive gait assessment to better understand sex-specific concussion recovery and support more individualised rehabilitation and return-to-play strategies for female athletes.

PRELUDE TO CHAPTER 3

Following the literature presented in Chapter 2, which explored the influence of menstrual cycle phases, hormonal fluctuations, and gait parameter alterations on concussion outcomes and their potential implications for secondary musculoskeletal injury and recurrent concussion, Chapter 3 presents a prospective observational pilot case study among naturally menstruating healthy females. The aim of this study was to examine changes across three menstrual cycles and identify menstrual cycle-related hormonal changes and corresponding gait alterations across different menstrual phases.

This study is one of the first to investigate the relationship between menstrual cycle-related hormonal changes and gait alterations in females. It was specifically designed to improve understanding of how hormonal changes across the menstrual cycle relate to gait parameters in naturally menstruating females, providing preliminary evidence to inform future research in females following concussion.

Data collection was conducted between October 2025 and March 2026 to provide preliminary findings and direction for a larger-scale future study addressing the major research aims. This study focused on naturally menstruating females, with an aim to include both healthy participants and females within seven days post-concussion. However, due to time limitations and difficulty recruiting concussed naturally menstruating females within the required timeframe, only ten healthy naturally menstruating females were recruited.

For healthy participants, data were collected across three menstrual cycles, with assessments conducted during four phases of each cycle to identify hormonal changes and corresponding gait alterations. Each participant completed three walking tasks to assess gait alterations, while Plantiga insole sensors were used to measure spatiotemporal gait parameters.

Although this study did not provide definitive conclusions due to the small sample size and absence of concussed participants, it provided an important framework for future large-scale research. The findings may help identify patterns of hormonal variation and gait across the menstrual cycle, providing a foundation for future research investigating their potential implications for concussion recovery, secondary musculoskeletal injury, and recurrent concussion. Such research may ultimately contribute to the development of cycle-informed training and rehabilitation approaches to support injury prevention, recovery outcomes, and athletic performance in females.

CHAPTER 3: A PROSPECTIVE OBSERVATIONAL PILOT CASE STUDY - WALKING PATTERNS AND HORMONAL CYCLES IN HEALTHY FEMALES

Overview

Introduction: This study aimed to explore preliminary trends in the relationship between menstrual cycle-related hormonal changes and gait alterations in healthy females.

Design: A prospective observational pilot case study was conducted between 1 October 2025 and 15 March 2026.

Methods: Participants were followed up to three consecutive menstrual cycles over 12-week period. At each visit, they completed a series of questionnaires, performed three different walking tasks (single-task, dual-task, and head-turns), and underwent blood sample collection. Data were analysed according to sex, age, body mass index (BMI), sport participation, history of MSK injury or concussion, menstrual phase and cycle characteristics, hormonal levels, and gait parameters.

Results: The study included 10 healthy female participants with no history of musculoskeletal injury or concussion, and no new injuries were recorded during the data collection period. Hormone concentrations demonstrated the expected fluctuations across menstrual cycle phases, with significant phase-related changes observed in oestradiol, progesterone, LH, FSH, testosterone, TSH, and DHEA. However, no significant differences were found in gait parameters, cognitive dual-task costs, or motor dual-task costs across menstrual cycle phases under any walking condition. Overall, gait performance remained stable despite normal menstrual cycle-related hormonal fluctuations.

Conclusion: This pilot study showed that although hormonal concentrations varied significantly across the menstrual cycle, no significant differences in gait performance were detected across menstrual phases in healthy females. Given the small sample size and limited statistical power, these non-significant findings should be interpreted cautiously and should not necessarily be taken as evidence of no effect. Further research with larger cohorts, particularly in individuals with mild traumatic brain injury (mTBI), and females with different menstrual-cycle characteristics, is required to better investigate these relationships.

3.1 Introduction

Concussion is a mild traumatic brain injury caused by direct or indirect biomechanical forces to the head or body, resulting in temporary disruption of brain function and a range of physical, cognitive, emotional, and sleep-related symptoms, often without structural abnormalities detectable on standard neuroimaging (McGroarty et al., 2020). Research suggests female athletes are more susceptible to concussions, with prolonged symptoms and higher rates of secondary musculoskeletal (MSK) injuries compared to males (McGroarty et al., 2020). This vulnerability may be influenced by hormonal fluctuations and balance impairments, both of which impact recovery and elevate the likelihood of recurrent injuries.

Oestrogen and progesterone, hormones primarily linked to reproductive functions, also influence brain bioenergetics, hydration, metabolism, and thermoregulation (Rettberg et al., 2014). Women experience considerable hormonal shifts across different life stages, and fluctuations during the menstrual cycle (lasting 21–35 days) may occur concurrently with concussion symptoms, potentially influencing physical, cognitive, and emotional recovery. Concussions can disrupt menstrual cycles via hypothalamic pituitary ovarian (HPO) axis interference, potentially worsening neurological outcomes for those injured during high-progesterone phases like the luteal phase (Wunderle et al., 2014). A 2017 cohort study reinforced the importance of monitoring menstrual patterns post-concussion, as abnormal cycles can indicate potential health impacts and guide treatment adjustments (Snook et al., 2017).

Menstrual-cycle characteristics can also vary between individuals and across cycles, and menstrual phase alone may not fully represent an individual's hormonal or physiological state. Menstrual irregularity, polycystic ovary syndrome (PCOS), and hormonal contraceptive use can alter endogenous hormone profiles (Caliskan Guzelce et al., 2019; Hewett et al., 2007) may influence neuromuscular function, including muscle activation, postural control, joint stability, and coordination(Colenso-Semple et al., 2023; Elliott-Sale et al., 2021; McNulty et al., 2020). Therefore, these factors are important considerations when examining relationships between menstrual phase, hormonal concentrations, gait, and injury risk in female athletes.

Evidence also suggests that preovulatory phase is associated with MSK injury risk, particularly ligament injuries (Hewett et al., 2007). Female athletes in the preovulatory phase appear to face greater anterior cruciate ligament (ACL) injury risk. Additionally, oral contraceptive use has been linked to improved knee stability and reduced impact forces, suggesting a potential protective role when combined with neuromuscular training (Herzberg et al., 2017). A qualitative study further highlighted the prevalence of ligament sprains, dislocations, and strains across the menstrual cycle, emphasising the importance of cycle-informed training strategies to reduce injury risk (Chou et al., 2023).

Dynamic balance impairments have also been identified as a key contributor to concussion recurrence. For example, rugby players with poorer performance on the Y-Balance Test demonstrated a threefold increased risk of sports-related concussion, independent of prior concussion history (Johnston et al., 2019). Persistent postural control deficits following clinical recovery suggest ongoing neurophysiological impairment. Similarly, individuals with previous concussions have shown altered gait patterns, including increased double-leg stance time and reduced walking speed, which may elevate the risk of both recurrent concussion and MSK

injury due to reduced stability (Martini et al., 2011). These gait changes are particularly evident under dual-task conditions, where cognitive demands further challenge motor control (Chou et al., 2023).

Although neurocognitive deficits are often subtle in the acute phase of concussion recovery, impairments in concentration, working memory, immediate recall, and processing speed have been reported (Kevin et al., 2001). Longer-term concussion effects may also disrupt the HPO axis, influencing hormonal regulation, menstrual patterns, and physical performance. Such hormonal dysregulation may further contribute to injury susceptibility, highlighting the need for research exploring the interaction between endocrine function, neuromotor control, and injury risk (Lynall et al., 2020).

Despite the growing recognition that both hormonal fluctuations across the menstrual cycle and persistent gait alterations may influence injury risk in female athletes following concussion, limited research has examined how these factors interact in relation to secondary musculoskeletal injury and recurrent concussion risk. This lack of evidence highlights an important gap in understanding sex-specific mechanisms that may contribute to injury during recovery. Therefore, further investigation is needed to clarify these relationships and inform more targeted rehabilitation approaches. To address this gap, the present prospective observational pilot case study aimed to examine the associations between menstrual cycle-related hormonal changes and gait alterations in healthy females. Specifically, the study aimed to: (1) examine menstrual cycle-related hormonal changes across menstrual cycle phases; (2) identify gait alterations across the menstrual cycle phases; and (3) examine the relationship between menstrual cycle-related hormonal changes and gait alterations.

3.2 Ethical consent

Ethical approval for the study was reviewed and approved by the Southern Health and Disability Ethics Committees on 27 June 2025 (Reference: 2025 AM 21888) and locality approval was obtained from the Auckland University of Technology Ethical Committee (AUTEC) on 26 August 2025 for three years until 26 August 2028 (Reference 25/152). Informed consent was obtained from the participants before proceeding with the data collection.

3.3 Design

Participants

Participants included naturally menstruating females aged 18 years and older who provided informed consent to participate in this study. Initially, the study aimed to recruit 10 females with concussion (mTBI group) and 10 healthy control participants. However, due to the limited availability of naturally menstruating females with concussion within the study timeframe, only 10 healthy control participants were successfully recruited. Healthy participants were identified as naturally menstruating females based on their self-reported menstrual status.

Given the small sample size, this pilot study primarily focused on assessing feasibility, conducting descriptive analyses, and generating preliminary estimates rather than performing formal hypothesis testing. The findings from this study will inform the design and methodology of future large-scale studies. Participants

were eligible for inclusion if they were female, aged 18 years or older, naturally menstruating with regular menstrual cycles, residing in Auckland, New Zealand, and willing and able to attend study sessions and provide blood samples. Participants were excluded if they were younger than 18 years of age, unable to provide informed consent, pregnant, or using oral contraceptives or other hormonal medications that could affect menstrual regulation. Individuals with irregular menstrual cycles, severe neurological or other significant medical conditions, or musculoskeletal conditions that could limit gait or balance performance were also excluded. Recruitment was conducted through social media, posters displayed in healthcare settings such as general practice and physiotherapy clinics, concussion clinics, and word-of-mouth. Interested individuals contacted the research team and underwent eligibility screening prior to providing informed consent.

Procedures

Pre-session procedure

Participants who expressed interest in the study through the recruitment process were invited to have a brief discussion with the research team after reading the Participant Information Sheet (PIS). During this discussion, participants were given the opportunity to ask questions and clarify any concerns regarding the study procedures. Eligible participants were then provided with the date, time, and location of their first study visit. Participants were instructed to wear comfortable clothing that allowed access to the antecubital fossa (crook of the arm) for blood collection and to wear appropriate footwear suitable for walking assessments.

Baseline assessment

At the baseline visit, written informed consent was obtained by the researcher after explaining the study procedures and answering all participant questions to their satisfaction. Participants were provided with a signed copy of the consent form and the PIS. Following consent, each participant was assigned a unique sequential study identification (ID) number, which was used for all study documentation, sample labelling, and data storage to maintain confidentiality.

Demographic and health-related data were collected, including ethnicity, age, date of birth, sex at birth, gender, medical and surgical history, current medications, concomitant health conditions, highest educational qualification, and vital signs. Anthropometric measurements were obtained according to International Society for the Advancement of Kinanthropometry (ISAK) protocols. Body mass was measured using calibrated Seca scales, and standing height was measured using a stadiometer.

General health questionnaire and interview (GHQ)

A brief health interview was conducted by the researcher to identify current medications, previous medical diagnoses, and any new or previous musculoskeletal injuries or concussions. Participants also completed general health questionnaire during each visit. The interview required approximately 5 minutes, while the questionnaire took approximately 10 minutes to complete.

Menstrual cycle tracking and phase determination

Participants self-reported the date of their last menstrual period, the number of bleeding days, and usual menstrual cycle length. This information was used to estimate menstrual cycle phases, including menstruation, follicular phase, ovulation, and luteal phase using calendar-based method. All menstrual data were recorded and managed using REDCap software, which automatically calculated menstrual cycle phase based on a predefined 28–32-day cycle. Cycle phase determination was based on a calendar-based method, including the midpoint of the bleeding phase, defined as halfway through the reported bleeding window; the midpoint of the follicular phase, defined as halfway between the end of bleeding and estimated ovulation; estimated ovulation, calculated by counting backward 50% of the cycle length from the anticipated next menstrual period; and the midpoint of the luteal phase, defined as seven days after estimated ovulation (Schmalenberger et al., 2021; Wideman et al., 2013). Participants were informed of their next scheduled visit date and time accordingly. Menstrual-cycle phases were subsequently confirmed using the corresponding measured hormone concentrations, including oestradiol, progesterone, LH, and FSH, using the phase-specific reference ranges provided by the laboratory for the Abbott ARCHITECT analyser (Stricker et al., 2006). Where possible, all study visits were conducted at the same time of day to minimise potential circadian variation and reduce measurement bias; however, some sessions were rescheduled based on participant availability.

Gait assessment

Gait performance was assessed using Plantiga insole sensor pods containing inertial measurement units (IMUs) that recorded triaxial accelerometer and gyroscope signals at 416 Hz. The Plantiga processing platform automatically identified foot-ground interaction events, including initial contact and toe-off, from the recorded inertial signals and generated trial-level gait outcomes.

Before testing, participants were instructed to remove watches, mobile phones, and heavy outerwear to avoid interference with natural movement. Prior to data collection, appropriate footwear and correctly fitted Plantiga insoles were verified for each participant. To maintain consistency across testing sessions, participants wore the same footwear at all time points. A participant-specific account was created in the Plantiga mobile application using the participant's study ID, date of birth, and gender.

Participants completed gait assessments under three task conditions: single-task walking (ST), dual-task walking (DT), and head-turn walking (HT). In the single-task condition, participants walked at a self-paced speed. In the dual-task condition, participants walked while performing a cognitive task involving serial subtraction, specifically counting backward by threes from a randomly selected number provided by the researcher. For this purpose, 24 random numbers between 400 and 500 were generated using Microsoft Excel and selected via a lottery method, with different number sequences used at each visit to minimise familiarity bias.

In the head-turn condition, participants walked while alternately turning their head left and right in response to auditory cues delivered via a metronome mobile application set at 33 beats per minute to provide a slow, controlled pace for standardised head-turn movements. Rhythmic auditory cueing has been used in dual-task gait paradigms to standardise temporal structure and modulate attentional demand during locomotor

tasks (Hausdorff et al., 2008; Kline & Burdick, 1980; Thaut et al., 2007). For all conditions, participants walked in a straight line along a 7-metre walkway toward a cone, turned around, and returned to the starting point. They were instructed to begin and stop immediately following the researcher's verbal commands to ensure consistent timing and data collection.

Gait variables were derived from the recorded accelerometer and gyroscope signals using the Plantiga system's automated processing algorithms, which identify gait and foot-ground interaction events and calculate the corresponding gait measures. The selected variables were informed by gait measures examined in previous concussion research and were chosen to capture key aspects of walking, including temporal and performance characteristics, gait stability, mechanical loading and propulsion, and stride-to-stride gait consistency, to assess potential changes in gait across walking conditions and menstrual cycle phases (Fino et al., 2018; Lee et al., 2013; Mitchell & Cronin, 2023). Ground contact time (s) was defined as the duration between foot contact and toe-off, representing the time the foot remained in contact with the ground. Walking speed (m/s) represented the average forward walking speed during the walking trial. Double support ratio (%) represented the proportion of the gait cycle during which both feet were simultaneously in contact with the ground. Vertical impact (g) was defined as the peak acceleration in the world-vertical direction occurring during foot-ground contact and represented impact loading at landing. Vertical push-off (g) was defined as the peak acceleration in the world-vertical direction during the propulsion phase preceding toe-off. Swing duration variability (s) and stride length variability (m) represented stride-to-stride variability in swing duration and stride length, respectively, across successive gait cycles recorded during each walking trial. All gait variables were automatically derived from multiple gait cycles by the Plantiga processing algorithms and reported as trial-level outcomes for subsequent statistical analysis. The primary gait variables, demographic data and hormonal measures collected in this study are presented in Table 3.

Table 3 Summary of study variables collected.

Demographic/ Health data	**Gait variables**	**Hormonal measures**
Age	Ground contact time	Oestradiol
Sex	Walking speed	Testosterone
Height	Swing duration variability	Progesterone
Weight	Stride length variability	Luteinizing hormone (LH)
Menstrual cycle characteristics	Double support ratio	Follicle-stimulating hormone (FSH)
Cycle length	Vertical impact	Prolactin
General Health Questionnaire (GHQ) score	Vertical push-off	Thyroid-stimulating hormone (TSH)
	DTC-Cognitive	Free thyroxine (FT4)
	DTC-Motor	Total thyroxine (TT4)
		Cortisol
		Dehydroepiandrosterone sulphate (DHEA-S)

Note: DTC = dual-task cost; GHQ = General Health Questionnaire.

DTC-Cognitive represents the percentage change in cognitive task performance during dual-task conditions compared with single-task cognitive performance, thereby quantifying the impact of performing a concurrent motor task on cognitive performance. DTC-Motor represents the percentage change in gait performance during dual-task conditions compared with single-task walking performance, reflecting the cost of adding a cognitive task to motor performance. Dual-task cost was calculated for cognitive and motor performance as follows:

DTC-Cognitive (%) = (ST cognitive − DT cognitive) / ST cognitive × 100.

DTC-Motor (%) = (ST gait − DT gait) / ST gait × 100.

Blood sample collection

Blood samples were collected following gait assessment at AUT Millennium (AUTM) using standard venepuncture techniques. Participants were advised to maintain adequate hydration prior to blood collection and were given sufficient rest before sampling. Screening for allergies to latex, isopropyl alcohol, and adhesive materials was conducted prior to collection. Venous blood was drawn into 10 mL K2EDTA (dipotassium ethylenediaminetetraacetic acid) anticoagulant tubes, with a maximum total blood volume of 40 mL collected per participant.

Standard venepuncture procedure

Venepuncture was performed by suitably trained personnel using veins within the antecubital fossa (basilic, cephalic, or median cubital vein). After verbal confirmation, a tourniquet was applied approximately 10 cm above the selected site, and the skin was cleaned with 70% isopropyl alcohol.

The tourniquet was not applied for longer than one minute to minimise haemoconcentration. Following successful needle insertion and blood flow confirmation, the tourniquet was released, and blood was collected into the labelled tubes. After collection, participants were bandaged and instructed to avoid lifting heavy objects or strenuous arm activity for at least one hour. All blood specimens were labelled using the participant's study ID, collection date, and collection time. Tubes were gently inverted 8–10 times immediately after collection to ensure proper mixing with Ethylenediaminetetraacetic acid (EDTA).

Sample processing and storage

Blood samples were centrifuged within two hours of collection. If immediate processing was not possible, samples were refrigerated at 4°C for no longer than 24 hours. Samples were centrifuged at 2100 relative centrifugal force (RCF) for 10 minutes at 4°C. Plasma was carefully separated without disturbing the buffy coat and transferred into 1.5 mL Eppendorf tubes for long-term storage at −80°C. A maximum of two freeze–thaw cycles were permitted for each aliquot. All processed samples were stored at AUTM in accordance with laboratory health and safety protocols. All three components, including follow-up general health screening, gait assessment, and blood sample collection, were conducted at each study visit for all participants.

Hormonal analysis and assays

Before analysis, frozen plasma samples were thawed and brought to room temperature. Samples were pipetted into reagent cups and loaded onto the Abbott Architect ci4100 system, an integrated diagnostic analyser combining clinical chemistry and immunoassay testing. The Architect ci4100 system was used to quantify hormone concentrations for research purposes only. Calibration and quality control standards were supplied by Abbott Laboratories and performed by qualified laboratory technicians according to manufacturer guidelines. Calibration was conducted once per reagent lot or following maintenance of critical system components. The hormonal assay results were used to examine hormonal fluctuations across the menstrual cycle and their relationship with gait parameters in healthy participants.

3.4 Statistical analysis

Data were analysed using IBM SPSS Statistics (Version 31.0). Descriptive statistics were used to summarise participant demographics, hormonal concentrations, and gait parameters across menstrual-cycle phases. Continuous variables were reported using means, medians, standard deviations (SD), and ranges, as appropriate.

A linear mixed-effects model (LMM) was used to analyse hormonal concentrations and gait parameters while accounting for repeated measurements collected from the same participant across menstrual-cycle phases and menstrual cycles. Menstrual-cycle phase and menstrual cycle were included as fixed effects and repeated factors. Participant identification number was specified as the subject variable, and a compound symmetry covariance structure was selected to account for the correlation between repeated observations within participants. No additional random-effects structure was specified. Estimated marginal means (EMMs) were calculated to compare outcomes across menstrual-cycle phases and cycles, with Bonferroni-adjusted pairwise comparisons conducted where appropriate. Statistical significance was set at $p < 0.05$.

There were no missing values within completed study visits. However, participants contributed different numbers of complete menstrual cycles because not all participants were able to complete the planned three-cycle follow-up period. Four participants contributed one complete menstrual cycle, two participants contributed two complete cycles, and four participants contributed three complete cycles. No imputation of unavailable menstrual-cycle data was performed. All available observations were retained and analysed using the LMM, which accommodates repeated measurements and unequal numbers of observations across participants.

Prior to analysis, data were screened for normality, homogeneity of variance, and potential outliers using box plots and histograms. A small number of potential outliers were identified; however, these represented plausible physiological observations and were therefore retained. Model residuals were assessed using Normal Q-Q plots and plots of residuals versus fitted values. Residuals demonstrated acceptable agreement with expected normal distribution, with no substantial deviations from normality or evidence of heteroscedasticity observed. The repeated-measures structure accounted for the correlation between observations within participants, while participants were treated as independent observational units. Model fit was evaluated using Akaike's Information Criterion (AIC), corrected Akaike's Information Criterion (AICC), Bayesian Information

Criterion (BIC), and consistent Akaike's Information Criterion (CAIC), with lower values indicating better relative model fit.

3.5 Results

3.5.1 Description of the study sample

The study included 10 healthy female participants with no history of musculoskeletal injury or concussion. Participant characteristics are summarised in Table 4. Data were collected across multiple menstrual cycles depending on participant availability. Hormonal level data for one complete menstrual cycle were available for four participants (n = 4), while two participants (n = 2) completed two full menstrual cycles, and four participants (n = 4) completed three full menstrual cycles.

For gait assessment, 12 completed visits were available for seven participants (n = 7), eight completed visits were available for 1 participant (n = 1), and four completed visits were available for two participants (n = 2). The mean age of the participants was 28.5 ± 8.3 years (95% CI: 22.55–34.45; range: 20–42 years). Notably, no secondary injuries were recorded during the data collection period among the healthy female participants (n = 10, 100%).

Table 4 Characteristics of participants.

Variable	**Value (Healthy controls)**
Age (Years), mean ± SD	28.5 ± 8.3
Age range (min–max)	20-42
Ethnicity, n (%)	
NZ European	7 (58.3%)
Māori	1 (8.3%)
Asian	3 (25.0%)
Middle Eastern/ Latin American/African	1 (8.3%)
Average cycle length (days), mean ± SD	27.5 ± 2.4
Weight (kg), mean ± SD	66.0 ± 13.9
Height (cm), mean ± SD	166.9 ± 7.1
BMI (kg/m²), mean ± SD	23.6 ± 4.3
Sport participation, n (%)	Organised sports, 6 (60%) No participation in organised sports, 4 (40%)
Training hours per week, mean ± SD	5.4 ± 2.2

3.5.2 Comparison of hormonal concentrations across menstrual phases

A linear mixed-effects model was used to examine within-subject differences in hormonal concentrations across the four menstrual phases: menses, follicular, ovulation, and luteal. Table 5 presents the hormonal concentrations across the four menstrual phases.

Table 5 Hormonal concentrations across menstrual phases (mean ± SE).

Hormone group	Hormone	Menses	Follicular phase	Ovulation phase	Luteal phase	p-value
Reproductive	Oestradiol (pmol/L)	105.5 ± 40.7	178.3 ± 41.2	365.6 ± 40.7	297.2 ± 40.7	**<.001**
	Testosterone (nmol/L)	1.07 ± 0.1	1.12 ± 0.1	1.3 ± 0.1	1.1 ± 0.1	**0.004**
	Progesterone (nmol/L)	0.8 ± 1.6	0.6 ± 1.6	1.6 ± 1.6	13.3 ± 1.6	**<.001**
	LH (mIU/mL)	3.6 ± 1.3	4.9 ± 1.4	11.2 ± 1.3	4.9 ± 1.3	**<.001**
	FSH (mIU/mL)	5.5 ± 0.6	6.3 ± 0.6	6.3 ± 0.6	3.8 ± 0.6	**0.013**
	Prolactin (mIU/L)	174.5 ± 25.1	220.3 ± 25.3	233.2 ± 25.1	217.3 ± 25.1	0.063
Thyroid	TSH (uIU/ml)	0.8 ± 0.1	0.9 ± 0.1	1.1 ± 0.1	0.9 ± 0.1	**0.029**
	FT4 (pmol/L)	12.5 ± 0.2	13.0 ± 0.2	12.7 ± 0.2	12.6 ± 0.2	0.406
	TT4 (nmol/L)	6.6 ± 0.2	6.7 ± 0.2	6.6 ± 0.2	6.5 ± 0.2	0.886
Adrenal	Cortisol (nmol/L)	206.4 ± 23.9	237.4 ± 24.2	244.8 ± 23.9	213.1 ± 23.9	0.325
	DHEA-S (ug/mL)	6.8 ± 0.9	6.7 ± 0.9	7.6 ± 0.9	6.9 ± 0.9	**0.033**

Significant variations were observed in several reproductive hormones across the menstrual cycle. Oestradiol levels increased significantly across phases ($p < 0.001$), rising from menses to a peak during ovulation, before slightly decreasing during the luteal phase. Progesterone also showed a significant phase-dependent variation ($p < 0.001$), remaining low during menses, follicular, and ovulation phases, with a marked increase during the luteal phase. Testosterone levels differed significantly across phases ($p = 0.004$), with the highest concentration observed during ovulation. Similarly, LH demonstrated significant variation ($p < 0.001$), peaking during the ovulation phase, while FSH showed smaller but significant changes across phases ($p = 0.013$). Prolactin levels showed an increasing trend from menses to ovulation; however, this did not reach statistical significance ($p = 0.063$).

Among thyroid hormones, TSH varied significantly across menstrual phases (p = 0.029), with the highest concentration recorded during ovulation. In contrast, FT4 (p = 0.406) and TT4 (p = 0.886) did not show significant phase-related changes. For adrenal hormones, cortisol concentrations remained stable across the menstrual cycle and did not differ significantly between phases (p = 0.325). However, DHEA-S demonstrated a significant variation across menstrual phases (p = 0.033), with the highest levels observed during ovulation. Figures 4, 5 and 6 present the mean concentrations of progesterone, luteinising hormone (LH), follicle-stimulating hormone (FSH), and oestradiol across menstrual cycle phases.

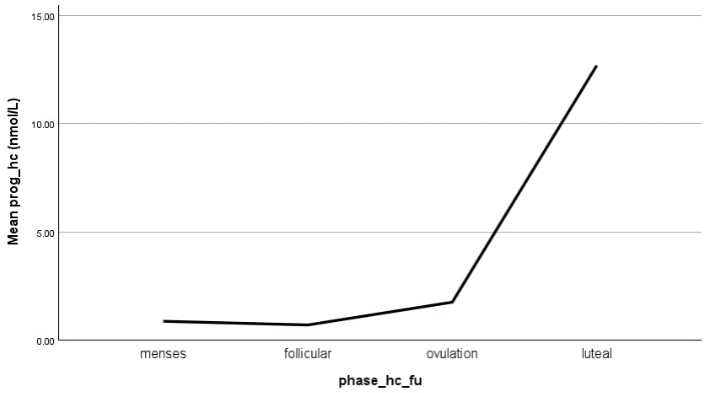

Figure 4 Mean concentrations of progesterone across menstrual cycle phases.

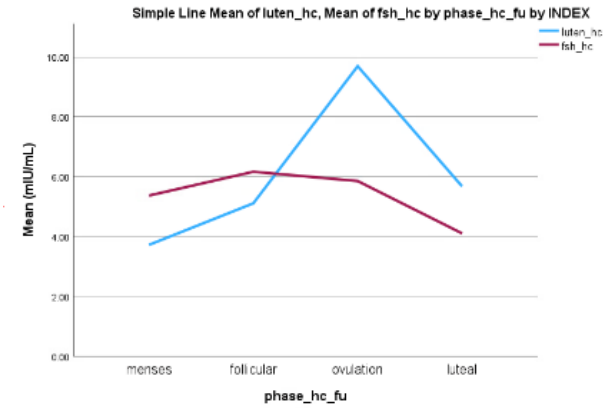

Figure 5 Mean concentrations of luteinising hormone (LH), and follicle-stimulating hormone (FSH) across menstrual cycle phases.

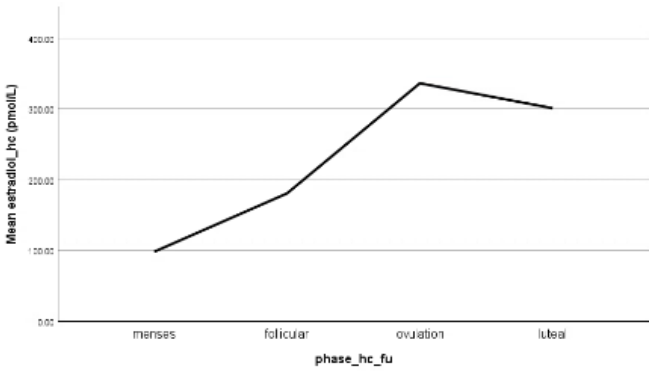

Figure 6 Mean oestradiol concentrations across menstrual cycle phases.

3.5.3 Pairwise comparisons between menstrual phases

Post hoc pairwise comparisons with Bonferroni adjustment were performed for hormones that showed significant overall phase effects in the linear mixed model. Table 6 summarizes pairwise comparisons of significant hormonal differences across menstrual phases. For oestradiol, concentrations were significantly higher during ovulation compared to menses (mean difference = −260.1 pmol/L, SE = 51.9, $p < 0.001$) and follicular phase (mean difference = −187.3 pmol/L, SE = 52.2, $p = 0.004$), and were also significantly higher during the luteal phase compared to menses (mean difference = −191.6 pmol/L, SE = 51.9, $p = 0.003$).

Progesterone concentrations were significantly elevated during the luteal phase compared to menses (mean difference = 12.4 nmol/L, SE = 2.1, $p < 0.001$), follicular phase (mean difference = 12.7 nmol/L, SE = 2.1, $p < 0.001$), and ovulation (mean difference = 11.6 nmol/L, SE = 2.1, $p < 0.001$). LH concentrations were significantly higher during ovulation compared to menses (mean difference = −7.6 mIU/mL, SE = 1.8, $p < 0.001$) and the follicular phase (mean difference = −6.2 mIU/mL, SE = 1.8, $p = 0.009$), while LH levels decreased significantly from ovulation to the luteal phase (mean difference = 6.2 mIU/mL, SE = 1.8, $p = 0.008$). For FSH, significantly higher concentrations were observed during the follicular and ovulation phases compared to the luteal phase (mean difference = 2.4 mIU/mL, SE = 0.8, $p = 0.026$ and $p = 0.025$, respectively). Testosterone concentrations were significantly higher during ovulation compared to menses (mean difference = 0.27 mIU/mL, SE = 0.07, $p = 0.004$) and the follicular phase (mean difference = 0.22 mIU/mL, SE = 0.07, $p = 0.023$). TSH was significantly higher during ovulation compared to menses (mean difference = −0.303 uIU/ml, SE = 0.102, $p = 0.026$).

Table 6 Pairwise comparisons of significant hormonal differences across menstrual phases.

Hormone	Comparison	Mean diff	SE	p-value
Oestradiol (pmol/L)	menses vs ovulation	-260.1	51.9	**<.001**
	menses vs luteal	-191.6	51.9	**0.003**
	follicular vs ovulation	-187.3	52.2	**0.004**
Progesterone (nmol/L)	luteal vs menses	12.4	2.1	**<.001**
	luteal vs follicular	12.7	2.1	
	luteal vs ovulation	11.6	2.1	
LH (mIU/mL)	menses vs ovulation	-7.6	1.8	**<.001**
	follicular vs ovulation	-6.2	1.8	**0.009**
	ovulation vs luteal	6.2	1.8	**0.008**
FSH (mIU/mL)	follicular vs luteal	2.4	0.8	**0.026**
	ovulation vs luteal	2.4	0.8	**0.025**
Testosterone (nmol/L)	ovulation vs menses	0.27	0.07	**0.004**
	ovulation vs follicular	0.22	0.07	**0.023**
TSH (uIU/ml)	menses vs ovulation	-.303	0.102	**0.026**

3.5.4 Gait variables across menstrual phases (single-task (ST) condition)

Kinetic gait parameters

No significant differences were observed in kinetic gait parameters across menstrual phases. Average vertical impact (left: p = 0.891; right: p = 0.853) and vertical push-off (left: p = 0.771; right: p = 0.781) remained stable across menses, follicular, ovulation, and luteal phases. Table 7 summarizes kinetic gait parameters for the ST condition.

Table 7 Kinetic gait parameters (mean ± SE).

Variable	Menses	Follicular phase	Ovulation phase	Luteal phase	p-value (phase)
Average vertical impact (left) (g)	2.40 ± 0.17	2.37 ± 0.17	2.38 ± 0.17	2.35 ± 0.17	0.891
Average vertical impact (right) (g)	2.48 ± 0.15	2.48 ± 0.15	2.43 ± 0.15	2.44 ± 0.15	0.853
Average vertical push-off (left) (g)	2.79 ± 0.14	2.73 ± 0.14	2.73 ± 0.14	2.70 ± 0.14	0.771
Average vertical push-off (right) (g)	2.70 ± 0.12	2.60 ± 0.12	2.64 ± 0.12	2.63 ± 0.12	0.781

Spatiotemporal gait parameters

Similarly, spatiotemporal gait parameters showed no significant phase related differences. Average walking speed (p = 0.809), ground contact time (p = 0.165), and double support ratio (p = 0.599) remained consistent across all menstrual phases. Table 8 summarizes spatiotemporal gait parameters for the ST condition.

Table 8 Spatiotemporal gait parameters (mean ± SE).

Variable	Menses	Follicular phase	Ovulation phase	Luteal phase	p-value (phase)
Average speed (m/s)	1.23 ± 0.04	1.22 ± 0.04	1.22 ± 0.04	1.21 ± 0.04	0.809
Average ground contact time (s)	0.64 ± 0.01	0.64 ± 0.01	0.64 ± 0.01	0.65 ± 0.01	0.165
Average double support ratio (ratio)	0.28 ± 0.008	0.28 ± 0.008	0.28 ± 0.008	0.29 ± 0.008	0.599

Gait variability parameters

Gait variability measures also demonstrated no statistically significant differences across menstrual phases. Stride length variability (left: $p = 0.107$; right: $p = 0.261$) and swing duration variability (left: $p = 0.664$; right: $p = 0.552$) remained stable across the cycle. Table 9 summarizes gait variability parameters for the ST condition.

Table 9 Gait variability parameters (mean ± SE).

Variable	Menses	Follicular phase	Ovulation phase	Luteal phase	p-value (phase)
Stride length variability (left) (m)	0.17 ± 0.01	0.16 ± 0.01	0.16 ± 0.01	0.16 ± 0.01	0.107
Stride length variability (right) (m)	0.23 ± 0.01	0.22 ± 0.01	0.23 ± 0.01	0.23 ± 0.01	0.261
Swing duration variability (left) (s)	0.02 ± 0.003	0.02 ± 0.003	0.02 ± 0.003	0.02 ± 0.003	0.664
Swing duration variability (right) (s)	0.02 ± 0.003	0.02 ± 0.003	0.02 ± 0.003	0.02 ± 0.003	0.552

3.5.5 Gait variables across menstrual phases (dual-task (DT) condition)

Kinetic gait parameters

Under dual-task walking conditions, no significant differences were observed in kinetic gait parameters across menstrual phases. Average vertical impact of both limbs remained stable across menses, follicular, ovulation, and luteal phases (left: $p = 0.454$; right: $p = 0.986$). Similarly, vertical push-off showed no significant phase-related changes for either limb (left: $p = 0.996$; right: $p = 0.714$). Table 10 summarizes kinetic gait parameters for the DT condition.

Table 10 Kinetic gait parameters (mean ± SE).

Variable	Menses	Follicular phase	Ovulation phase	Luteal phase	p-value (phase)
Average vertical impact (left) (g)	2.16 ± 0.09	2.12 ± 0.09	2.06 ± 0.09	2.13 ± 0.09	0.454
Average vertical impact (right) (g)	2.24 ± 0.17	2.24 ± 0.17	2.22 ± 0.17	2.24 ± 0.17	0.986
Average vertical push-off (left) (g)	2.50 ± 0.12	2.48 ± 0.12	2.49 ± 0.12	2.49 ± 0.12	0.996
Average vertical push-off (right) (g)	2.47 ± 0.12	2.35 ± 0.12	2.36 ± 0.12	2.38 ± 0.12	0.714

Spatiotemporal gait parameters

Spatiotemporal gait parameters also showed no statistically significant differences across menstrual phases during dual-task walking. Average speed ($p = 0.79$), ground contact time ($p = 0.82$), and double support ratio ($p = 0.806$) remained consistent across all phases of the menstrual cycle**.** Table 11 summarizes spatiotemporal gait parameters for the DT condition.

Table 11 Spatiotemporal gait parameters (mean ± SE).

Variable	Menses	Follicular phase	Ovulation phase	Luteal phase	p-value (phase)
Average speed (m/s)	1.1 ± 0.06	1.09 ± 0.06	1.08 ± 0.06	1.09 ± 0.06	0.79
Average ground contact time (s)	0.68 ± 0.02	0.68 ± 0.02	0.69 ± 0.02	0.69 ± 0.02	0.82
Average double support ratio (ratio)	0.3 ± 0.01	0.3 ± 0.01	0.3 ± 0.01	0.3 ± 0.01	0.806

Gait variability parameters

Gait variability measures demonstrated no significant menstrual phase effects under dual-task conditions. Stride length variability showed no significant differences for either limb (left: $p = 0.236$; right: $p = 0.632$). Similarly, swing duration variability remained stable across phases (left: $p = 0.457$; right: $p = 0.722$). Table 12 summarizes gait variability parameters for the DT condition.

Table 12 Gait variability parameters (mean ± SE).

Variable	Menses	Follicular phase	Ovulation phase	Luteal phase	p-value (phase)
Stride length variability (left) (m)	0.17 ± 0.01	0.15 ± 0.01	0.16 ± 0.01	0.17 ± 0.01	0.236
Stride length variability (right) (m)	0.21 ± 0.007	0.21 ± 0.007	0.21 ± 0.007	0.21 ± 0.007	0.632
Swing duration variability (left) (s)	0.03 ± 0.004	0.03 ± 0.004	0.03 ± 0.004	0.03 ± 0.004	0.457
Swing duration variability (right) (s)	0.02 ± 0.004	0.02 ± 0.004	0.03 ± 0.004	0.02 ± 0.004	0.722

3.5.6 Gait variables across menstrual phases (head-turn (HT) condition)

Kinetic gait parameters

Under head turn walking conditions, no significant differences were observed in kinetic gait parameters across menstrual phases. Average vertical impact of both limbs remained stable across menses, follicular, ovulation, and luteal phases (left: $p = 0.914$; right: $p = 0.530$). Similarly, vertical push-off showed no significant phase-related changes for either limb (left: $p = 0.965$; right: $p = 0.879$). Table 13 summarizes kinetic gait parameters for the HT condition.

Table 13 Kinetic gait parameters (mean ± SE).

Variable	Menses	Follicular phase	Ovulation phase	Luteal phase	p-value (phase)
Average vertical impact (left) (g)	2.16 ± 0.08	2.15 ± 0.08	2.14 ± 0.08	2.18 ± 0.08	0.914
Average vertical impact (right) (g)	2.27 ± 0.14	2.22 ± 0.14	2.21 ± 0.14	2.3 ± 0.14	0.530
Average vertical push-off (left) (g)	2.77 ± 0.1	2.78 ± 0.1	2.73 ± 0.1	2.78 ± 0.1	0.965
Average vertical push-off (right) (g)	2.7 ± 0.12	2.6 ± 0.12	2.6 ± 0.12	2.6 ± 0.12	0.879

Spatiotemporal gait parameters

Spatiotemporal gait parameters also showed no statistically significant differences across menstrual phases during head turn walking. Average speed ($p = 0.713$), ground contact time ($p = 0.858$), and double support ratio ($p = 0.339$) remained consistent across all phases of the menstrual cycle. Table 14 summarizes spatiotemporal gait parameters for the HT condition.

Table 14 Spatiotemporal gait parameters (mean ± SE).

Variable	Menses	Follicular phase	Ovulation phase	Luteal phase	p-value
Average speed (m/s)	1.14 ± 0.05	1.13 ± 0.05	1.14 ± 0.05	1.15 ± 0.05	0.713
Average ground contact time (s)	0.66 ± 0.01	0.65 ± 0.01	0.65 ± 0.01	0.66 ± 0.01	0.858
Average double support ratio (ratio)	0.3 ± 0.01	0.3 ± 0.01	0.2 ± 0.01	0.3 ± 0.1	0.339

Gait variability parameters

Gait variability measures demonstrated no significant differences across menstrual phases under head turn conditions. Stride length variability (left: $p = 0.542$; right: $p = 0.668$) and swing duration variability (left: $p = 0.868$; right: $p = 0.868$) remained stable across menses, follicular, ovulation, and luteal phases. Table 15 summarizes gait variability parameters for the HT condition.

Table 15 Gait variability parameters (mean ± SE).

Variable	Menses	Follicular phase	Ovulation phase	Luteal phase	p-value (phase)
Stride length variability (left) (m)	0.15 ± 0.01	0.14 ± 0.01	0.15 ± 0.01	0.15 ± 0.01	0.542
Stride length variability (right) (m)	0.21 ± 0.008	0.20 ± 0.008	0.20 ± 0.008	0.20 ± 0.008	0.668
Swing duration variability (left) (s)	0.02 ± 0.003	0.02 ± 0.003	0.02 ± 0.003	0.02 ± 0.003	0.868
Swing duration variability (right) (s)	0.02 ± 0.003	0.02 ± 0.003	0.02 ± 0.003	0.02 ± 0.003	0.868

3.5.7 Cognitive and motor dual-task cost across menstrual phases

Cognitive dual-task cost (DTC-C)

No significant differences were observed in cognitive dual-task cost across menstrual phases ($p = 0.619$). Although minor fluctuations were observed, DTC-C remained relatively stable across menses, follicular, ovulation, and luteal phases, with no clear phase-dependent pattern.

Motor dual-task cost (DTC-M)

Similarly, motor dual-task cost did not show any significant differences across menstrual phases ($p = 0.549$). DTC-M values remained consistently negative across all phases, indicating a comparable reduction in gait performance under cognitive loading regardless of menstrual phase. Table 16 summarizes the results of cognitive and motor dual-task cost across menstrual phases.

Table 16 Cognitive and motor dual-task cost across menstrual phases (mean ± SE).

Variable	Menses	Follicular phase	Ovulation phase	Luteal phase	p-value (phase)
DTC-Cognitive (%)	-13.48 ± 6.32	-6.46 ± 6.32	-2.88 ± 6.32	-11.55 ± 6.32	0.619
DTC-Motor (%)	-11.23 ± 3.68	-12.38 ± 3.68	-14.19 ± 3.68	-12.05 ± 3.68	0.549

Overall, across single-task (ST), dual-task (DT), and head turn (HT) conditions, no significant differences were found in kinetic, spatiotemporal, or gait variability parameters across menstrual phases. Similarly, cognitive and motor dual-task cost (DTC) showed no significant phase-related changes. Overall, gait performance remained stable across all walking conditions throughout the menstrual cycle.

3.6 Discussion

The present prospective observational pilot study was conducted with a small sample size and was designed primarily to assess feasibility, perform descriptive analyses, and generate preliminary estimates rather than undertake formal hypothesis testing. Accordingly, the findings are intended to inform the design and methodology of future larger-scale studies.

Overall, the study identified expected cyclical variations in several reproductive hormones across the menstrual cycle, while gait performance remained largely unchanged across menstrual phases. Significant phase-related differences were observed in several reproductive hormones, including oestradiol, progesterone, testosterone, LH, FSH, prolactin, TSH, and DHEA-S, reflecting normal endocrine fluctuations across the cycle. However, not all hormonal measures followed this pattern. FT4, TT4, and cortisol did not show significant phase-related differences, indicating relative stability across menstrual phases for these hormones. Despite these clear endocrine variations, gait outcomes remained stable across menstrual phases. No significant differences were observed in kinetic, spatiotemporal, or gait variability measures across ST, DT, or HT conditions. Similarly, cognitive and motor dual-task costs showed no significant phase-related changes. The following sections discuss these findings in relation to the existing literature and their potential implications for injury risk and gait performance.

3.6.1 Menstrual cycle-related hormonal changes across menstrual phases

The present study demonstrated significant hormonal changes across the menstrual cycle in healthy, naturally menstruating females. Specifically, oestradiol concentrations increased from menses to ovulation before declining during the luteal phase, reflecting the expected pre-ovulatory surge associated with follicular maturation and ovulation. This pattern is consistent with established endocrine physiology and supports the accuracy of menstrual phase classification within the study cohort (Reed & Carr, 2018). Similar cyclical variations in oestradiol have been reported in previous studies of naturally menstruating females(Chen et al., 2021; Ott et al., 2024), suggesting that the hormonal profiles observed in the present study are representative of normal menstrual cycle physiology.

The observed changes in oestradiol may have important implications for musculoskeletal function and injury susceptibility (Pfeifer et al., 2024). Previous research has suggested that elevated oestrogen concentrations can influence connective tissue properties and ligament laxity (Herzberg et al., 2017; Martini et al., 2011). For example, Herzberg et al. (2017) reported an increased likelihood of ACL injury during the pre-ovulatory phase, while Martini et al. (2011) observed a greater proportion of muscle tendon injuries during the late follicular phase. Similarly, (Pfeifer et al., 2024) proposed that elevated oestrogen concentrations may

increase susceptibility to myofibre injury while simultaneously influencing inflammatory responses following tissue damage. Collectively, these findings suggest that fluctuations in oestradiol may contribute to alterations in tissue biomechanics and neuromuscular function throughout the menstrual cycle.

In addition to oestradiol, progesterone demonstrated significant phase-related variation, remaining low during menses, the follicular phase, and ovulation before increasing markedly during the luteal phase. This pattern is consistent with the physiological role of the corpus luteum and reflects normal reproductive endocrine regulation (Reed & Carr, 2018). Beyond its reproductive functions, progesterone has been proposed to influence injury-related outcomes and recovery processes (Chen et al., 2021; Yu et al., 2001). Experimental evidence suggests that progesterone promotes fibroblast proliferation and collagen synthesis, potentially reducing ligament laxity and enhancing tissue repair (Yu et al., 2001). Furthermore, (Chen et al., 2021) reported that progesterone may exert neuroprotective effects through improved cerebral perfusion and modulation of physiological stress responses. However, the role of progesterone remains incompletely understood. (Ott et al., 2024) reported greater concussion symptom severity and poorer neurocognitive performance during the luteal phase despite elevated progesterone concentrations, indicating that hormonal influences on injury risk and recovery are likely multifactorial and may involve interactions with other physiological mechanisms.

Beyond oestradiol and progesterone, significant phase-related differences were also observed in LH, FSH, testosterone, TSH, and DHEA-S. These findings further demonstrate the coordinated endocrine changes that occur across the menstrual cycle and are consistent with previous studies describing cyclical modulation of multiple hormonal systems in naturally menstruating females (Chen et al., 2021; Ott et al., 2024). Together, these hormonal variations reinforce the physiological validity of the menstrual phase classification used in the present study and highlight the complex endocrine environment that characterises the menstrual cycle.

In contrast, FT4, TT4, and cortisol did not demonstrate significant phase-related variation, suggesting relative stability of these hormones across the menstrual cycle. The absence of significant changes in these markers indicates that not all endocrine systems are influenced to the same extent by menstrual cycle phase. Taken together, the hormonal findings of the present study demonstrate expected cyclical variation in key reproductive hormones alongside relative stability in several non-reproductive hormones, providing a comprehensive overview of endocrine changes across the menstrual cycle in healthy females.

Although the present study was not designed to directly evaluate injury outcomes, the observed hormonal fluctuations provide preliminary evidence supporting the biological plausibility of menstrual cycle-related influences on injury susceptibility. However, whether these endocrine changes translate into measurable alterations in movement patterns or injury risk remains unclear. To explore this possibility, the present study also examined gait performance across menstrual phases.

3.6.2 Gait variables across menstrual phases

Despite the significant hormonal fluctuations observed throughout the menstrual cycle, no significant differences were identified in gait performance across menstrual phases under single-task, dual-task, or head-turn walking conditions. Kinetic variables, including average vertical impact and vertical push-off, remained stable throughout the cycle. Similarly, spatiotemporal measures, gait variability parameters, and cognitive and

motor dual-task costs demonstrated no significant phase-related changes. These findings suggest that menstrual cycle-related hormonal fluctuations may not substantially influence gait performance in healthy females under controlled laboratory conditions.

The current findings align with previous research suggesting that healthy individuals generally maintain stable gait performance despite physiological fluctuations. Howell et al. (2020) reported that healthy controls demonstrated consistent gait performance across walking conditions, whereas gait deficits were more evident among individuals recovering from concussion. Further evidence comes from Jain et al. (2024), who found no significant differences in several gait measures between healthy and concussed participants despite reductions in performance during dual-task conditions. Together, these findings suggest that gait performance is generally stable in healthy individuals and may not be substantially affected by physiological or cognitive challenges.

Several explanations may account for the absence of phase-related differences in gait outcomes. Healthy females may possess sufficient neuromuscular adaptability to maintain stable gait performance despite hormonal variation. Additionally, the gait variables assessed in the present study may not have been sensitive enough to detect subtle menstrual cycle-related changes. Finally, the small sample size may have limited the ability to detect small but potentially meaningful effects.

Collectively, the findings indicate that although significant hormonal changes occur throughout the menstrual cycle, these fluctuations do not appear to result in measurable changes in gait performance in healthy females. This provides important preliminary evidence for future investigations examining whether hormonal influences become more apparent under conditions involving greater physiological or neurological challenge.

3.7 Study limitations and future directions

Several limitations should be considered when interpreting the findings of this pilot study. First, the small sample size limited statistical power and reduced the ability to detect subtle phase-related differences in gait performance. Second, menstrual cycle phase classification partially relied on participant-reported cycle information, which may be subject to recall error and variability in cycle length.

Data collection was conducted within a laboratory environment using relatively short walking trials. Although this approach provided a controlled assessment environment, it may not accurately reflect movement patterns observed during daily activities or sporting participation. Additionally, variability in participant characteristics, including sport participation levels and training exposure, may have contributed to heterogeneity in gait outcomes. Frequent blood sampling across the study period also created a participant burden and may have influenced retention and compliance. Future research should investigate gait performance in more ecologically valid settings using wearable technologies and longer duration movement assessments. Real world monitoring may provide greater insight into functional movement behaviours than short laboratory-based walking trials conducted in controlled environments. Such approaches may improve understanding of how hormonal fluctuations influence movement patterns during everyday activities and sporting participation.

Building on these considerations, future studies should recruit larger cohorts to improve statistical power and enhance the generalisability of findings. The use of more objective menstrual cycle monitoring methods,

including ovulation testing and more frequent hormonal assessments, may further improve the accuracy of menstrual phase classification. Additionally, future studies may benefit from examining sport-specific populations, as movement demands and injury mechanisms vary considerably across different sports. Longitudinal studies incorporating repeated assessments across multiple menstrual cycles would further enhance understanding of within-person variability and the influence of hormonal fluctuations on movement patterns and injury risk.

3.8 Conclusions

This prospective observational pilot study investigated menstrual cycle-related hormonal changes and gait alterations across menstrual cycle phases in healthy females. Significant phase-related differences were observed in several reproductive hormones, particularly oestradiol, progesterone, LH, FSH, and testosterone, confirming expected endocrine variations across the menstrual cycle. However, no significant differences were identified in kinetic, spatiotemporal, gait variability, or dual-task gait measures across menstrual phases. These findings suggest that menstrual cycle-related hormonal changes do not appear to substantially influence gait alterations in healthy females under controlled laboratory conditions. Although hormonal variation may influence physiological processes associated with injury susceptibility, such effects were not reflected in measurable changes in gait within the present cohort. The findings provide preliminary evidence to inform future larger-scale studies investigating hormonal influences on neuromuscular function and injury risk in female populations.

CHAPTER 4: DISCUSSION AND CONCLUSION

4.1 Discussion

This thesis aimed to investigate the potential relationship between menstrual cycle related hormonal changes and gait alterations in females following concussion, including their potential implications for secondary musculoskeletal injury and recurrent concussion. The research comprised two components: a scoping review examining the existing literature on how menstrual cycle phases, hormonal changes, and gait alterations are related to concussion outcomes and their potential implications for secondary musculoskeletal injury and recurrent concussion, and a prospective observational pilot study investigating menstrual cycle-related hormonal changes and gait alterations across menstrual cycle phases in healthy females.

The scoping review identified a substantial gap in the literature regarding the interaction between menstrual cycle-related hormonal changes, gait alterations, and secondary MSK injury risk and recurrent concussion following concussion. While previous studies have independently examined hormonal influences on concussion outcomes and gait deficits following concussion, no studies directly investigated how menstrual cycle-related hormonal changes and gait alterations interact to influence secondary injury risk in females. The pilot study was therefore aimed to provide preliminary data addressing this gap by characterising hormonal and gait changes across menstrual phases.

Together, the findings indicated that significant hormonal fluctuations occur throughout the menstrual cycle, particularly in reproductive hormones such as oestradiol, progesterone, LH, FSH, and testosterone. However, despite these hormonal variations, gait performance remained relatively stable across menstrual phases under single-task, dual-task, and head-turn walking conditions. These findings suggested that although hormonal changes may influence physiological processes relevant to injury susceptibility, their direct influence on gait performance in healthy females may be limited.

It is important to consider the differences in design and purpose when interpreting the findings of the pilot study alongside the evidence identified in the scoping review. The scoping review examined existing research on menstrual cycle-related hormonal changes, gait alterations, and concussion outcomes across diverse post-concussion populations and study designs. In contrast, the present study was a prospective observational pilot case study conducted in healthy females to characterise hormonal and gait changes across menstrual cycle phases. Although the study was initially intended to include naturally menstruating females following concussion, eligible participants could not be recruited within the study period. Therefore, the findings provide preliminary evidence regarding menstrual cycle-related hormonal and gait variation in a healthy population and should be interpreted as complementary to, rather than directly comparable with, findings from studies involving females with concussion.

4.1.1 Menstrual cycle-related hormonal changes and their potential implications for secondary injury risk in females

The pilot study demonstrated significant phase-related differences in reproductive hormone concentrations. Oestradiol concentrations peaked during ovulation, while progesterone concentrations reached their highest

levels during the luteal phase. These findings are consistent with established endocrine patterns and support the validity of menstrual phase classification within the study.

The findings also aligned with evidence identified in the scoping review suggesting that hormonal fluctuations may influence musculoskeletal and neurological function. Several studies have reported associations between menstrual cycle phase and injury occurrence. Herzberg et al. (2017) observed an increased likelihood of ACL injury during the pre-ovulatory phase, while Martini et al. (2011) reported a greater proportion of muscle–tendon injuries during the late follicular phase. Similarly, Pfeifer et al. (2024) suggested that elevated oestrogen concentrations may increase susceptibility to myofibre injury, potentially through effects on tissue structure and inflammatory responses. The present study confirmed the occurrence of substantial hormonal fluctuations that may provide a biological basis for these observations. Elevated oestrogen concentrations during ovulation have been associated with increased ligament laxity and altered connective tissue properties, which may contribute to injury susceptibility. Furthermore, Parker et al. (2024) reported that elevated relaxin concentrations during the late luteal phase may increase ACL injury risk through effects on ligament integrity.

Progesterone may also influence injury-related outcomes. Experimental evidence suggested that progesterone promotes collagen synthesis and fibroblast activity, potentially contributing to tissue repair and reduced ligament laxity (Yu et al., 2001). In addition, Chen et al. (2021) reported that progesterone may exert neuroprotective effects through modulation of cerebral blood flow and physiological stress responses. These findings supported the hypothesis that hormonal fluctuations may influence both musculoskeletal and neurological processes relevant to injury risk and recovery.

However, findings across the literature remained inconsistent. The scoping review identified conflicting evidence regarding the role of progesterone and menstrual cycle phase in concussion outcomes. Wunderle et al. (2014) suggested that injuries occurring during the luteal phase may be associated with poorer outcomes, whereas Roby et al. (2023) reported limited evidence supporting a clear relationship between menstrual phase and recovery. Similarly, Ott et al. (2024) observed greater symptom severity and poorer neurocognitive performance during the luteal phase despite elevated progesterone concentrations, while Goeckner et al. (2025) found no significant association between hormone levels and recovery outcomes. Taken together, the findings of both the scoping review and pilot study suggested that hormonal fluctuations are biologically meaningful and may contribute to injury susceptibility and recovery processes. However, current evidence remains insufficient to establish direct causal relationships between specific hormonal profiles and injury outcomes.

4.1.2 Gait performance across menstrual phases

A major finding of the pilot study was the absence of significant differences in gait performance across menstrual phases. Kinetic, spatiotemporal, gait variability, and dual-task cost measures remained relatively stable throughout the menstrual cycle despite significant hormonal variation. When considered alongside existing literature, these findings align with studies in healthy populations reporting limited gait variability under both single- and dual-task conditions. For example, Howell et al. (2020) found minimal gait differences in healthy individuals during single-task walking, while Jain et al. (2024) reported that both healthy and

concussed participants demonstrated reduced gait performance under dual-task conditions, with no consistent between-group differences in some gait variables. These findings suggest that gait performance in healthy populations may remain relatively stable despite changes in task demand or physiological state.

In contrast, studies involving concussed populations have reported persistent gait alterations, particularly during dual-task conditions. Howell et al. (2020) observed slower walking speed and altered gait dynamics in individuals recovering from concussion, while David et al. (2020) reported increased gait determinism, laminarity, and trapping time following injury. Further evidence indicates that Lowe et al. (2023) and Messerschmidt et al. (2021) demonstrated that females exhibit greater dual-task gait deficits than males following concussion. Importantly, these alterations are generally interpreted as reflecting neurological impairment and disrupted cognitive–motor integration rather than hormonal influences alone.

The absence of gait changes across menstrual phases in the pilot study suggests that hormonal fluctuations in isolation may not meaningfully influence gait performance in healthy females under controlled laboratory conditions. In contrast, gait abnormalities reported in concussion populations are more likely to be associated with central nervous system dysfunction, impaired postural control, and increased cognitive–motor interference following injury. However, this interpretation should be considered cautiously given the exploratory nature of the study and the small sample size. The limited sample size may have reduced the sensitivity of the analysis to detect small phase-related changes in gait that could nevertheless be meaningful. Thus, the observed stability in gait measures should not be interpreted as definitive evidence that menstrual-cycle-related hormonal fluctuations have no influence on gait. It is possible that subtle phase-related gait changes may exist but were not detectable using the assessment methods employed. More sensitive biomechanical tools, longer-duration monitoring, or real-world movement analysis may be required to capture minor neuromotor variations associated with hormonal fluctuations.

Overall, these findings suggest that healthy females can maintain relatively stable gait performance across the menstrual cycle, potentially reflecting neuromuscular compensation mechanisms that preserve locomotor control despite endocrine variation. Future research incorporating larger cohorts and both healthy and concussed females with matched control groups would help clarify whether menstrual cycle phases interact with concussion status to influence gait control and secondary injury risk. Longitudinal designs with repeated measures across multiple menstrual cycles would further strengthen understanding of within-person variability and improve detection of subtle phase-dependent effects.

4.2 Limitations and future directions

A key limitation of this thesis was the absence of naturally menstruating concussed female participants in the pilot study, primarily due to the limited study timeframe and inability to recruit individuals who met the inclusion criteria within this period. As a result, the intended comparison between healthy and concussed females could not be completed. This significantly limits the ability to directly examine the interaction between concussion status, menstrual cycle related hormonal changes, and gait alterations, thereby restricting conclusions regarding secondary musculoskeletal injury risk following concussion.

The scoping review also highlighted important limitations within the existing literature. Only a small number of studies meeting the inclusion criteria (n = 11), reflecting the emerging nature of this research area. In addition, the included studies were highly heterogeneous in terms of study design, outcome measures, participant characteristics, and methods used to assess menstrual cycle phase and hormonal status. Many relied on self-reported cycle data or inconsistent hormonal assessment protocols, increasing the risk of misclassification and limiting comparability across studies. Furthermore, very few studies examined gait outcomes in relation to menstrual cycle phases, and none investigated the combined interaction between menstrual cycle-related hormonal changes, gait alterations, and secondary MSK injury risk and recurrent concussion following concussion. The pilot study itself also had methodological constraints. The small sample size may have reduced the sensitivity of the study to identify subtle menstrual cycle-related changes in gait, and the findings should therefore be considered preliminary. In addition, gait assessment was conducted in a controlled laboratory environment using short-duration walking trials, which may not fully capture the complexity and variability of real-world locomotor behaviour.

An additional consideration for the generalisability of the present findings is that the pilot study primarily included females with regular, naturally occurring menstrual cycles. This criterion was used to facilitate more consistent identification of menstrual-cycle phases and interpretation of cyclical hormonal changes. However, menstrual irregularity, PCOS, and hormonal contraceptive use can alter endogenous hormonal profiles and may influence physiological and neuromuscular responses (Colenso-Semple et al., 2023; Elliott-Sale et al., 2021; McNulty et al., 2020). Therefore, the findings may not be directly generalisable to females with irregular menstrual cycles, PCOS, or those using hormonal contraceptives.

To address these limitations, future research should adopt large-scale longitudinal designs including both healthy and concussed female cohorts. The integration of wearable inertial sensor technology and extended real-world monitoring is recommended to improve ecological validity and better reflect functional gait behaviour in naturalistic settings. In addition, future work should directly investigate the combined influence of menstrual hormones and gait changes on concussion recovery and secondary musculoskeletal injury risk, including both healthy and concussed groups, thereby addressing a critical gap identified across both the scoping review and pilot study components of this thesis.

4.3 Conclusion

This thesis combined a scoping review and a prospective observational pilot case study aimed at investigating the relationship between menstrual cycle related hormonal changes and gait alterations in relation to secondary musculoskeletal injury risk and recurrent concussion in females. The scoping review highlighted limited and heterogeneous evidence, with no studies directly examining the interaction between menstrual cycle phase, hormonal changes, gait alterations, and secondary MSK injury risk following concussion, indicating a clear gap in the literature. The pilot study demonstrated significant hormonal fluctuations across the menstrual cycle in healthy females; however, no significant differences in gait performance were observed across phases under controlled laboratory conditions. The absence of concussed participants limits the ability to extend these findings to post-concussion populations. As a preliminary pilot investigation, the observed lack of significant

gait differences should be interpreted cautiously rather than as definitive evidence that menstrual-cycle-related hormonal fluctuations have no influence on gait. Overall, the findings suggest that while hormonal fluctuations are biologically significant, their influence on gait performance in healthy females remains uncertain, and further research is required to explore their interaction with concussion and injury risk.

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APPENDICES

Appendix A – AUTEC ethics

Auckland University of Technology Ethics Committee
(AUTEC)

26 August 2025

Patria Hume
Faculty of Health and Environmental Sciences

Dear Patria

Re Ethics Application: **25/152 Walking patterns, hormonal cycles, and injury risks in female athletes' post-concussion (WHIP)**

Thank you for your responses to AUTEC's conditions.

Your ethics application has been approved for three years until 26 August 2028.

Standard Conditions of Approval

1. The research is to be undertaken in accordance with the Auckland University of Technology Code of Conduct for Research and as approved by AUTEC.
2. All public facing documents must have the AUTEC approval number and be of a high standard of spelling and grammar. Dates on the Information Sheet(s) and Consent Form(s) must be consistent.
3. Any amendments to the project must be approved by AUTEC prior to being implemented.
4. A progress report is due annually on the anniversary of the approval date.
5. A final report is due at the expiration of the approval period, or, upon completion of project.
6. Any serious or adverse events must be reported to AUTEC, this includes unforeseen issues that might affect continued ethical acceptability of the project.
7. AUTEC grants ethical approval only. You are responsible for obtaining management permission for access from any institution or organisation at which your research is being conducted and you need to meet all ethical, legal, public health, and locality obligations or requirements for the jurisdictions in which the research is being undertaken.

The application number and title need to be referenced on all correspondence related to this project.

All forms are available online http://www.aut.ac.nz/research/researchethics

For any enquiries, please contact the Secretariat at ethics@aut.ac.nz
(This is a computer-generated letter for which no signature is required)

The AUTEC Secretariat
Auckland University of Technology Ethics Committee

Cc: sarniphy@gmail.com; anja.zoellner@aut.ac.nz; [redacted]; [redacted]; christi.essex@autuni.ac.nz; maggie.sandleback@aut.ac.nz

Auckland University of Technology, D-88, Private Bag 92006, Auckland 1142, New Zealand.
T: +64 9 921 9999 ext. 8316: E: ethics@aut.ac.nz: www.aut.ac.nz/researchethics

Appendix B – HDEC ethics

Health and Disability Ethics Committees

Health and Disability Ethics Committees
Ministry of Health
133 Molesworth Street
PO Box 5013
Wellington
6011
hdecs@health.govt.nz

Ethics reference: 2025 AM 21888

27 June 2025

Professor Patria Hume

17 Antares Place
Mairangi Bay
Auckland 1142
New Zealand

Tēnā koe Professor Hume

APPROVAL OF AMENDMENT

Study title: Determining optimal sampling times for mTBI blood biomarkers for prediction of recovery in females

I am pleased to advise that this amendment was **approved** by the Southern Health and Disability Ethics Committee (the Committee). This decision was made through the post-approval pathway.

Further information and assistance

Please contact the HDECs Secretariat at hdecs@health.govt.nz or visit our website at www.ethics.health.govt.nz for more information, as well as our General FAQ and Ethics RM manual.

Nāku noa, nā

Mr Dominic Fitchett

Chair

Southern Health and Disability Ethics Committee

Encl: Appendix A: documents submitted

Appendix A: Documents submitted

Document Type	File Name	Date	Version
PAF	Clean WHN Study 1 mTBI blood sample HDEC PIS 240625 version 4	24/06/2025	4
PAF	Clean WHN Study 1 mTBI blood sample HDEC CONSENT 240625 version 4	24/06/2025	4
PAF	TC WHN Study 1 mTBI blood sample HDEC PIS 240625 version 4	24/06/2025	4
PAF	TC WHN Study 1 mTBI blood sample HDEC CONSENT 240625 version 4	24/06/2025	4
PAF	Minor Amendments Letter 240625	24/06/2025	4

Appendix C - Participant information sheet

Participant Information Sheet

Date Information Sheet Produced: 30th July 2025

Project title: Walking patterns, hormonal cycles, and injury risks in female athletes' post-concussion

This project is part of the Women's Health and Neuroscience Research Programme study "Determining optimal sampling times for concussion blood biomarkers for prediction of recovery in females" approved by HDEC 2024 EXP 21888, and registered with ANZCTR ACTRN12625000260426.

Research Team

Co-ordinating Investigator: Professor Patria Hume

Co-Principle Investigators: Dr Nusratnaaz Shaikh
Co-Associate Investigators: Dr. Anja Zoellner, Dr Christi Essex, Marguerite Sandleback
Primary Researcher: Sarniya Moganathas
Sponsor: Auckland University of Technology

Study Sites: AUTM: AUT Millennium in Mairangi Bay, Auckland, AUT-city: AUT City Campus in Auckland CBD, ASM: Axis Sports Medicine in Auckland

Contact phone number: AUCKLAND: Sarniya Moganathas 022 519 1726

Ethics committee ref.:

An Invitation

Tēnā koe (greetings to you). My name is Sarniya Moganathas and I am conducting my masters research at AUT. On behalf of the research team, you are invited to participate in this research study that aims to observe preliminary trends in how hormonal cycle changes correlate with gait pattern alterations and their potential contribution to secondary musculoskeletal injuries and repeated concussions upon return to play in female athletes. You are invited to participate in the study because you have sustained a recent head injury (mTBI group = mild traumatic brain injury/concussion) or are a part of the non-concussed control group.

Participation is voluntary and your decision to participate or not will not affect any healthcare you or your whānau will receive. If you agree to participate you will be assessed at AUT Millennium in Auckland.

We will collect some basic information about you, such as your age, height, weight, lifestyle information, any medical conditions you have, what medications you currently take, your ethnicity, your menstrual cycle. We will also ask you to fill some surveys about your head injury, do some blood tests, and gait monitoring. During the lab/clinic visits, we will collect a venous blood sample. We will collect several samples, with your permission, across your menstrual period. This is to determine how the hormonal fluctuation affect the gait patterns and likelihood of secondary musculoskeletal injuries and repeated concussion. They might be a good markers for the diagnosis.

The data collected within this research will be analysed only by the research team and will be presented as de-identified group data before being published.

Please ensure you read the information below and understand this prior to partaking in the study. If you have further questions or concerns, you can contact the research team using the details at the bottom of this document.

This Participant Information Sheet will help you decide if you'd like to take part. It sets out why we are doing the study, what your participation would involve, what the benefits and risks to you might be, and what would happen after the study ends. We will go through this information with you and answer any questions you may have. You do not have to decide today whether or not you will participate in this study. Before you decide you

may want to talk about the study with other people, such as family, whānau, friends, or healthcare providers. Feel free to do this.

If you agree to take part in this study, you will be asked to sign the Consent Form. You will be given a copy of both the Participant Information Sheet and the Consent Form to keep.

This document is 8 pages long. Please make sure you have read and understood all the pages.

WHAT IS THE PURPOSE OF THE STUDY?

This study aims to improve how we detect and manage risk of secondary musculoskeletal injuries after concussion by using hormonal testing and gait parameter analysis. Sport related concussion injuries often occur, especially female athletes are more susceptible to concussions, with prolonged symptoms and higher rates of secondary musculoskeletal (MSK) injuries compared to males. So far, the only way to diagnose if someone has a mild or moderate traumatic brain injury (often called a concussion) is by scanning their brain with a CT or MRI scanner and performing several medical exams. There can be quite a delay between someone being injured and when they can be seen by medical specialist and given a CT scan.

Scientists have been trying to develop blood tests and gait analysis methods that would help to diagnose a fast and easy way to diagnose the recovery stage from concussion and prevent further injuries than how we currently do it. This research is trying to test if menstrual hormone's levels and gait parameters are useful to monitor risk probability of repeated concussions and secondary MSK injuries.

Additionally, females seem to suffer from longer and worse symptoms after mTBI/concussion. To identify if the menstrual cycle (period) plays a role in the severity of mTBI symptoms, blood tests will be performed to look at the levels of female sex hormones. The levels of these hormones after injury may help to determine why females have worse symptoms compared with males and will help to design cycle specific gait and balance training to prevent further injuries and improve their performance.

HOW WAS I IDENTIFIED TO PARTICIPATE IN THIS SURVEY?

You would be identified for this study by a healthcare professional who has diagnosed you with a mTBI/concussion. This professional may inform you about this study themselves, or they may ask your permission for a member of the study team to contact you about this study. Or if you have not experienced a concussion, you are a non-contact sport athlete that was identified to be a part of the non-concussed group.

HOW DO I AGREE TO PARTICIPATE IN THIS STUDY?

You can contact a member of the study team to participate in this study. If you would like to get involved, a member of the study team will walk you through this document and answer any questions you or your whānau might have. You have the option of either doing an online consent form or completing the consent form when you attend the first session.

WHAT WILL MY PARTICIPATION IN THE STUDY INVOLVE?

You will need to be willing to fill in some forms that ask you about your sex, age, ethnicity, concussion history, last menstrual date, and some general health questions . This is to make sure we know there are no conditions or medication that might affect the testing. You must also be willing to provide several blood samples. This will only be taken by someone trained to do so. There are also some non-invasive tests of your gait that will also happen.

- You will need to come to one of our clinics (ideally within 7 days post-concussion, then on menstrual cycle days; Menstrual Phase: Days 1–5, Post-Menstrual Phase: Days 6–13, Mid-Cycle (Ovulation): Day 14, Pre-Menstrual Phase: Days 15–24 , for three months). Three menstrual cycles will be tracked. . This is a total of up to 12 visits and these dates will be provided for you.

None of the research tests are diagnostic, they will not be recorded in your medical files. They are for research only. Here is a list of the test we would like to perform:

- Height and weight (5 min)
- General Health Questionnaire and Menstrual cycle questions
- Surveys (20 min): Brain Injury Screening Tool-10, Perceived Recovery Scale, Glasgow Outcome Scale Extended.
- Walking assessment (Gait) (10 min) whilst using Plantiga pressure insole sensors placed in your shoes.
- Blood sample collection (10 min).

This research includes basic information such as your ethnic group, age range, and sex. It is possible that this research could one day help people in the same groups as you. However, it is also possible that research findings could be used inappropriately to support negative stereotypes, stigmatize, or discriminate against members of the same groups as you. The study team will take utmost care with regards to this but are unable to control how other people choose to use the information we may publish.

There is also a small risk the study team may make an incidental finding that might affect your health. In this case, we would immediately report the result to your General Practitioner (GP). GP notification of clinically significant abnormal results is a mandatory component of study participation. Therefore, GP contact details are requested on the consent form.

If you feel you need counselling or support due to taking part in this research study AUT Health Counselling and Wellbeing is able to offer three free sessions of confidential counselling support for adult participants in an AUT research project. These sessions are only available for issues that have arisen directly as a result of participation in the research and are not for other general counselling needs. To access these services, you will need to:

- drop into our centres at WB219 or AS104 or phone 921 9992 City Campus or 921 9998 North Shore campus to make an appointment. Appointments for South Campus can be made by calling 921 9992.
- let the receptionist know that you are a research participant and provide the title of our research and the name and contact details as given in this Information Sheet.

You can find out more information about AUT counsellors and counselling on http://www.aut.ac.nz/being-a-student/current-postgraduates/your-health-and-wellbeing/counselling.

VOLUNTARY PARTICIPATION AND WITHDRAWAL FROM THIS STUDY

Your participation is voluntary, and you can withdraw from the study at any time. You do not have to give us a reason why and it will not affect any care you receive in the future. If you choose to withdraw from the study, you will be offered the choice between having any data that is identifiable as belonging to you removed or allowing it to continue to be used. You may also request that any blood samples be removed and disposed of. You may also have any blood samples that have not yet been analysed returned to you by written arrangement with the study team.

HOW IS THE STUDY DESIGNED?

This is an observational study; no drugs or therapeutics are offered.

As a mTBI patient you may receive advice from healthcare professional during the course of the study. We want to analyse your data to help us determine which of these measures might be helping to take decision regarding the likelihood of secondary musculoskeletal injury or repeated concussion risk and whether they can be used to track your recovery over time.

WHO CAN TAKE PART IN THE STUDY?

You have been invited to take part in this study because you are aged 18_years and older and have recently suffered a mild traumatic brain injury (mTBI). You will be able to start the study if you meet the inclusion criteria outlined below.

To take part in the study you need to be aged 18 or older, reside in New Zealand, be able to provide informed consent, able to provide a blood sample and have sustained a recent mTBI/concussion.

You can NOT take part in the study if you have a suspected or confirmed neurodegenerative conditions or dementia, including, but not limited to, dementia (Alzheimer's disease, and vascular, Lewy body, or frontotemporal dementia), Parkinson's, Huntington's, motor neurone disease and mild cognitive impairment, and no history of acute neurological events or structural brain abnormalities including TBI, stroke, seizure, epilepsy, chronic headache, and brain tumour and no unstable severe medical conditions for example; cancer, severe coagulopathy, terminal illness, end-stage organ failure, acute kidney dysfunction, chronic kidney dysfunction or renal failure. If you are unable to provide informed consent due to cognitive impairment or language barriers, or unwilling to provide a blood sample or track the menstrual cycle or monitor gait parameters, or unable to walk independently for 2 minutes continuously also cannot take part in the study.

WHAT WILL HAPPEN TO MY DATA?

Your data will be de-identified in all analysis, reports and publications. Once the data have been analysed and findings have been produced, removal of your data may not be possible.

We would also like to use the data we collect here for future studies. At this stage we cannot tell you exactly what that research might be other than it would be related to mTBI. Your data will be de-identified before it is used. We will ask you to sign an additional consent form if this is the case.

WHAT WILL HAPPEN TO MY BLOOD?

When your blood is drawn, it will be tested for Follicle stimulating hormone, Estrogen, Luteinizing hormone, Progesterone Hormones. We will take no more than 40 mL of blood. For reference, a donation to NZ Blood is 457 mL, ten times as much as we will take. You must alert the study team before we take any blood if you wish your samples to be disposed of with Karakia or if you wish them returned to you. We cannot post these samples by mail so you will need to return to the site where you donated the blood to collect them or to AUTM if they have been sent for storage.

Here is a list of all the tests we will perform on your blood:

- Follicle stimulating hormone
- Estrogen
- Luteinizing hormone
- Progesterone

  If you consent to the use of your blood for future research related to mTBI and brain health, we will keep some of your blood secure in a freezer. We will ask you to sign an additional consent form if this is the case.

WHAT ARE THE POSSIBLE RISKS OF THIS STUDY?

Although routine blood collection is generally safe you may experience some pain or discomfort when having your blood taken. Your arm may be sore for a few hours afterwards. There is a minor risk of bruising or haematoma, which is when blood pools under the skin causing a dark bump to form. These are not likely to seriously affect you but they may be distressing. If you have concerns about anything involving the collection of blood you are welcome to contact the study team and have them respond to any concerns you might have.

Some of the walking tests may make participants unsteady when their balance is challenged. This will be addressed by ensuring that participants have close supervision and guarding by the assessor as required during all tests. If there are any tests that participants do not feel comfortable attempting, they do not need to perform these. The assessor will monitor how they are feeling throughout each procedure, and they will be able to stop the session at any stage.

What are the benefits?

A possible benefit to you is that you will receive the result from the Brain Injury Screening Tool (BIST) and walking test that are used by GPs and Physiotherapists in clinical practice and may help you to track your progress or any change in your movements. The main benefit of your participation is that it will increase our understanding of whether these gait parameters and hormonal levels can help to detect the changes in the balance and walking pattern after concussion and the risk of secondary musculoskeletal injuries or repeated concussion. If this method is useful and accurate, it could serve as a valuable tool for confirming the risks involved in the post-concussion return to play decisions and tracking recovery after concussion in the broad community.

How will my privacy be protected?

Study data will be recorded in an online database called REDCap. This database is securely hosted at AUT and is inaccessible to people other than the study team. Results will be provided as de-identified group data. All data collected during the study will be in storage at the AUT SPRINZ ethics storage room. All data will be stored under the AUTEC policy, which secures this data for ten years.

Individual information will not be shared or discussed. Although efforts will be made to protect your privacy, absolute confidentiality of your information cannot be guaranteed. Even with coded and anonymised information, there is no guarantee that you cannot be identified. The risk of people accessing and misusing your information (e.g. making it harder for you to get or keep a job or health insurance) is currently very small but may increase in the future as people find new ways of tracing information.

What if something goes wrong?

If you were injured in this study, you would be eligible to apply for compensation from ACC just as you would be if you were injured in an accident at work or at home. This does not mean that your claim will automatically be accepted. A claim would have to be lodged through an ACC registered health professional. If your claim is accepted, you will receive funding to assist in your recovery. If you have private health or life insurance, you may wish to check with your insurer that taking part in this study won't affect your cover.

What will happen to my information?

When you consent to take part in this study, you will be assigned a unique identifier number (e.g. CH001) so that your information can be de-identified for analysis. This number will be noted on your consent form. During this study the study team will record information about you and your study participation. This includes your consent form.

We may collect survey information either online through our REDCap database or hardcopy when you attend your visit. The choice is yours. That information, along with the clinical measurements and the results of our blood tests will be stored on REDCap.

All of the research data collected in this study will be stored on a secure and password protected cloud service (OneDrive). Only Professor Patria Hume, Dr Nusratnaaz Shaikh, Dr. Anja Zoellner and Sarniya Moganathas and will have access to your identifiable research data.

For research purposes, a master spreadsheet will be created with all information that could identify you removed, your unique identifier number will be used to record data in this spreadsheet. Only this second de-identified spreadsheet will be shared with the rest of the research team and used for analysis and in publications/presentations.

Identifiable Information

Identifiable information is any data that could identify you (e.g. your name, date of birth, or address). Only Professor Patria Hume and members of the study team will have access to your identifiable information.

De-identified (coded) information

To make sure your personal information is kept confidential, information that identifies you will not be included in any report generated by Professor Patria Hume. Instead, you will be identified by a code which keep your name secretly, so that you can be identified by your coded data if needed.

The following people may have access to your de-identified (coded) information:

- Biostatistician assisting in final data analysis.
- Other members of the Study Team
- Abbott Diagnostics (USA) if you consent to it

The results of the study may be published or presented, but not in a form that would reasonably be expected to identify you.

Security and storage of your information

All of your personal information will be stored on a secure password protected cloud service (OneDrive). Only named members of the research team will have access to this information.

Research data will be stored on REDCap, a cloud-based database hosted at AUT. This database shares all of AUT's cybersecurity protections. Only the study team will have access to this database. Digital data will be stored on the AUT R drive, the folder that has already been set up for the wider research project

For research purposes, you will be assigned a unique identifier code so that your data can be de-identified. A second master spreadsheet will be created with all information that could identify you removed. Only this second de-identified spreadsheet will be shared with the rest of the research team and used for analysis and used in publications/presentations. Analysis and reporting of data will be de-identified. All reporting of data in reports and publications will be de-identified.

Rights to access your information

You have the right to request access to your information held by the research team. You also have the right to request that any information you disagree with is corrected.

Please indicate on your consent form if you would like to receive a summary of the results of your tests. If you have any questions about the collection and use of information about you, you should ask Sarniya Moganathas.

Rights to withdraw your information

You may withdraw your consent for the collection and use of your information at any time, by informing Sarniya Moganathas, Dr Nusratnaaz Shaikh, Dr Anja Zoellner, or Professor Patria Hume. If you withdraw your consent, your study participation will end, and the study team will stop collecting information from you. If you agree, information collected up until your withdrawal from the study will continue to be used and included in the study. You may ask for it to be deleted when you withdraw, unless you withdraw after the study analyses have been undertaken.

Are there any cultural considerations?

You may hold beliefs about sacred and shared values about your tissue samples and/or data originating from this tissue. The cultural issues associated with sending your data overseas and/or storing your tissue and data should be discussed with your family/whānau as appropriate. If you wish to know more about the study, we can arrange for a member of the study team to come and talk to you and your whānau. Your samples can be disposed of with Karakia, you only need to ask the study team when you come to clinic.

Personal and health information is a tāonga and will be treated accordingly. The following data sovereignty principles are in place to ensure that the data generated from this research are protected and may benefit

Māori now and into the future. The principles included are whakapapa, whanaungatanga, kotahitanga, manaakitanga and kaitiakitanga. This research programme is also establishing a Māori consultation committee to assist with the design and presentation of this and future research.

Whakapapa: We understand that all data have genealogy and special relationships to the community. We collect demographic and ancestry data to ensure these data still hold relevance to the communities they come from.

Whanaugatanga: The data kept in Aotearoa will be stored securely and accessed only by research staff members who maintain confidentiality, it is collected only from those participants who have consented to the collection – and participants are encouraged to consult with whānau if that is culturally appropriate for them. The distribution of any results will be done in a way that recognises the reciprocity of the participant-researcher relationship.

Kotahitanga: This research, whilst it may not offer personal benefit, will hopefully contribute to collective benefit. You information will be used in a way that benefits more than just the researchers, but people affected by mTBI.

Manaakitanga: Free and informed consent and clear communication between yourself and study team is a hallmark of respect and Manaakitanga. We acknowledge the personal investment you make by participating and the importance of reciprocity of the research process.

Kaitakitanga: this research is designed to empower and discover. Māori data will be accessible where required and any analysis specific to gender or ethnicity will contribute to overcoming historic data inequality.

What happens after the study or if I change my mind?

If you wish to withdraw from the study at any point you can contact either Dr Nusratnaaz Shaikh, Dr. Anja Zoellner, Professor Patria Hume or another member of the Study Team if that is easier. If you do withdraw from the study, you can choose to either, allow the information collected up until your withdrawal to continue to be used or you may ask for it to be deleted when you withdraw. If you withdraw after the study analyses have been undertaken, it may not be possible to remove your data/information.

Will I receive feedback on the results of this research?

The overall study results will be published on the AUT SPRINZ SKIPP website Sports Kinesiology Injury Prevention and Performance - SPRINZ - AUT (https://sprinz.aut.ac.nz/areas-of-expertise/sports-kinesiology-injury-prevention-and-performance).

Within 120 days of the study finishing a 1–2-page summary of the research findings will be available. If you would like to receive a copy, please indicate on the consent form, and provide a contact email address.

Who is funding the study?

There are no direct financial costs to you as a participant in this study. This research is part funded by Abbott Laboratories (USA), a medical device and research company.

Each participant will receive $20 voucher for each of the clinic visits.

What do I do if I have concerns or further questions about this research?

If you have any concerns regarding this project, please contact the lead investigator – Professor Patria Hume, patria.hume@aut.ac.nz mobile 021 805 591.

For Māori cultural support contact please contact Dr Doug King, doug.king@aut.ac.nz.

Concerns regarding the conduct of this study should be notified to the Executive Secretary of AUTEC, ethics@aut.co.nz (+649)9219999 ext 6083.

If you want to talk to someone who isn't involved with the study, you can contact an independent health and disability advocate on:

Phone:	0800 555 050
Fax:	0800 2 SUPPORT (0800 2787 7678)
Email:	advocacy@advocacy.org.nz
Website:	https://www.advocacy.org.nz/

WHO HAS APPROVED THE STUDY?

This study has been approved by an independent group of people called the AUT Ethics Committee (AUTEC), who check that studies meet established ethical standards.

Approved by Health and Disability Ethics Committee on 14th March 2025

Reference number HDEC 2024 EXP 21888.

Note: The Participant should retain a copy of this form.

Appendix D – Consent form

Consent Form

Project title: Walking patterns, hormonal cycles, and injury risks in female athletes' post-concussion

This project is part of the Women's Health and Neuroscience Research Programme study "Determining optimal sampling times for concussion blood biomarkers for prediction of recovery in females" approved by HDEC 2024 EXP 21888, and registered with ANZCTR ACTRN12625000260426.

Research Team
Co-ordinating Investigator: Professor Patria Hume
Co-Principle Investigators: Dr Nusratnaaz Shaikh
Co-Associate Investigators: Dr. Anja Zoellner, Dr Christi Essex, Marguerite Sandleback
Primary Researcher: Sarniya Moganathas

- ○ I have read and understood the information provided about this research project in the Information Sheet dated 30th July 2025
- ○ I have had an opportunity to ask questions and to have them answered.
- ○ I have had the opportunity to use a legal representative, whānau/ family support or a friend to help me ask questions and understand the study.
- ○ I am satisfied with the answers I have been given regarding the study and I have a copy of this consent form and information sheet.
- ○ I understand that taking part in this study is voluntary (my choice) and that I may withdraw from the study at any time without being disadvantaged in any way.
- ○ I understand that if I withdraw from the study then I will be offered the choice between having any data that is identifiable as belonging to me removed or allowing it to continue to be used. However, once the findings have been produced, removal of my data may not be possible.
- ○ I consent to the research staff collecting and processing my information, including information about my health.
- ○ I agree to an approved auditor appointed by the New Zealand Health and Disability Ethics Committees, or any relevant regulatory authority or their approved representative reviewing my relevant medical records for the sole purpose of checking the accuracy of the information recorded for the study.
- ○ I understand that my participation in this study is confidential and that no material, which could identify me personally, will be used in any reports on this study.
- ○ I understand the compensation provisions in case of injury during the study.
- ○ I consent to my General Practitioner (GP) or current provider being notified of clinically significant abnormal results. This is a mandatory component of study participation. Therefore, GP contact details are requested here: _____

(please tick one): Yes ○ No ○

I agree to take part in this research.

(please tick one): Yes ◯ No ◯

I wish to receive a summary of the results from the study.

(please tick one): Yes ◯ No ◯

I consent to be contacted by study team about future opportunities to participate in future, related research.

(please tick one): Yes ◯ No ◯

I consent to Abbott Diagnostics (USA) having access to my deidentified data.

(please tick one): Yes ◯ No ◯

I give permission to the study team for the future unspecified use of my deidentified research data.

(please tick one): Yes ◯ No ◯

I wish to have a karakia for my sample when this is disposed.

(please tick one): Yes ◯ No ◯

Participant's signature:.....

Participant's name:.....

Participant's Contact Details (if you wish to have your individual results):

Email: .....

Date: .....

Declaration by member of research team:

I have given a verbal explanation of the research project to the participant and have answered the participant's questions about it. I believe that the participant understands the study and has given informed consent to participate.

Researcher's name: _____

Signature: _____ Date: _____

Approved by Health and Disability Ethics Committee on 14th March 2025

Reference number HDEC 2024 EXP 21888.

Note: The Participant should retain a copy of this form.

Appendix E - An example of a search string: EBSCO

1) How do menstrual cycle changes influence in secondary injury risk (lower limb injuries and concussion) in female athletes after a concussion injury?
   i) (“menstrual cycle” OR “menstrual phase*” OR “hormonal fluctuation*” OR hormone* OR estradiol OR estrogen OR progesterone OR follicular OR luteal OR ovulatory OR premenstrual OR amenorrhea OR oligomenorrhea) AND (“secondary injury” OR reinjur* OR “subsequent injury” OR recurrence OR “recurrent injury” OR “repeat injury” OR “injury recurrence” OR “contralateral injury”) AND (“lower limb” OR “lower extremity” OR knee OR ankle OR hip OR thigh OR hamstring OR quadriceps OR groin OR calf OR “anterior cruciate ligament” OR ACL OR MCL OR meniscus) AND (female* OR women OR girl* OR “female athlete*” OR “physically active female*” OR “non-athlete females” OR “non-athlete women”) AND ("post-concussion" OR "following concussion" OR "after concussion")
   ii) (“menstrual cycle” OR “menstrual phase*” OR “hormonal fluctuation*” OR hormone* OR estradiol OR estrogen OR progesterone OR follicular OR luteal OR ovulatory OR premenstrual OR amenorrhea OR oligomenorrhea) AND (“secondary injury” OR reinjur* OR “subsequent injury” OR recurrence OR “recurrent injury” OR “repeat injury” OR “injury recurrence” OR “contralateral injury”) AND (concussion OR “mild traumatic brain injury” OR mTBI OR “concussion symptom*” OR “concussion recovery” OR “symptom resolution” OR “return to play” OR “return to sport” OR RTP OR “time to recovery”) AND (female* OR women OR girl* OR “female athlete*” OR “physically active female*” OR “non-athlete females” OR “non-athlete women”) AND ("post-concussion" OR "following concussion" OR "after concussion")

2) Which gait pattern alterations, influence risk of secondary injuries in females after a concussion injury?
(gait OR walking OR locomotion OR "spatiotemporal parameters" OR "stride length" OR "step length" OR "step time" OR "stride time" OR "cadence" OR "gait velocity" OR "gait speed" OR balance OR stability OR "postural control" OR kinematic* OR kinetic* OR "joint angles" OR "joint moments" OR "foot progression" OR "gait symmetry" OR "stride variability" OR "stance time" OR "swing time" OR "double support time") AND (“secondary injury” OR reinjur* OR “subsequent injury” OR recurrence OR “recurrent injury” OR “repeat injury” OR “injury recurrence” OR “contralateral injury”) AND (female* OR women OR girl* OR “female athlete*” OR “physically active female*” OR “non-athlete females” OR “non-athlete women”) AND ("post-concussion" OR "following concussion" OR "after concussion")

3) How do menstrual cycle related hormonal changes, and gait pattern alterations influence the risk of secondary injury risk (lower limb injuries and concussion) for females after mTBI?
(“menstrual cycle” OR “menstrual phase*” OR “hormonal fluctuation*” OR hormone* OR estradiol OR estrogen OR progesterone OR follicular OR luteal OR ovulatory OR premenstrual OR amenorrhea OR oligomenorrhea) AND (gait OR walking OR locomotion OR "spatiotemporal parameters" OR "stride length" OR "step length" OR "step time" OR "stride time" OR "cadence" OR "gait velocity" OR "gait speed" OR balance OR stability OR "postural control" OR kinematic* OR kinetic* OR "joint angles" OR "joint moments" OR "foot progression" OR "gait symmetry" OR "stride variability" OR "stance time" OR "swing time" OR "double support time") AND (“secondary injury” OR reinjur* OR “subsequent injury” OR recurrence OR “recurrent injury” OR “repeat injury” OR “injury recurrence” OR “contralateral injury”) AND (female* OR women OR girl* OR “female athlete*” OR “physically active female*” OR “non-athlete females” OR “non-athlete women”) AND ("post-concussion" OR "following concussion" OR "after concussion")

Appendix F - Blood sample report

WOMEN'S HEALTH AND NEUROSCIENCE RESEARCH PROGRAMME

Scan the QR code or email brain@aut.ac.nz to refer a friend

WHIP Concussion Study: Your Individual Biomarker Panel Results

We are pleased to provide you with your complete results, including the blood biomarker analyses and gait analyses. Please note that these results are for research purposes only. At the completion of the study a fact sheet will be available on the AUT WHN website.

Participant details

Sex:	Female
Age at blood draw:	29
Reproductive status (self-reported):	Naturally menstruating (regular period, no hormonal contraceptives)
Start Date	21/10/2025

Interpreting out-of-range results

If results fall outside the usual laboratory reference range, they will be marked with an asterisk (*) in this summary. If there is no asterisk, the results do not match the specific patterns or markers that our clinical team has pre-specified for proactive general practitioner notification (including the combined sex hormone patterns described below, or abnormal ferritin, vitamin B12, vitamin D, TSH, or free T4 levels). We will not contact your general practitioner about these findings; however, you are welcome to discuss them with your usual healthcare provider if you have any questions or concerns.
Unless otherwise stated, the reference ranges shown below are those typically used for a naturally menstruating adult female of this age.

The average of your three menstrual cycles are reported.

Inflammation panel

Analyte	Luteal Phase	Menses	Follicular Phase	Ovulation	Units	Reference Range
Ferritin	25.65	22.48	25.75	18.85*	ng/mL	20–170 ng/mL.
Cortisol	232.3	199.6	127.3*	260.3	nmol/L	Morning: 170–500 nmol/L
CRP	<0.10	0.16*	<0.10	<0.10	mg/L	<5 mg/L.

For more information on the WHN studies see https://sprinz.aut.ac.nz/brain-testing. For further information contact brain@aut.ac.nz.

Vitamin panel

Analyte	Luteal Phase	Menses	Follicular Phase	Ovulation	Units	Reference Range
Vitamin D	36.6*	39.7*	43*	42.6*	nmol/L	50–150 nmol/L (optimal).
Active B12	171.1	160.2	167.5	131.8	pmol/L	≥58 pmol/L (deficiency unlikely).
Magnesium	0.74	0.72	0.77	0.79	mmol/L	0.7–1.0 mmol/L.

Thyroid function panel

Analyte	Luteal Phase	Menses	Follicular Phase	Ovulation	Units	Reference Range
TSH	1.56	0.79	0.55	1.42	mIU/L	0.27–4.20 mIU/L.
Free T4	11.93*	12.01	12.13	12.64	pmol/L	12–22 pmol/L.
Total T4	6.43	6.68	6.54	6.65	µg/dL	4.87–11.72 µg/dL.

Sex hormone panel[¥]

Analyte	Luteal Phase	Menses	Follicular Phase	Ovulation	Units	Reference Range
Estradiol	800	72	191	369	pmol/L	5–850 pmol/L (follicular), 150–1450 (mid-cycle), 85–1250 (luteal), 70-180 (menses)
FSH	6.76	4.32	4.65	3.23	IU/L	3–10 IU/L (early follicular & follicular), 4–25 (mid-cycle), 2–8 (luteal)
LH	9.48	3.14	4.11	20.43	IU/L	2–8 IU/L (early follicular & follicular), 10–75 (mid-cycle), 2–8 (luteal)
Prolactin	371.56	299.05	144.69	348.66	mIU/L	<630 mIU/L.
Progesterone	6.6	0.8	0.5	1.1	nmol/L	<1-2 nmol/L (early follicular), <1-3 nmol/L (follicular phase), ~ 1-5 nmol/L (pre-ovulatory), ~10-50+ nmol/L (luteal phase)
Testosterone	1.39	0.99	1.32	1.78	nmol/L	<3 nmol/L.
DHEA-S	6.73	5.29	6.49	7.63	µmol/L	3.9–11.5 µmol/L.

[¥]A low result for estradiol, testosterone, FSH, and LH is only considered abnormal if all are low. A high result for FSH, LH, and prolactin is only considered abnormal if all are high. Abnormal results will be flagged. If you have concerns, please consult with an appropriate healthcare professional. Interpretation of some markers uses age, sex, and self-reported reproductive status. Results do not constitute a clinical diagnosis.

Key for biomarker acronyms

CRP: C-reactive protein; **TSH:** Thyroid-stimulating hormone; **Free T4:** Free thyroxine; **Total T4:** Total thyroxine; **FSH:** Follicle-stimulating hormone; **LH:** Luteinising hormone; **DHEA-S:** Dehydroepiandrosterone sulphate

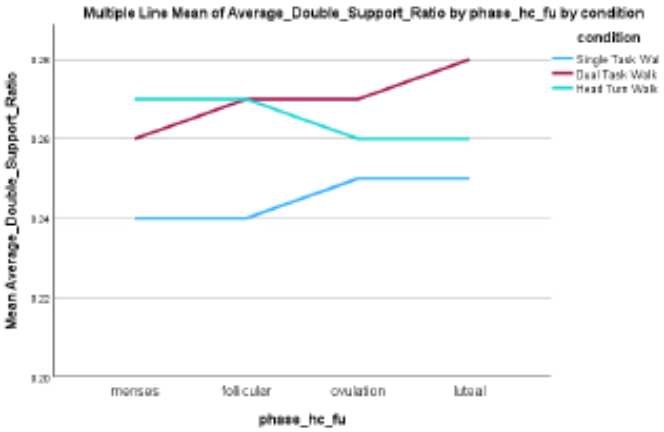
For more information on the WHN studies see https://sprinz.aut.ac.nz/brain-testing. For further information contact brain@aut.ac.nz.

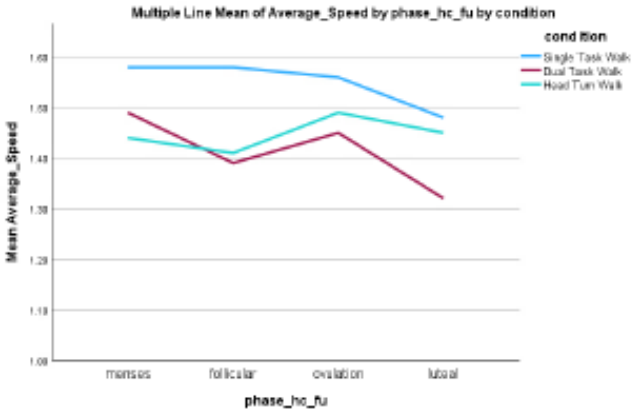
Walking Task Performance Interpretation

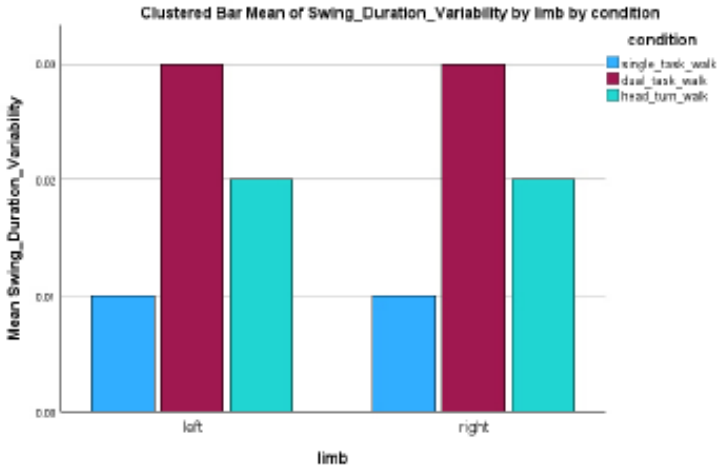
This section explains how your walking changes for three types of walking tasks:

- **Normal walking (single task)** – walking at your usual pace
- **Dual-task walking** – walking while counting backwards
- **Head-turn walking** – walking while turning your head and counting backwards

These tasks help us understand how your brain and body work together when walking becomes more challenging. Your results are shown in graphs comparing these tasks to your normal walking. The results are also shown across four menstrual cycle phases: menses, follicular, ovulation, and luteal.







For more information on the WHN studies see https://sprinz.aut.ac.nz/brain-testing. For further information contact brain@aut.ac.nz.

Interpretation

- Compared to normal walking, small but **clearer differences** were observed **during dual-task** and head-turn walking.
- Dual-task walking showed slight changes, indicating the effect of increased attention demands on walking performance.
- **Head-turn** walking showed **small variations**, likely due to added coordination and balance challenges.
- Across menstrual phases, minor fluctuations were visible, but no consistent pattern was observed.
- No phase showed a clear or meaningful change in walking performance.
- **Left & right limb:** Small variations were observed across tasks, with slightly more change during dual-task and head-turn walking, but overall performance remained stable.

👉 Overall, walking remains stable, with small variations during more challenging tasks, which are expected.

Summary

- Walking is generally consistent across tasks.
- **Slight differences** are more noticeable during complex tasks.
- Menstrual cycle shows minimal influence on walking.
- Overall walking performance remains **stable and normal**.

For more information on the WHN studies see https://sprinz.aut.ac.nz/brain-testing. For further information contact brain@aut.ac.nz.

Appendix G - General Health Questionnaire (GHQ)

mTBI Screening, Demographics, and GHQ

Study Registration Number

_____

Data collection method:

- ◯ Self report (participant)
- ◯ Interview (researcher)

Demographic Information

What ethnic group or groups do you identify with (select all that apply)

- ☐ NZ European
- ☐ Māori
- ☐ Pacific Peoples
- ☐ Asian
- ☐ Middle Eastern/ Latin American/African
- ☐ Other Ethnicity

Please specify

_____

What is your highest level of education?

- ◯ Primary School
- ◯ High School/ Secondary
- ◯ College, Professional, or Training Qualification
- ◯ University
- ◯ Other

Please specify

_____

Inclusion / Exclusion Criteria Screening

Have you ever experienced a severe Traumatic Brain Injury?

- ◯ Yes
- ◯ No

(A severe traumatic brain injury is a previous brain injury e.g. skull fracture or brain bleed that ended up with a hospital admission (e.g. admitted to ward))

Previous Traumatic Brain Injury: INELIGIBLE

Have you ever been diagnosed with, or do you have any history of, neurological or neurodegenerative conditions including dementia, Parkinson's, Huntington's, motor neurone disease, mild cognitive impairment, stroke, seizure, epilepsy, chronic headache, or brain tumour?

- ◯ Yes
- ◯ No

History of neurological or neurodegenerative conditions: INELIGIBLE

Do you have any unstable, severe medical conditions, including, but not limited to, cancer, severe coagulopathy, terminal illness, end-stage organ failure, acute kidney dyfunction, chronic kidney dysfunction or renal failure?

- ◯ Yes
- ◯ No

History of unstable, severe medical conditions: INELIGIBLE

General Health Information

Height (in centimetres; researcher to complete) Optional for < 24 hours post-concussion at Hutt Valley ED only Remove shoes and socks Stand straight: Ensure the participant is standing with their back against the stadiometer, feet together, and heels touching the base. Head position: Ensure the participant is looking straight ahead, ensuring the head is in a neutral position (not tilted up or down). Take a deep breath: Ensure the participant stands tall and takes a deep breath in to ensure their posture is fully upright. Measurement: lower the stadiometer's headpiece until it gently touches the top of the participant's head and record the height.	_____ (cm)
Weight (in kilograms; researcher to complete) Optional for < 24 hours post-concussion at Hutt Valley ED only Remove heavy clothing. Ask the participant to step onto the scale and stand still with their weight evenly distributed on both feet. Measurement: record the weight once the scale reading stabilises.	_____ (kg)
BMI Optional for < 24 hours post-concussion at Hutt Valley ED only	_____
Do you have any history of mental health conditions?	◯ Yes ◯ No
Select all mental health issues that apply	☐ Depression ☐ Anxiety ☐ OCD ☐ PTSD ☐ Eating Disorders ☐ Alcohol/other drug addiction ☐ Other
Other mental health condition	_____
Do you have an autoimmune condition?	◯ Yes ◯ No

What is your autoimmune condition?	_____
Do you have diabetes?	◯ Yes ◯ No
What type of diabetes do you have?	◯ Type 1 ◯ Type 2
Have you experienced a physical injury other than your concussion the last 3 months?	◯ Yes ◯ No
Please describe your injury	_____
Have you had surgery in the last 3 months?	◯ Yes ◯ No
What was your surgery for?	_____
Do you currently experience insomnia or significant sleep disturbances?	◯ Yes ◯ No
On average, how many hours have you slept per night over the last month?	_____ (hours)
Approximately how many hours did you sleep last night?	_____ (hours)
Approximately how many hours ago did you consume your last meal	_____ (hours)
Which of the following best describes your smoking status?	◯ Never smoked ◯ Ex smoker ◯ Occasional smoker ◯ Regular smoker
How many cigarettes do you typically smoke per day?	_____ (cigarettes per day)
Which of the following best describes your vaping habits?	◯ Never vaped ◯ Ex vaper ◯ Occasional vaper ◯ Regular vaper
On average, how often do you consume a drink containing alcohol?	◯ Never ◯ Monthly or less ◯ Two to four times a month ◯ Two to three times a week ◯ Four or more times a week

Please indicate how many standard drinks you consume on a typical day when drinking	◯ 1 or 2 ◯ 3 or 4 ◯ 5 or 6 ◯ 7 to 9 ◯ 10 or more (standard drinks)
Have you consumed alcohol within the last 5 hours?	◯ Yes ◯ No
How many hours ago did you stop consuming alcohol?	_____ (hours)
How many standard drinks did you have?	_____ (standard drinks)
Have you experienced an infection, respiratory illness, or allergic reaction in the last 2 weeks?	◯ Yes ◯ No
Please specify	◯ Infection ◯ Respiratory illness ◯ Allergic reaction
Are you currently taking any medication other than contraceptives?	◯ Yes ◯ No
Please list the medications you are taking	_____

Sports Participation Information

Are you currently involved in organised sports?	◯ Yes ◯ No
Please indicate your level of competition	◯ Recreational (train but do not participate in competition) ◯ Amateur (compete but do not expect to win) ◯ High-level athlete (qualify and compete at National Championship level) ◯ Elite non-professional (qualify and compete at the international level) ◯ Professional (full-time paid)
What sport or sports do you play?	_____
Is this a team sport?	◯ Yes ◯ No
What position do you typically play?	_____

Physical Activity Information

Please indicate your level of physical activity	◯ Sedentary (minimal to no regular exercise) ◯ Lightly active (occasional exercise, e.g., 1-2 times per week) ◯ Moderately active (regular exercise, e.g., 3-4 times per week) ◯ Highly active (consistent exercise regimen, e.g., 5 or more times per week)
On average, how many hours per week do you train, compete, or exercise?	_____ (hours)
How many hours ago did you last engage in physical activity or exercise?	_____ (hours ago)
How would you rate the intensity of your last exercise session?	◯ Light ◯ Moderate ◯ Vigorous
Please provide a short description (e.g., what physical exercise did you engage in, why have you rated it the intensity you have?)	_____

Menstrual and Reproductive Information

Which of the following best describes your current menstrual and reproductive status? Please select all that apply.	☐ Naturally menstruating ☐ Utilising hormonal contraceptives, e.g., combined pill ☐ Currently pregnant ☐ Currently or recently postpartum (within one year of having given birth) ☐ Amenorrhea (absent menstrual cycle at a time that is not related to menopause or pregnancy) ☐ Perimenopause ☐ Menopause ☐ Post-menopause ☐ I don't know ☐ Other (if more than two) ☐ Not applicable (for example, gender does not reflect assigned sex at birth)
What was the start date of your most recent period? (if applicable)	_____
Number of bleeding days	_____
Cycle length (days)	_____
days_since_bleed	_____
cycle_day	_____

Menstrual Phase	_____
Are you currently using a contraceptive?	◯ Yes ◯ No
Which form of contraceptive are you currently using?	◯ Combined Oral Contraceptive Pill ◯ Progesterone only pill ◯ Rod Implant ◯ IUD - Hormonal (combined) ◯ IUD - Progesterone Only ◯ Depo-Provera ◯ Other
Please specify	_____